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## **The Allan Walker Plenary Lecture: Pesticides in Soil & Water**

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In 2003 Professor Allan Walker BSc. Ph.D. DSc. was awarded an honorary membership of the European Weed Research Society (EWRS) in “*recognition of a long established reputation for excellence in the field of weed science and in appreciation of a long, active and outstanding contribution to the European Weed Research Society*”. Allan was a world-renowned scientist who spent his 35-year career specialising in pesticide and environmental interactions. His expertise was also recognised in 1993 by the British Crop Protection Council who awarded him their Medal for “*Outstanding Services to Crop Protection Research in the United Kingdom*” and in 2003 when he was awarded the DowAgroSciences, Career Achievement Award “*In recognition of contribution to the advancement of science throughout a distinguished career*” presented at the 12<sup>th</sup> International Congress of Pesticide Chemistry, Piacenza, Italy. Allan applied his expert scientific knowledge to support the regulatory and policy roles of UK Government and the European Union and served on several pesticide advisory committees. Academia also acknowledged his scientific excellence when he was awarded a DSc (Soil Science), by the University of Nottingham in 1986 when he reviewed his work to identify ‘*Factors influencing the activity and persistence of herbicides in soil*’. In 2000 he was appointed Visiting Professor, at Cranfield University at Silsoe. Allan published extensively throughout his career and was author or co-author of at least 219 publications comprising: 4 books (editor or joint editor), 110 refereed papers in scientific journals, 8 chapters in books, 74 papers in conference proceedings, 23 popular articles in farming magazines and other reports.

The overall theme of Allan’s research throughout was to gain a quantitative understanding of the interactions between the various physical, chemical and microbial processes that control the biological activity and environmental dynamics of pesticide residues. He introduced many new concepts to the study of pesticide behaviour in soils and thus to the protection of the wider environment. In his early work, he was one of the first scientists to apply the established theories of nutrient availability in soils to the behaviour of soil-applied organic chemicals (1, 2, 3). This was driven by a need to understand the reasons for the variability in pesticide efficacy at the field scale and the need to improve commercial practices. He discovered that uptake from soil approximated to a simple flow process, and that any factor that controls the concentration of pesticide in the soil solution (adsorption/desorption relationships; degradation rates) or the availability of this solution to the plant (soil moisture content; relative distribution of chemical and roots in the soil) will influence biological activity. The research demonstrated how pesticide activity in the field is controlled by shoot-zone uptake of the chemical (2), by absorption of chemical by a small proportion of the total root system, and by the soil moisture relationships of the surface soil layers during early periods of seedling growth (3). The research as a whole contributed to a much wider appreciation and understanding of the factors controlling the variation in pesticide efficacy under practical use conditions.

Allan’s innovative contribution was to develop a detailed mechanistic simulation model – PERSIST (4) to forecast the dynamics of pesticide residues in the variable field environment from readily available data (4, 5, 6, 7, 8). The algorithms are now used in many more complex models of pesticide fate and behaviour, particularly those used in the regulatory process for

the calculation of soil and water PECs. PERSIST is now being used to assess the potential influence of global warming on pesticide degradation for over 200 UK sites.

Allan identified that there can be considerable spatial variation in pesticide sorption and degradation within apparently uniform fields (9) and he was again one of the first to recognise the potential use of advanced geostatistical techniques to evaluate the scale and structure of this variation (10). These techniques, in combination with appropriate models, allow “hot spots” prone to pesticide leaching and thus potential surface or groundwater contamination to be identified, and can be used to map these vulnerable areas at the farm or small catchment scale. This information can then be used to modify pesticide application regimes within precision agriculture management systems. His most recent work investigated microbial aspects of pesticide metabolism in soils (11, 12, 13). The rates of metabolism of readily degradable organic compounds have long been known to be enhanced by repeated application (14, 15), but Allan broke new ground by demonstrating that this can also occur with recalcitrant pesticides if sufficient selection pressure is applied (13, 16,). His team also demonstrated a link between species diversity in the soil with the ability to degrade a particular compound and the ease of induction and stability of enhanced biodegradation phenomena in soils. Strains of bacteria were isolated and identified that metabolise four of the eight pesticides found as significant water contaminants in Northern Europe (13, 17) and the genetic mechanisms identified (19). Bacteria with the ability to degrade several other persistent pesticides from different chemical classes have also been identified and characterised (16, 17, 19). They provide a means of bioremediation of contaminated environments, as he demonstrated for the persistent herbicide napropamide (11). Allan was key in the work to develop an understanding of pesticide fate in on-farm biobeds.

Allan enjoyed interacting with young and emerging scientists and enjoyed his role as supervisor of a number of highly successful PhD students. His reputation led to invitations to speak or provide consultancy in Europe and other countries including Malaysia, Brazil and South Africa. He leaves us with a rich legacy of science, friendship and humour.

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## **Pattern and potentiality of fungal isolates from soil-rhizosphere and from young *Orobanche crenata* infesting *Vicia faba* fields in South of Egypt**

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The Mediterranean basin countries account for nearly 25% of both the total global area planted to faba bean (*Vicia faba* L.) and its production. The average yield from 1991 to 2001 in Egypt was remarkably high (9423-9565kg/ha) and was approximately double the average for developing countries (4610-4659 kg/ha). Abiotic factors such as drought, high temperature, inadequate supply of nutrients and biotic factors such as microorganisms, parasitic weeds and nematodes, play important roles in reducing *Vicia faba* yields. Infestation of the parasitic weed *O. crenata* Forsk. (broom rape) in food legumes especially in faba bean is of high significance. While not the major agricultural problem in this area, it nevertheless deserves attention. The parasite was present throughout the whole of the Beni-Suef governorate (South of Cairo) infesting 53 % of the faba bean fields surveyed and resulting in 23% reduction in yield. Fields of faba bean highly infested with *O. crenata* were investigated in the governorate of Beni-Suef. 188 fungal isolates, obtained from the rhizosphere area of *Vicia faba* parasitised by *O. crenata* were compared to 26 isolates obtained from the youngest possible infected shoots of the parasite for a preliminary evaluation of the isolated fungi as biocontrol agents. This article records their identification and their *in vitro* bioassay variation against germination of *O. crenata* seeds.



## **Toxins produced by pathogenic fungi of grass weeds as potential natural herbicides**

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The ban of some dangerous herbicides, and the loss of efficacy of others due to the appearance of resistant weeds render the need for new active compounds particularly urgent, especially for the control of grass weeds which are among the worst weeds in many crops in the world. Among the possible sources of natural compounds, fungal pathogens of weeds appear to be particularly interesting. Toxins of plant pathogens could be used as new natural herbicides, both in their native forms, or as derivatives and analogues. For this purpose, many pathogens of grass weeds were collected, and their ability to produce toxic metabolites was ascertained. A strain of *Drechslera siccans* proved to be particularly interesting. This communication will describe the optimisation of the production *in vitro* of the phytotoxins produced by this fungus, its isolation from the fungal culture filtrates and its chemical and biological characterisation. The possible use of this toxin in integrated strategies for grass weed control will also be discussed.

## Fungal metabolites for management of *Orobanche ramosa*

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Extensive surveys carried out in Southern Italy allowed us to isolate many pathogenic fungi from diseased plants of *Orobanche ramosa*, a parasitic weed heavily infesting several important crops in the Mediterranean area. These fungi were evaluated both as biocontrol agents of this weed and as producers of natural herbicides. The ability of fifty-three strains to produce bioactive metabolites in solid and liquid cultures was ascertained, in order to identify metabolites inhibiting the germination of *O. ramosa* seeds. Among them, the organic extracts from liquid cultures of one strain of *Fusarium compactum* and one of *Myrothecium verrucaria* caused total inhibition of germination. In this communication, the isolation, identification and biological activity both of verrucarins, roridin A, isotrichoverrin B produced from *M. verrucaria*, and of neosolaniol monoacetate from *F. compactum*, will be described. Their potential use in biological and integrated control of *O. ramosa* will also be discussed.

## Phylogeny, pathogenicity and diversity of biocontrol agents for Alismataceae weeds in Australia and Korea.

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Plants species in the Family Alismataceae are significant weeds of rice in Australia and south East Asia. In Korea and Japan, *Sagittaria trifolia* is considered the most important weed of rice while other members of the family including *Damasonium minus* (starfruit), *Alisma lanceolatum*, *S. montivedensis* and *A. plantago-aquatica* are weeds of concern in Australia. These weeds are difficult to control due to their herbicide tolerance/resistance and their season-long emergence. A related weed species, *S. gramineae* is an important weed in irrigation supply channels in southern Australia and because of water quality issues, it has very limited chemical control options. In Australia and Korea, parallel research programs are investigating the use of inundative biological control of these weeds using the plant pathogenic fungi, *Rhynchosporium alismatis* and *Plectosporium tabacinum*. Recent studies in Australia involving the sequencing of the internal transcribed spacer from *R. alismatis* revealed a close relationship with the *Plectospheralla* genus and so the fungus has been transferred to the species *Plectosporium alismatis*. This finding demonstrated the close phylogenetic relationship between the organisms under investigation in the two countries.

During 2004, a survey of Alismataceae weeds in the southern regions of South Korea was undertaken to obtain isolates of fungi belonging to *Plectosporium* sp., which have potential as biocontrol agents for several weeds in NSW rice crops. From a total of 158 leaf samples of rice weeds collected over 10 days, forty five pure cultures were returned to Australia (under AQIS quarantine permits) and were cultured from single spores. Isolates that were morphologically similar to *Plectosporium* were tested for pathogenicity to *Alisma lanceolatum*, *Damasonium minus*, *Alisma lanceolatum* and *Sagittaria gramineae* in glasshouse experiments. When compared to the Australian isolates, those from Korea were less virulent, only causing small lesions on emergent leaves. Preliminary data from the sequencing of the internal transcribed spacer of a number of the isolates confirm their identity as species of *Plectosporium*, with at least one isolate showing 100 % homology with the sequence of *Plectosporium alismatis*.

## **Environmental fate of *Phoma macrostoma*, a fungus for broadleaf weed control in turfgrass**

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Several isolates of *Phoma macrostoma* have demonstrated bioherbicidal activity against dandelions and other broadleaf weeds in turfgrass and this technology is being developed into a microbial weed control product by Agriculture & Agri-Food Canada and The Scotts Company. Because the fungus is applied at higher levels than those occurring in nature, we investigated aspects of environmental fate in order to assess the potential risk. A DNA probe highly specific to isolates of *P. macrostoma* with bioherbicidal activity was developed to detect the fungal DNA in plant and soil samples. Field experiments were conducted to monitor the presence or absence of *P. macrostoma* in space and time. The fungus had limited mobility in both the horizontal and vertical soil profiles and did not persist in plants or soil after 4 months. Studies under greenhouse conditions were conducted to determine the fate in susceptible and resistant plant species. The results showed that both resistant and susceptible plant species were colonized after application of the fungus to soil, but that the frequency of isolation from the rhizosphere declined after 10 days. These results suggest that *P. macrostoma* would have minimal environmental impact.



## **Can mycelial inoculum be an alternative to conidia in the case of *Stagonospora cirsii* J.J. Davis, a potential biocontrol agent of *Cirsium arvense*?**

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The pycnidial fungus, *Stagonospora cirsii* is being evaluated as a potential bioherbicide for control of *Cirsium arvense*. One of the main features of this microorganism is dependence of pycnidial formation on near ultraviolet light. It is a constrain for large scale conidia production. In this study we compared temperature and pH requirements for germination of conidia and mycelium of *S. cirsii* C-163/6, and biocontrol efficacy of conidial and mycelial inoculum of the fungus. The mycelium was produced on autoclaved millet in the darkness, conidia were obtained on pearl barley under NUV. Optimum temperature for conidia germination was about 27°C, for mycelium the optimum was lower (24°C). Optimal pH for germination of conidia lied between 6 and 7. The mycelial inoculum propagated better at pH 5-6. It seems that mycelial type of inoculum will fit better to the environment than conidial inoculum of *S. cirsii*. In addition we studied efficacy of mycelial and conidial inoculum of the fungus applied on leaves of the weed or on soil surface. Millet colonized with the mycelium was dried, ground and applied at the rate 1 g/dm<sup>2</sup>. For soil application, the concentration of conidial inoculum was 1·10<sup>6</sup> conidia per g of soil, and for foliar application the concentration was 5·10<sup>6</sup> conidia/ml (1 ml/plant). Plants of *C. arvense* at the rosette stage were inoculated and subjected to 24-h period of 100% relative humidity. By the 14-th day post inoculation the best results (more than 50% reduction of fresh biomass and dry weight of roots) were observed when mycelial inoculum of *S. cirsii* was deposited either on leaves of the weed or on the soil surface. Formulation of the mycelium in oil-based emulsion improved efficacy of *S. cirsii*.

## **Towards commercialisation of the fungus *Sclerotinia sclerotiorum* as a mycoherbicide for *Ranunculus acris* in dairy pasture**

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*Ranunculus acris* (giant buttercup), a perennial species of European origin, has spread widely in dairy pastures in New Zealand. It caused a loss in milk solids revenue of \$156 million in 2001-02 despite the use of the herbicides MCPA, MCPB, thifensulfuron-methyl and flumetsulam. MCPA and MCPB, once useful herbicides, now fail to control giant buttercup due to herbicide resistance. An additional problem with both MCPA and thifensulfuron-methyl is the damage they cause to clovers in treated pastures. These problems along with the high rainfall common in dairying regions (2 to 3 m/y), and the high-value of dairy production, have created an ideal environment for the development of *Sclerotinia sclerotiorum* as a mycoherbicide for use against *R. acris*. This plant pathogenic pathogen does not attack clovers and there is no increase in disease risk in susceptible crops downwind of a treated pasture because the spore-trapping ability of dairy swards greatly limits the escape and aerial dispersal of the ascospores of the fungus that are formed in the treated pasture. Funded by the New Zealand government, the New Zealand dairy industry, and a joint-venture company, we have shown that control of *R. acris* to a level acceptable to dairy farmers (60% reduction) is attainable at an application rate of 50 kg/ha of a novel mycelium-based formulation applied with conventional farm spreader machinery. The effects remain evident for at least one year giving potential production benefits in two milking seasons. Registration and commercialisation of a mycoherbicide product are currently under consideration by a leading New Zealand fertilizer company.

## SolviNix: R&D of a Bioherbicide for Tropical Soda Apple

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Tropical soda apple (TSA; *Solanum viarum*) is a highly invasive noxious weed in Florida and the southeastern USA. We have shown that *Tobacco mild green mosaic tobamovirus* (common name: tobacco mild green mosaic virus; TMGMV), a plant virus that occurs worldwide on some *Nicotiana* spp., kills TSA upon infection by eliciting a lethal hypersensitive response. Typically, two to three weeks after virus inoculation, TSA plants of all ages wilt suddenly and die quickly and completely without regrowth. The TSA-killing ability of TMGMV is rather specific to this plant-virus interaction. We have successfully field tested TMGMV in several locations in Florida with repeatable, nearly 100% weed-kill. The virus could be applied over large areas, such as open ranch lands, with a tractor-mounted spray boom or a wiper applicator. TSA can be spot-treated with a backpack-sprayer or a pressure-jet spray. In wooded areas and over uneven terrain, an ATV-mounted spot-sprayer or a wiper could be used. Based on an extensive host-range study we have conducted, the virus is clearly adapted to plants in the Solanaceae. Of nearly 420 plants in 57 families tested, including 175 solanaceous plants, 68% were immune or resistant to TMGMV. Among plants outside the Solanaceae, 98% were immune or resistant. Among the susceptible plants (including asymptomatic and symptomatic plants), only peppers (*Capsicum* spp.) and tobaccos (*Nicotiana* spp.), are of any real concern. However, the risk to peppers and tobaccos and the other susceptible plants is negligible and manageable since the virus has no natural vectors and does not spread except through physical contact. TMGMV does not infect humans, animals, birds, fish, or insects and hence poses no risks to these organisms. We are currently following a small-business technology-transfer model to develop and register this bioherbicide. This R&D model, which may have broad applicability to other bioherbicide projects, will be discussed in detail. We intend to register TMGMV as the world's first commercial viral bioherbicide.

## Survival of *Stagonospora cirsii* J.J. Davis, a leaf pathogen of *Cirsium arvense*, in the soil

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The fungus, *Stagonospora cirsii* is a potential biocontrol agent of *Cirsium arvense*. In preliminary experiments, the pathogen infected the weed plants at soil surface application. The objective of this study was to evaluate survival and population dynamics of *S. cirsii* in the sterile soil. An artificial soil mixture (peat: sand 3: 1) and a field sod-podzol soil were used in the experiments. Survival of *S. cirsii* in the soils was studied with an adapted technique using membrane filters. In both soils the fungus survived on the filters for 4 weeks and more, but the survival rate of soil surface inoculum of *S. cirsii* was higher than survival rate of the inoculum incorporated into the soil at the depth of 3 cm. In the soil or on its surface conidia of *S. cirsii* germinated and formed mycelium. In 3-4 weeks the contents of the mycelial cells became granular, the most of the cells looked collapsed. The population dynamics of the fungus in the sterile soil was studied by a dilution technique. For two initial concentrations of *S. cirsii* ( $10^3$  and  $10^6$  conidia per g of the soil), the CFU “decay” curves were similar: from decreasing CFUs (first 15 days) to a low constant level (15-90 days), and to the period of population growth (month 6). The fungus is not soil-borne, however, it is possible that selection of some individuals of *S. cirsii* capable of living in the sterile soil took place. [This work was funded by EU, project FOOD-CT-2003-001687, 2E-BCAs in Crops.]



## **Do *Chondrostereum purpureum* and *Fusarium tumidum* have potential as mycoherbicides for gorse?**

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*Chondrostereum purpureum* and *Fusarium tumidum* occur naturally on gorse (*Ulex europaeus* L) in New Zealand. Both pathogens have been associated with disease in this woody weed in the pastures and natural environments that it invades, and we have begun to explore their potential as mycoherbicides. Here we discuss the results of two experiments (Bourdôt et al. 2005) in which the response of gorse to these pathogens was evaluated. In the first experiment we found that summer-autumn (Feb-May) or late winter-early spring (Aug-Sept) applications of agar cultures of *C. purpureum* were effective on decapitated gorse stems, halving stem stump survival (from an average of 56% to 29%). In the second experiment, in which *F. tumidum* spores were applied in an invert emulsion to the shoots on gorse stems regenerating following decapitation, with and without wound treatment with *C. purpureum*, there was no evidence of synergism between the two fungi. Each independently reduced the density of regenerating shoots on the decapitated stems by 39-63% averaged over the 12 months following their respective applications. In neither experiment did the effects progress beyond the treated stems. The results confirm that both pathogens have potential as mycoherbicides for gorse. The contrasting modes of action of these two fungi (*C. purpureum* invading wounds on woody tissue and *F. tumidum* affecting young foliage) could be exploited by applying them to gorse plant stumps and regrowth foliage respectively. New research is planned to investigate this approach.

Bourdôt, G.W.; Barton, J.; Hurrell, G.A.; Gianotti, A.; Saville, D. 2005: *Chondrostereum purpureum* and *Fusarium tumidum* independently reduce regrowth in gorse (*Ulex europaeus*). *Biocontrol Science and Technology* Submitted March 2005

## **Selective inhibition and promotion of ryegrass, wheat and canola by rhizobacteria from Australian soils**

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Deleterious rhizobacteria (DRB) have been demonstrated to selectively suppress the growth of several annual weeds in North America; however, limited research has been conducted on the use of DRBs for biological control of weeds in Australia. Thirty six rhizobacteria isolates, from several Australian soils with varying crop histories, were screened for impacts on shoot and root growth of rigid ryegrass (*Lolium rigidum*), wheat (*Triticum aestivum* 'Janz') and canola (*Brassica rapa*). Rhizobacteria were grown in nutrient broth for 48 hours at 25C then diluted to approximately  $1 \times 10^7$  cfu/ml. Ten seeds of each species were placed in Petri dishes with 5 ml of diluted rhizobacteria cultures. Root and shoot growth were measured when roots in water controls reached approximately 40 mm in length. Data were converted to percent of the controls and compared using analysis of variance; means were separated using a least significant difference procedure. Means were also subjected to cluster analysis. Root and shoot growth responses differed among species and isolates. Several isolates promoted root and shoot growth; fewer isolates inhibited both root and shoot growth. Isolates were also identified that selectively inhibited ryegrass root growth with little or no effect on wheat or canola. Although further research will be needed to identify isolates with adequate efficacy and safety for field use, these data demonstrated that it is possible to select rhizobacteria from native Australian soils that selectively promote or inhibit ryegrass, canola, and wheat root or shoot growth.

## Spray retention and its impact on bioherbicide efficacy

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Spray retention is often used as an indicator in herbicide delivery, but little is known about retention characteristics of fungal spores used for weed biocontrol. This study examined spore retention of three bioherbicide agents, *Pyricularia setariae*, *Colletotrichum* sp., and *C. gloeosporioides* f. sp. *malvae*, on their respective weed targets: green foxtail, scentless chamomile and round-leaved mallow. Spore suspensions, containing a sodium fluorescein tracer dye (2.5 ml/L), were applied at 500, 1000 and 2000 L/ha using a cabinet sprayer, and the liquid volumes as well as spores retained on the plants were quantified. On all three weed species, liquid and spore retention showed a high degree of correlation with increasing application volumes although differences existed depending on the weed and volume used. Liquid retention reflected spore retention more correctly on green foxtail and scentless chamomile but might overestimate the number slightly on round-leaved mallow, possibly due to different plant morphology and architecture as well as spray run-off patterns. There was a general trend for finer droplets to result in higher spray retention at the same application volume, but this retention difference was not consistently translated into efficacy enhancement for the three weed-bioherbicide systems tested. Possibly, higher retention increases are required for a more significant and consistent improvement in weed biocontrol.

## **Culture stability of *Phomopsis cirsii*, a potential biocontrol agent of *Cirsium arvense***

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A common problem of biocontrol agents is the loss of virulence during upscaling in submerged culture. In order to obtain a strain of *Phomopsis cirsii* with high stability, one-spore cultures of different Nordic strains of *P. cirsii* having different aggressiveness were prepared. Different culture traits, compatibility and DNA-profiles of the one-spore cultures were then compared. The aggressiveness of selected one-spore isolates was compared with that of the “mother” culture. Loss of aggressiveness was studied for correlation with changes in the DNA-profile.



## ***Nigrospora oryzae* associated with shoot tip death of *Arundo donax***

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Giant reed, *Arundo donax* L. [Poaceae], is a recent target for biological control. Plant stand density of giant reed in the Mediterranean basin is lower compared to California and Texas where it is considered an invasive weed. Observations of natural stands in France, Italy, Greece, Cyprus, and Morocco commonly show a shoot tip dieback. Dissection of the dead flag leaf reveals abundant large black spores at the point of necrosis on all samples. Based upon spore size these were identified as *Nigrospora oryzae*. This was confirmed by sequencing the ITS1 and ITS2 regions and comparing the sequences with a confirmed identified isolate acquired from a repository in France. Abundant large black spores can also be observed in older tissue of *A. donax* sampled in New Mexico, but the conidial size was smaller than any previously recorded *Nigrospora* spp. In addition, sequences of the New Mexico isolate do not match with the *N. oryzae* samples collected from giant reed in the Mediterranean. This leads to the conclusion that this is a previously undescribed *Nigrospora* sp. not pathogenic to giant reed. Attempts to fulfill Koch's postulate and infect *A. donax* with the *N. oryzae* isolates have been unsuccessful using classical inoculation techniques. Frequently observed in both Mediterranean and U.S. locations in the giant reed tissue is a mite species that is capable of translocating *Nigrospora* spores on its back. In cotton, it was shown previously that a mite was needed in order for infection to occur with *N. oryzae*. Work has been started to demonstrate this synergism in *A. donax* as well.

## Does wounding of gorse plants enhance *Fusarium tumidum* infection?

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Gorse (*Ulex europaeus* L.) is a serious weed in New Zealand. Wounding of gorse plants was assessed to determine if it enhances infection by *Fusarium tumidum* Sherb. This is an integral part of a long-term objective, using insects as deliberate vectors to disseminate spores of this pathogen to control gorse. Feeding and oviposition activities of these insects on the weed may provide wound sites for fungal entry, which could enhance infection. Plants used in this experiment were 1, 2, 4 and 8 months old. A fixed number of wounds per unit plant size were made in their stems, spines and leaves using needles. The plants were sprayed with a suspension of  $10^6$  *F. tumidum* conidia/mL immediately after wounding. The mean dry weights of wounded plants, which were not sprayed with the pathogen and that of the untreated control, were similar across all age groups at  $5.6 \pm 0.70$  (SEM) g/plant. Wounding enhanced *F. tumidum* infection of all the gorse plants irrespective of their age at treatment. Wounded plants (1-4 months old) which were treated with the pathogen were shorter and had lower dry matter weight than the untreated control ( $P < 0.001$ ). Plant mortality by *F. tumidum* infection was higher in the 1 and 2-month old wounded plants compared with non-wounded plants. Wounding increased tip dieback infection in the 4 and 8-month old plants but not in the younger plants of which both wounded and non-wounded plants had 100% tip dieback. It is clear that wounding of older tissue will be required to facilitate *F. tumidum* infection of mature gorse plants.

# Pattern and potentiality of fungal isolates from soil rhizosphere and from young *Orobanche crenata* infesting *Vicia faba* fields in the south of Egypt

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## Introduction

World production of faba bean (*Vicia faba* L.) is concentrated in nine regions and especially in the Mediterranean and in the Egyptian Nile valley. Pathogens and the parasitic plant *Orobanche crenata* Forsk are among biotic factors constraining crop productivity and quality. Reductions in height, root length, number of leaves and mature pods of infested faba bean, were recorded by Abdel-Hafeez (1981). In Egypt, attention has been drawn to the broomrape problem in the mid 1960s when faba bean production dropped because of heavy infestation with *O. crenata*. ICARDA (1990) estimated the Egyptian yield loss in faba bean due to *O. crenata* as 5–24%. This paper records the variation in fungal pattern of soil rhizosphere and of *crenata* broomrape in infested fields of faba bean in Beni-Suef governate and records the results *in vitro* for their culture filtrates bioassay on *O. crenata* seed germination.

## Materials and methods

**Field survey.** A field survey conducted in the governrate of Beni-Suef (south of Cairo) in the early winter of 2001 showed fields with moderate level of infestation. *Crenata* broomrape was present in nearly half of the faba bean fields surveyed and fields with high infestation (2–3 *Orobanche* shoots per host plant) have been also recorded.

**Fungal isolation from *Vicia faba* rhizosphere and from young *O. crenata* shoots.** Fungi were isolated from young stalks of the parasite to assure high proportion of primary pathogens. Isolation from the soil rhizosphere was carried out by collecting soil from the studied localities to fill 43 pots (7 kg pot<sup>-1</sup>). Three sets of pots in addition to the control were used where set 1 contained *O. crenata* seeds mixed thoroughly into the soil to a depth of 10–12 cm while set 2 contained the host seeds planted at a depth of 10 cm. In set 3, soil was mixed with *O. crenata* seeds before planting of *Vicia faba*. Isolation of fungi was carried out every 15 d from duplicate pots until the podding stage of *Vicia faba*. Areas below the hypocotyl were detached, dipped into sterile distilled water and continuously shaken for 10 min; 0.5 ml of each dilution (10<sup>-1</sup> – 10<sup>-3</sup>) was inoculated on to plates of potato dextrose agar (PDA), sealed and incubated at 28°C for 1 week. Fungal colonies with different macroscopic characteristics were sub-cultured and pure cultures were maintained on PDA slants at 4°C.

**Preparation of *O. crenata* seeds for bioassay.** To a solution of Tween 20 (0.1% w/v), *O. crenata* seeds (3 mg) were added, 150 µl of this suspension was spread on to agar plates and number of seeds weighing 1 mg was calculated. Seeds in miracloth bags were immersed in 80% ethanol for 1 min then in 1% NaOCl for 10 min before rinsing in sterile distilled water and drying overnight. Seeds were kept in aluminium foil at 22°C for 19 d.

**Culture filtrate bioassay.** Fungal isolates were grown in still culture on 200 ml of liquid Czapeck-Dox and cations agar and 1 ml of each was removed under sterile conditions at 3 d intervals up to 2 weeks. Filtrates were sterilized using 0.22 µm filters. One ml of the test filtrate and of GR24 (1 µg ml<sup>-1</sup>) was used to wet the GF/D in each petri dish. Plates were sealed and incubated for 1 week and germination percentage scored.

**Macroscopic and microscopic identification of fungal isolates.** Active fungal isolates were identified using slide culture technique. Inoculated Petri dishes were sealed, incubated at 25°C and examined. Standard manuals, e.g. Booth (1971) were used for isolate identification.

## Results and discussion

Fungal isolates (188) from *Vicia faba* soil rhizosphere and 26 from the young parasite shoots were obtained and identified. Different *Aspergillus* and *Fusarium* species were isolated from the rhizosphere and from the parasite shoots. *Drechslera*, *Penicillium* and *Myrothecium* species were common in the soil rhizosphere, while *Alternaria* species were isolated from *Orobanche* shoots but not from rhizosphere flora. The methods used did not allow the identification of some isolates to the species level. Screening the culture filtrates of these isolates against the germination of *O. crenata* seeds gave 70 active isolates with medium or high potentiality for *O. crenata* seed control (Table 1). The rest of rhizosphere isolates (causing < 100% inhibition) were not considered in addition to some potential isolates from the parasite shoots which inhibited seed germination either by 16–24% (eight isolates: two of *F. moniliform*, one of *A. glaucus* and of *A. versicolor* and four of *A. niger*) or by 29–38% (six isolates of *Alternaria*). Highest inhibition values were recorded for the 3-day-old culture filtrates of all tested isolates compared to those recorded for the 6-, 9-, 12- and 15-day old cultures (except values recorded for 10 *Myrothecium* isolates). Culture duration affected the germination inhibition activity of the tested isolates and reduced the number of active isolates (having a medium or a high potential on assay) between 28 and 53%. Macroscopic and microscopic characteristics revealed a similarity of some isolates and allowed their grouping later. An example is *M. verrucaria* which comprised 10 of the 58 highly active isolates of which the culture filtrates of six of them completely inhibited germination of *O. crenata* seeds when they were 9-, 12- and 15-day old and even when filtrates were diluted up to 16-fold. Their studied characteristics showed them to be similar and the DNA sequence data of them provided detailed information to the species level (El-Kassas, 2003). There is no literature on treating *Orobanche* seeds by the 33 isolates of *Drechslera spicifera*, *Aspergillus flavus*, *A. fumigatus*, *A. wentii* or *Penicillium* species recorded.

Table 1. Activity of rhizosphere and *O. crenata* isolates on *O. crenata* seed germination

No. of isolates from	Fungi identified	% of <i>O. crenata</i> seeds germinated	Potential*
<b>Rhizosphere</b>			
1	<i>Drechslera spicifera</i>	0%	H
14	<i>A. flavus</i>	0%	H
1	<i>A. fumigatus</i>	0%	H
11	<i>A. niger</i>	0%	H
15	<i>A. wentii</i>	0%	H
2	<i>Penicillium</i> sp.	0%	H
4	<i>Fusarium</i> sp.	0%	H
10	<i>Myrothecium</i> sp.	0%	H
<b><i>O. crenata</i></b>			
3	<i>F. oxysporum</i>	29%	H
3	<i>F. oxysporum</i> Schlecht.	38%	M
6	<i>F. solani</i> (Mart.) Sacc.	45%	M

\* Potential of 3-day old cultures for *Orobanche* control: H, high (70–100%); M, medium (30–69%)

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# Toxins produced by pathogenic fungi of grass weeds as potential natural herbicides

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## Introduction

Research carried out to evaluate the ability of grass weed pathogenic fungi to produce toxic metabolites to be used as new natural herbicides in their native forms or as derivatives and analogues led us to collect many strains from several fungal collections. Among them, some strains of the genus *Drechslera* proved to be of noteworthy interest (Fracchiolla, 2003). In particular, a strain of *Drechslera siccans*, a pathogenic fungus isolated from seeds of *Lolium perenne*, was able to produce phytotoxic metabolites, when grown in a minimal defined medium.

## Materials and methods

**Chemical.** Optical rotation was measured in  $\text{CHCl}_3$  solution on a JASCO 1010 digital polarimeter; IR spectra were determined as neat on a Perkin-Elmer Spectrum ONE FT-IR and UV spectra was recorded in MeCN solution on a Perkin-Elmer Spectrometer Lambda 25.  $^1\text{H}$ - and  $^{13}\text{C}$ -NMR spectra were recorded at 600, 400, and at 150 and 100 MHz, respectively, in  $\text{CDCl}_3$  on Bruker spectrometers. The same solvent was used as internal standard. Carbon multiplicities were determined by DEPT spectra. DEPT, COSY-45, HSQC, HMBC and NOESY experiments were performed using Bruker microprograms. EI MS were taken at 70 eV on a Fisons Trio-2000. HR Electrospray and ESI MS were recorded on a Cetaf Micromass and on a Perkin-Elmer API 100 LC-MS; a probed voltage of 5300 V and a declustering potential of 50 V were used. Analytical and preparative TLC were performed on silica gel (Merck, Kieselgel 60  $\text{F}_{254}$ , 0.25 and 0.50 mm, respectively) or reverse phase (Whatman, KC18  $\text{F}_{254}$ , 0.20 mm) plates; the spots were visualised by exposure to UV radiation and/or iodine vapours and by spraying 0.5% ninhydrin in  $\text{Me}_2\text{CO}$  and/or chromosulphuric acid followed by heating at  $110^\circ\text{C}$  for 10 min. CC: silica gel (Merck, Kieselgel 60, 0.040–0.063 mm). Solvent systems:  $\text{CHCl}_3$ -*iso*-PrOH (9:1); EtOAc; EtOAc-MeOH (4:1); EtOAc-MeOH (19:1); EtOH- $\text{H}_2\text{O}$  (1.5:1).

## Fungal strain, culture medium and growth conditions

A strain of *D. siccans* (Drechsler) Shoemaker, isolated from diseased seeds of *Lolium perenne* was kindly supplied by Dr. József Bakonyi, Plant Protection Institute, Hungarian Academy of Science, Budapest, and stored as a single spore culture (ITEM 6217) in the collection of the Istituto di Scienze delle Produzioni Alimentari, CNR, Bari, Italy. The fungus was maintained on potato-dextrose agar medium. For the production of toxic metabolites, Roux bottles (1 L) containing a mineral defined medium (200 ml) were seeded with mycelium fragments of actively growing colonies. The cultures were incubated under static conditions at  $25^\circ\text{C}$  in the dark for 4 weeks, then filtered, assayed for phytotoxic activity and lyophilised for the successive purification steps.

## Results and discussion

The ethyl acetate organic extracts of *D. siccans* (667 mg) obtained from culture filtrates (4 L) appeared to be very interesting for their phytotoxicity on host and non-host plants and for their chromatographic profile.

These extracts were fractionated using a combination of column and TLC (on silica gel and reverse phase) with different eluent systems to give the main phytotoxic metabolite as homogenous oil ( $2.4 \text{ mg L}^{-1}$ ).

The chemical structure of this main phytotoxin was determined using a combination of spectroscopic methods (IR, UV and essentially mono and bi-dimensional  $^1\text{H}$ - and  $^{13}\text{C}$ -NMR and MS) and it was characterized as 5,12a-trimethyl-2,5,5a,12a-tetrahydro-1*H*-naphtho[2',3':4,5]furo[2,3-*b*]azepin-2-one, a new phytotoxic trisubstituted naphthofuroazepinone, which was named drazezinone (Figure 1) (Evidente et al., 2005).

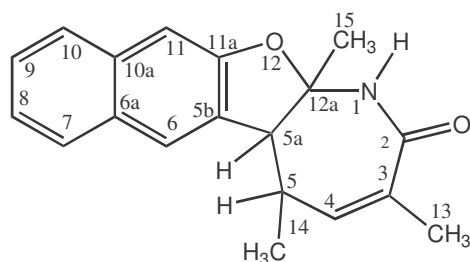


Figure 1. Chemical structure of drazezinone

Applied to wounded leaves, the toxin caused necrosis on almost all the species tested. The size of the necrosis produced ranged from very large, as in the case of *Urtica dioica*, to small ones, produced by applying the toxin to *Setaria viridis* and *L. perenne* leaves. The necrosis on *Euphorbia helioscopia* and *Mercurialis annua* leaves, both Euphorbiaceae, and *Chenopodium album* were also interesting. On the other hand, *Amaranthus retroflexus* and *Bromus* sp. were completely unaffected by the toxin. Drazezinone showed no antifungal (to *Geotrichum candidum*) and antibacterial (to *Pseudomonas syringae* and *Lactobacillus plantarum*) activities, and proved to have only low zootoxicity (to *Artemia salina* brine shrimps) (Evidente et al., 2005). Considering the original chemical structure of drazezinone, which is the first natural compound containing the naphthofuroazepin skeleton, its interesting phytotoxic activity, the low activity against fungi and bacteria, and the relatively low zootoxicity, further studies are in progress to evaluate its possible use as an environmentally friendly and safe herbicide.

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# Fungal metabolites for management of *Orobanche ramosa*

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## Introduction

Recently, many fungi were isolated from diseased *Orobanche ramosa* plants during extensive field surveys carried out within a national project, and some of them proved to be promising potential mycoherbicides for biological control of broomrapes (Boari & Vurro, 2004). Fifty-three isolates tested for virulence were also grown *in vitro* both on liquid and solid media with the main aim to find new metabolites having the ability to inhibit the induced germination of *O. ramosa* seeds. All the extracts from the liquid culture were assayed for the ability to inhibit seed germination, and most of them proved to be ineffective or only slightly active. Only the extracts produced by five strains (*Fusarium compactum*, *Myrothecium verrucaria*, *Alternaria* sp., *F. equiseti* and *Fusarium* sp.) were highly effective, causing the total or nearly complete inhibition of germination and were further considered as sources of new natural compounds (Abouzeid et al., 2004). In this study we have investigated the production, the purification and the chemical and biological characterization of the metabolites produced by *M. verrucaria* and *F. compactum*.

## Materials and methods

**Fungus.** *Myrothecium verrucaria* and *Fusarium compactum* were isolated during extensive surveys in fields in the South of Italy heavily infested by *O. ramosa* (Boari & Vurro, 2004). The isolates are stored in the collection of the Istituto di Scienze delle Produzioni Alimentari, CNR, Bari, Italy, as ITEM 6168 and ITEM 6160, respectively.

**Growth conditions.** A conidial suspension (1 mL, containing approximately 10<sup>6</sup> conidia) was added to 1 L Roux bottles containing 200 mL of the mineral medium M1-D for the production of toxic metabolites. The cultures were incubated under static conditions at 25 °C in the dark for 4 weeks.

**Chemical experiments.** The phytotoxic metabolites were purified from ethyl acetate extracts (100% inhibition of seed germination). *M. verrucaria* extracts were fractionated by CC eluted with CHCl<sub>3</sub>-*iso*-PrOH (19:1). The residues of seven homogeneous fractions were purified by some steps on preparative TLC in silica gel and reverse phase using different eluent systems [CHCl<sub>3</sub>-*iso*-PrOH (19:5) and (19:1), EtOAc-*n*-hexane (1.5:1) and (4:1), petroleum ether-Me<sub>2</sub>CO (1.5:1) and EtOH-H<sub>2</sub>O (1:1)]. The *F. compactum* extracts were fractionated by CC eluted with CHCl<sub>3</sub>-*iso*-PrOH (9:1) to yield eight groups of homogeneous fractions. Only the second fraction proved to be highly phytotoxic, causing 100% inhibition when assayed on *O. ramosa* seeds germination. The residue of this fraction, containing the main metabolite was further purified by TLC on silica gel [eluent petroleum ether-Me<sub>2</sub>CO (2.3:1)].

The physical and spectroscopic properties of the purified metabolites were determined. In particular: optical rotation was measured and infrared (IR), UV, <sup>1</sup>H and <sup>13</sup>C nuclear magnetic resonance (NMR), electron ionization (EI) and electrospray ionization MS (ESI) spectra were recorded.

## Results and discussion

*M. verrucaria* and *F. compactum* grown in liquid culture produced metabolites that inhibited the germination of *O. ramosa* seeds at 1-10 μM.

The phytotoxic metabolites were purified from ethyl acetate extracts of the fungal culture filtrates (100% inhibition of seed germination) and isolated using a combination of chromatographic technique (column and TLC). Their chemical structures were identified using a combination of spectroscopic methods (IR, UV, mono and bi- dimensional  $^1\text{H}$  and  $^{13}\text{C}$  NMR) and EI- and ESI-MS). From *M. verrucaria*, seven compounds were isolated in very low concentrations and identified as macrocyclic trichothecenes, namely verrucarins A, B, M and L acetate, roridin A, isotrichoverrin B and trichoverrol B (Figure 1). The main metabolite isolated from the same extracts was identified as verrucarin E, a disubstituted pyrrole (Figure 1). From *F. compactum* extracts, neosolaniol monoacetate was isolated and identified (Figure 1). It also belongs to the trichothecene family. All the trichothecenes proved to be potent inhibitors of *O. ramosa* seed germination and possess strong zootoxic activity when assayed on *Artemia salina* brine shrimps. Verrucarin E was inactive on both seed germination and zootoxic assay.

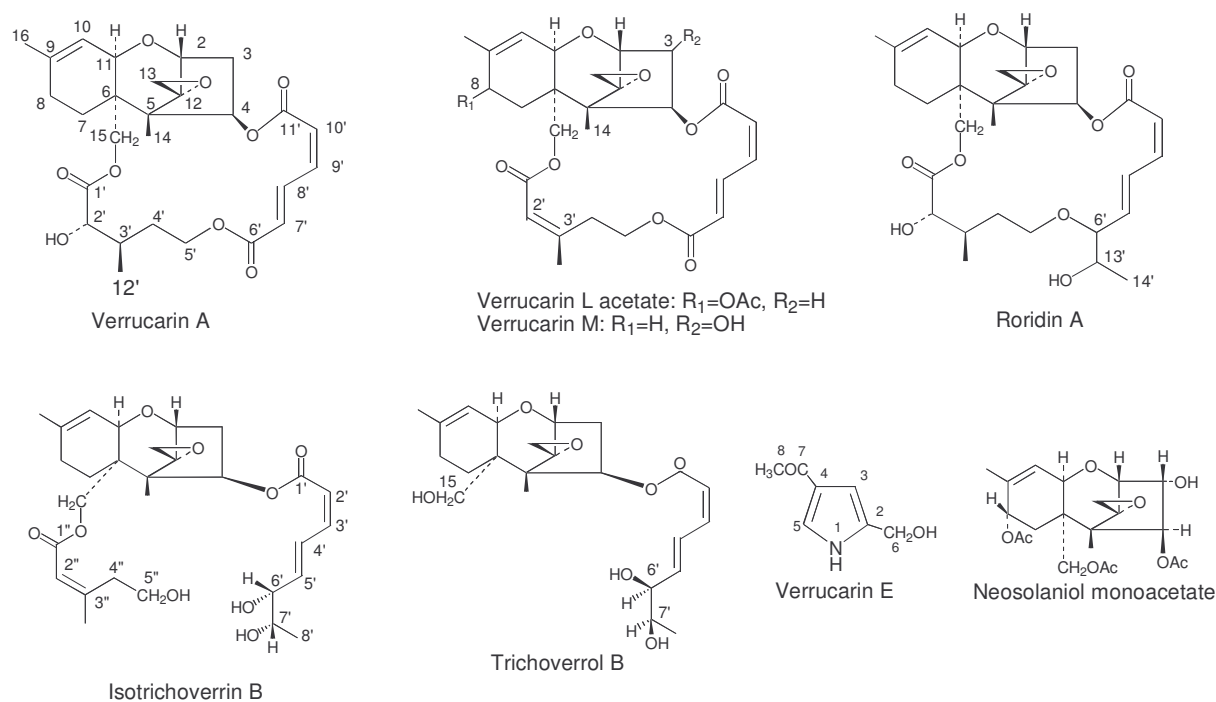


Figure 1. Chemical structures of metabolites isolated from *M. verrucaria* or *F. compactum*

This is the first time the above-mentioned metabolites have been reported to be produced by fungal strains isolated from infected tissues of *O. ramosa*. Their potential practical application as natural herbicides for the management of seed germination has been proposed (Andolfi et al., 2005).

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## Biological control of aquatic weeds of rice in Australia

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Eighty-five percent of Australia's annual rice production of 1.2 million tonnes is exported and as such contributes significantly to the gross national product of the country. Australia's rice production is comparatively pest and disease free with only a small number of relatively minor diseases and insects being recorded (Lanoiselet et al., 2001a; 2002). However, aquatic weeds have the potential to compete heavily with direct-seeded rice under Australian conditions. Plant species in the family Alismataceae including *Damasonium minus* (R.Br.) Buch., *Alisma lanceolatum* and *A. plantago-aquatica* are significant weeds of rice in Australia. Of these species, *Damasonium minus* or starfruit is regarded as the most important weed. The control of starfruit is almost exclusively reliant on the use of only one herbicide (Londax®), which has contributed to the emergence of herbicide-resistant weed biotypes throughout Australian rice growing areas. This resistance and the potential for the contamination of waterways by synthetic herbicides have spurred the search for alternative weed control strategies.

An endemic fungus, *Rhynchosporium alismatis*, was first observed causing necrotic leaf spots on *D. minus*, *A. lanceolatum* and *A. plantago-aquatica* in 1992 (Cother et al., 1994) and its potential as an inundative biological control agent was identified (Cother & Gilbert, 1994a, b). As part of host range studies, Cother (1999) reported that the fungus could cause symptoms on a range of aquatic species as well as crops such as cucumber, rockmelon, soybean and tomato under extremely severe conditions. However, even under these conditions the pathogen did not appear to have an effect on plant development. On *D. minus* the fungus causes dark necrotic lesions on the leaves, stems and petioles (Cother et al., 1994), which may lead to reduction in seed production and viability (Fox et al., 1999). Later studies by Jahromi et al. (2004) have also shown that the fungus may act as a mycoherbistat (Crump et al., 1999) by reducing plant growth without actually causing lesions on juvenile plants of *D. minus*.

The fungus produces both conidia and chlamydospores (Lanoiselet et al., 2001b). Early studies have shown that the media used has a marked affect on the number and infectivity of spores produced by the fungus (Jahromi et al., 1998; Cother & van de Ven, 1999) with lima bean agar proving to be the best medium for conidiospore production. A high concentration of malt extract (4.4 g L<sup>-1</sup>) as the sole carbon source and a high level of sodium nitrate as the sole nitrogen source (3.3 g L<sup>-1</sup>) were shown to increase chlamydospore production while agitation (150 rpm) enhanced conidial yields. Maximum chlamydospore production (5.09 x 10<sup>6</sup> ± 1.9 total chlamydospores mg DW<sup>-1</sup>) was achieved in cultures grown in a medium supplemented with 8.8 g L<sup>-1</sup> malt extract and 5.74 g L<sup>-1</sup> sodium nitrate (Cliquet et al., 2004). Germination of chlamydospores (90%) was significantly higher than germination of conidia (47%) after 2 days growth. Conidiospores of the fungus germinate and penetrate the leaves of the weed using appressoria within 4 hours of inoculation (Jahromi et al., 2002b) with the optimum temperature occurring between 25 and 30°C. Penetration through stomata occurs at low frequency and appears to be a random event. Secondary conidial formation occurred within 48 hours of inoculation. On species of Alismataceae considered to be non-hosts of *R. alismatis*, spore germination occurred but the rate of penetration and appressorial production was reduced (Pitt et al., 2004b).



The genetic diversity within *R. alismatis* as determined by ERIC-, REP- and ISSR-PCR was found to be low within and between populations of the fungus from southeastern Australia (Pitt et al., 2005). Sequencing of the internal transcribed spacer from the fungus revealed a close relationship with the *Plectospheralla* genus and so the fungus has been transferred to the species *Plectosporium alismatis* (Oudem.) W.M. Pitt, W. Gams & U. Braun (Pitt et al., 2004a). Similarly, the genetic diversity within *D. minus* was found to be low (Jahromi et al., 2002a). However, the genetic diversity with *Alisma lanceolatum* (an introduced species) was higher between populations within the rice growing area in Australia (Ash et al., 2004). This may reflect a number of introductions of the weed, which could harbour resistance to the pathogen.

The action of the fungus on the weed in both the glasshouse and the field has been shown to be enhanced by the addition of low rates of herbicides. Glasshouse experiments indicated evidence of synergism between the fungus and Londax<sup>®</sup>. Starfruit growth was suppressed when 1.5% of the recommended field rate was applied before fungal inoculation using a conidial suspension in water. Glasshouse and field trials showed that fungal inoculation can reduce the weed competition and indicated that fungal inoculation in combination with sublethal doses of Londax can be effective in reducing starfruit growth and its competition with rice (Jahromi et al., unpublished).

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## Inundative biological control of *Carthamus lanatus* in Australia

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*Carthamus lanatus* L. (saffron thistle) is a winter growing annual with erect rigid stems up to 1 m high (Meadley, 1965). It was introduced into Australia from the Mediterranean (Parsons, 1973). Since its introduction it has spread throughout Victoria, South Australia, Western Australia, Queensland and New South Wales (Peirce, 1990) and it is now considered the most economically important thistle in New South Wales (Briese, 1988; Sindel, 1996; Ayres, 1997). The species displays considerable phenotypic and genetic diversity in Australia (Peirce, 1990; Ash et al., 2003). Despite the implementation of control measures, saffron thistle has continued to spread (Sindel, 1996). The difficulty with controlling saffron thistle by conventional means has made this weed a potential target for biological control (Briese, 1988). Saffron thistle is both a target for classical (Briese, 1988) and inundative (Crump et al., 1996a, b) biological control. However, the close relationship to the cultivated crop plant safflower (*C. tinctorius*) has created difficulties in the implementation of the classical form of biological control (Wapshere, 1987; Morin & Jourdan, 2001). In this paper we report on progress on the development of a use of a species of *Phomopsis* as a mycoherbicide for this weed.

A species of *Phomopsis* has been isolated from the weed in Australia and is currently under investigation for use in a form of inundative biological control (Crump et al., 1996a, b). The fungus has been shown to be very aggressive on saffron thistle as well as most other species in the family Asteraceae including the crop plants lettuce and sunflower. Host range studies conducted in the glasshouse have shown that the fungus does not affect other crop plants tested including wheat, barley, rapeseed and many pasture species. Growth room experiments have also shown that the fungus requires an extended dew period of up to 24 h to infect and cause damage to many plants. Preliminary research focussed on the use of liquid formulations of the fungal mycelium but this was considered impractical in the field. This *Phomopsis* species sporulates poorly and so efforts have concentrated on the formulation of the mycelium of the fungus. The fungus can be produced in submerged culture or in semi-solid fermentation.

Semi-solid fermentation was undertaken using a range of whole and cracked grains (wheat, millet and rice). The fungus was found to colonise the millet entirely in 4 days and the intact seed coat provided structure to the colonised grain. The optimum drying time for the granules was determined and a shelf life of up to 6 months for the dried product at room temperature was found. Experimentation is continuing with the use of coatings applied to the colonised millet of low rates of herbicides (which have been previously found not to affect the growth of the fungus). This low tech and low cost method may be targeted at poorer communities for use of biological control for Asteraceae weeds.

An alternative protocol for formulation using microencapsulation has been developed. The successful spray-drying of particles establishes this new technology as one which may be utilised in the biological control arena. After viability of the fungus, the yield of the formulated fungus was considered most important, as this will greatly influence the cost of such a process. The optimum water activity of the formulations has also been established. This is a particularly important parameter that will affect the ability of the fungus to survive

and regenerate. It is also a factor in the exclusion of conditions where bacteria may flourish and therefore spoil the product. Continuing from this, the spray-drying of the fungi in gelatine capsules has also been developed. In this process, fungal particles are encapsulated before being spray-dried in a two-step process to further strengthen the capsules. These may serve as an artificial spore in the field environment. By the addition of outer layers and nutrients the capsules the fungal particles are more likely to survive and infect the target species.

Due to the variability in many of the morphological characteristics of *Phomopsis* species, researchers have placed emphasis on host affiliation to define species. However, the presence of two morphological types of *Phomopsis* on twigs and bark of *Ulmus* species in the United Kingdom and Italy was demonstrated (Brayford, 1990). These groups were also isolated from *Acer pseudoplanatus* L., *Fagus sylvatica* L. and *Fraxinus excelsior* L., demonstrating the potential of a *Phomopsis* species to infect more than one host, indicating that emphasis on host affiliation as a basis for defining species of *Phomopsis* is unreliable. This bias towards naming of species of *Phomopsis* based on host affiliation has led to an unnecessary proliferation of species (Rehner & Uecker, 1994). Interspecific variation is difficult to determine within a collection of *Phomopsis* isolates due to the plasticity of morphological and physiological characteristics, therefore other characteristics have been used. Thus, the variability in the species of *Phomopsis* isolated from saffron thistle was examined using PCR-based methods and this information used in selecting isolates in which the sequencing of the rDNA region was used to assist in the identification of the fungus. From this analysis it is hypothesised that the *Phomopsis* isolated from saffron thistle is a unique species, different from *Phomopsis* spp. found to be pathogenic on crop plants in Australia.

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# Is it safe to use fungal pathogens as classical biological control agents against weeds? Results from a retrospective analysis

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## Introduction

Given the misery that plant diseases can cause, e.g. the Irish Potato famine, it's not surprising that the practice of introducing exotic plant pathogens to control weeds makes some people nervous. However, it is their ability to cause devastating disease outbreaks that gives pathogens so much potential as biological control agents. Regulatory authorities weigh up the risks of harming nontarget species against the benefits of controlling a target weed, and results from host range testing are vital to this assessment, but how well can these tests, which are usually undertaken in a glasshouse, predict the behavior of pathogens outdoors in a new environment? Also, is it possible that a pathogen that appears to be safe in such tests could later evolve to attack a new host? The purpose of this study is to review the performance (with respect to nontarget attack) of the plant pathogens released as classical biological control agents to date, and compare that with predictions made on the basis of pre-release testing. This will allow the safety of this method of weed control to be assessed.

## Materials and methods

A list was compiled of all the pathogens that have been released as classical biological control agents against weeds. Information was then collected on their host-range test results and their behavior in the field after release. The resulting data were tabulated and used for a retrospective analysis (Barton (née Fröhlich), 2004).

## Results and discussion

If a 'project' is defined as the release of one exotic pathogen against one weed (or weed complex) in one country, then worldwide there have been 32 such 'projects' to-date. Twenty-six pathogens, all of them fungi, have been released against more than 26 species of weeds in seven countries. Nontarget damage in the field was observed in only three cases: (1) the gall rust *Uromycladium tepperianum* that was introduced to South Africa to control Port Jackson willow (*Acacia saligna*), formed a few abnormal galls on the nontarget species *Acacia cyclops*; (2) the rust *Puccinia carduorum*, an agent for musk or nodding thistle (*Carduus thoermeri*), formed a single pustule on a single plant of the nontarget globe artichoke (*Cynara scolymus*) in a field trial in the USA; and (3) the rust *Puccinia melampodii*, a pathogen of parthenium weed (*Parthenium hysterophorus*), caused symptoms on an Indian variety of *Calendula officinalis* in test plots in Australia.

Host range tests conducted on the Acacia gall rust before it was introduced to South Africa led to the conclusion that it would probably form galls occasionally on the nontarget species *Acacia cyclops* (Morris, 1987). While *A. cyclops* provides firewood to poorer communities in South Africa, it is an alien species with invasive tendencies and a decision was made that minor damage to that species was a price the authorities were willing to pay in order to bring *A. saligna* under biological control. Since its release in 1987 the rust has proven to be a highly effective biocontrol agent (Morris, 1997). It has sometimes caused abnormal galls on *A. cyclops*, but only where this species grows near heavily infected *A. saligna* plants. The

galls do not spread or multiply on *A. cyclops* and so this predicted nontarget damage is not of concern (M.J. Morris, Microbial Products, South Africa, personal communication).

Host range tests of the thistle rust (*P. carduorum*) showed that it could infect globe artichoke, which belongs to the same Family, Tribe and Subtribe as the target thistle, in the glasshouse (Politis et al., 1984). Further testing of the fungus was undertaken outdoors. A field trial ran for 2 years and during that time the only nontarget damage that occurred was a single rust pustule on one individual (out of 32 tested) of the artichoke (Baudoin et al., 1993). The fungus was subsequently released in the USA in 1987, and has spread well. It has not been found infecting artichoke, or any other nontarget species, since its release (W.L. Bruckart, USDA-ARS, Ft. Dietrick, MD, USA, personal communication).

There were plans to release the parthenium rust fungus (*P. melampodii*) in both Australia and India. Initial host range tests showed that it could infect some nontarget sunflower cultivars (Evans et al., 2001). After a risk assessment, Australian authorities decided the benefits of the introduction were likely to outweigh the risks and the rust was released in Australia in 1999. However, since no pathogens had ever been released as biocontrol agents in India, it was decided to do further testing, outdoors in Australia, before resolving on whether or not to release *P. melampodii* there (Evans et al., 2001). In field test plots the rust was found to cause disease symptoms on an Indian variety of *Calendula officinalis*. While this was the only nontarget damage ever observed as a result of the release of *P. melampodii* in Australia (A.J. Tomley, retired, formerly of Queensland Department of Natural Resources, Australia, personal communication), it was decided not to introduce it to India. It was feared that if *P. melampodii* attacked a crop plant, then the first release of a plant pathogen for biocontrol in India might also be the last (H.C. Evans, CABI Bioscience, UK, personal communication).

There were no examples in the literature reviewed of a pathogen introduced for biological control evolving to use a nontarget plant to a greater extent than was expected, or to be able to use a new host (Barton (née Fröhlich), 2004). The fact that plant pathogens evolve, as do all living organisms, does present risks of changes in host use. However, it must be remembered that the chances of an exotic pathogen evolving a broader host range after release are no greater than the chances that a native species will do so. In fact, it could be argued that exotic species pose less of a threat in this regard because they are less likely to be exposed to plants that were hosts during their previous evolutionary history (Van Klinken & Edwards, 2002).

Since the first deliberate release of a plant pathogen for classical biological control of a weed in 1971, there has not been a single case of unpredicted, post-release nontarget damage in the field. Thus, the answer to the query posed in the title of this article is ‘Yes’. As long as predictions of field host-range continue to be based on thorough host range testing and a good understanding of the taxonomy and ecology of the agent and the target weed, the use of plant pathogens should be a very low risk and environmentally benign method of weed control.

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## Evaluation of fungal pathogens for biocontrol of *Cirsium arvense*

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### Introduction

*Cirsium arvense* is a common weed in the European part of Russia (EPR) and a serious problem in spring cereals and some vegetable crops. The weed is difficult to eradicate without high doses of chemical herbicides. That is why it is a good target for development of a bioherbicide to facilitate weed control, especially in organic agriculture. The biocontrol potential of many fungal pathogens of *C. arvense* was evaluated in Europe, North America and New Zealand. The main pathogens are *Puccinia punctiformis* (Turner et al., 1986; Frantzen, 1994), *Sclerotinia sclerotiorum* (Brosten & Sands, 1986; Bourdôt et al., 1995), *Alternaria cirsinia* (Green & Bailey, 2000; Green et al., 2001), *Septoria cirsii* (Herschenhorn et al., 1993; Berestetskiy, 2000). A little is known about *Phoma* spp. (Bithell & Stewart, 2001; Guske et al., 1996), *Ramularia cynarae* and *Phomopsis cirsii*. Currently, none of these species has clear prospects of being used in practice. The main problems associated with them are low aggressiveness under natural conditions, difficulties in inoculum production, and wide host range. A possible solution is to find new pathogens/strains aggressive to the weed and compatible with fermentation technologies. Another way could be production and application of phytotoxic metabolites that depend on external factors to a lesser extent than living organisms. For these purposes, a collection of 140 fungal isolates was evaluated for virulence and phytotoxic activity.

### Materials and methods

Mycological herbarium material was sampled in different regions of EPR in 2002–2003. More than 200 fungi were isolated from dried diseased parts of *Cirsium arvense*. Of them, 140 isolates were tested by leaf disk bioassay for both virulence and phytotoxicity. Disks (diam. 1 cm) were cut from well-expanded leaves of *C. arvense*, placed in Petri dishes on moistened filter paper and subjected to 10  $\mu$ L of conidial suspension or culture filtrate. In the latter bioassay, the leaf disks were punctured. Symptoms were assessed 2–4 days after inoculation. Some selected isolates, which demonstrated both virulence and phytotoxic activity in the bioassays, were evaluated in greenhouse experiments on whole *C. arvense* plants. For each isolate, mycelial inoculum was produced on autoclaved millet. The well-colonized substrate was crushed and applied to the soil and plant base at the rate of 0.5–1 g pot<sup>-1</sup> (50–100 g m<sup>-2</sup>) with three plants (at the stage of 5–7 true leaves) produced from seeds.

### Results and discussion

The fungi *Septoria cirsii*, *Puccinia punctiformis*, *Ramularia cynarae* (syn.: *R. cirsii*) and *Ascochyta sonchi* were found to be common pathogens of *C. arvense* in EPR (Berestetskiy, 1997). Isolates tested belong to genera *Alternaria* (70 isolates) and *Fusarium* (30); others were referred to *Ascochyta sonchi* (10) and *Septoria cirsii* (20). Some isolates were preliminary identified as *Stagonosporopsis* sp. (10).

The most isolates tested from the genus *Alternaria* were from *A. tenuissima* (66). Culture filtrates of 26% isolates were phytotoxic; 10% of isolates were virulent. Identified *Fusarium* isolates belong to 15 species of the genus. It is interesting and important that *C. arvense* is a host plant for many toxigenic *Fusarium* spp. (e.g., *F. tricinctum*, *F. sporotrichioides*). A few isolates, C-23 (*F. proliferatum*), C-26 (*F. culmorum*), C-32 (*F. equiseti*), and C-57 (*F. oxysporum*), demonstrated high phytotoxicity by the leaf disc



bioassay. Isolates of pycnidial fungi were generally less phytotoxic but the most of them were virulent to the leaf disks of the weed. Summary data on phytotoxic activity of the studied fungi were: *Alternaria* > *Fusarium* > *Stagonosporopsis* > *Ascochyta* > *Septoria*. They did not correspond with virulence of the isolates: *Stagonosporopsis* > *Septoria* > *Ascochyta* > *Fusarium* > *Alternaria*.

Seven fungal isolates were selected for pathogenicity evaluation on whole plants. Considerable disease symptoms and fresh weight losses were caused by application of the following isolates: *F. oxysporum* (at the rate 50 g m<sup>-2</sup>), *Ascochyta sonchi* (100 g m<sup>-2</sup>) and *Stagonosporopsis* sp. (50–100 g m<sup>-2</sup>). Only infection of *C. arvense* by the last fungus led to significant reduction of dry weight of roots. The strains selected by Bailey et al. (2000) were able to kill the weed at the rate 250–500 g m<sup>-2</sup> in 4–6 weeks in greenhouse experiments. The experiments stress the potential of *Stagonosporopsis* sp. strains for biocontrol of *C. arvense*. The next step of the research is host-range study of selected isolates. For *Stagonosporopsis* sp., development of appropriate formulation is important.

Table 1. Effect of different pathogens on *Cirsium arvense* using application of dry powder preparation of inoculum on soil surface and plant base (14 dpi)

Fungal species	Isolate	Necrotic area, %		Fresh biomass, %		Dried roots weight, %	
				(control=100%)		(control=100%)	
		50 g m <sup>-2</sup>	100 g m <sup>-2</sup>	50 g m <sup>-2</sup>	100 g m <sup>-2</sup>	50 g m <sup>-2</sup>	100 g m <sup>-2</sup>
<i>Alternaria infectoria</i>	166	0.9 a	0 a	94.2	112.1	138.3	92.5
<i>A. tenuissima</i>	115	3.8 a	2.90 a	92.0	99.4	146.8	112.5
<i>Phyllosticta cirsii</i>	168	4.6 a	10.5 a	89.9	95.2	97.9	102.5
<i>Fusarium equisetii</i>	27	5.0 a	0.9 a	86.7	10.1	1192	105.0
<i>Ascochyta sonchi</i>	177	5.1 a	38.1 b *	73.4 *	79.7	1128	52.5
<i>Fusarium oxysporum</i>	57	24.6 b *	–	68.1 *	–	80.9	–
<i>Stagonosporopsis</i> sp.	163	73.3 c *	80.8 c *	36.7 *	27.0 *	46.8	22.5 *

Means following the same letter are not significantly different by Fisher's LSD test ( $P < 0.05$ ); means marked with asterisk are significantly different from control ( $P < 0.05$ ).

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# Biological control of *Orobanche ramosa* using *Fusarium* spp. and other fungi

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## Introduction

The genus *Orobanche* (broomrapes) includes many parasitic weeds species responsible for major losses to vegetables, legumes, sunflowers and many other plants. *Orobanche ramosa*, one of the main species of this genus, is a severe weed problem in fields cropped to Solanaceae, particularly tomato, tobacco, potato and eggplant, due to the withdrawal of water, minerals and organic compounds which leads to conspicuous yield losses. Difficulties in control of broomrape are due to the large amount of seeds produced, which are viable for years, and to the long damaging and unpredictable underground phase. Traditional control methods are inapplicable or only allow a modicum of control (Amsellem et al., 2001). Although biological control is considered an attractive approach for suppressing this parasitic weed, none of the proposed promising pathogenic isolates has reached the market. Considering the importance of *O. ramosa* in Italy, mainly on tomato, tobacco and cabbage, a research programme was funded by the Italian Ministry of Scientific Research to isolate and to identify pathogens of *Orobanche* spp. and to use them as biological control agents.

## Materials and methods

During extensive surveys in fields heavily infested by broomrapes in the south of Italy, 53 fungal strains belonging to 15 different species were isolated and stored in the ISPA collection.

For the assessment of pathogenicity and virulence all the strains were tested using a plastic-bag assay, by inoculating some broomrape tubercles with fresh colonies of the isolates and estimating the eventual appearance of symptoms (Boari & Vurro, 2004). The evaluation of symptoms was done by assigning a visual score ranging from 0, for nonpathogenic isolates, up to 3, for pathogens causing quick and complete necrosis of tubercles, swelling and loss of consistency.

The most effective strains were further evaluated in pot experiments in a greenhouse involving three different assays. Plastic pots were filled with about 3 kg of broomrape-free soil homogeneously mixed with 50 mg of *O. ramosa* seeds. Around 1 month after sowing tomato seeds, 100 ml of the suspension of conidia and mycelium was uniformly distributed on the soil surface of each pot. Four pots were inoculated for each isolate, with inoculum containing  $10^6$ – $10^7$  conidia ml<sup>-1</sup>.

In the first experiment, four strains were applied as described, and the number of emerged broomrape shoots and fresh and dry weight both of tomato plants and of broomrape shoots were determined. In the second experiment seven strains were tested. Among them, one strain of *Fusarium oxysporum* (FT2) was also inoculated a second time, 1 week after the first treatment. In the third experiment, five strains were assayed, with the isolate FT2 also applied 1 week before the planned treatment, or 1 week after. Number, length and fresh weight of emerged broomrape shoots were recorded, as well as fresh weight of tomato plants. Moreover, the soil from each pot was washed carefully to separate tomato roots and broomrape. The number of underground broomrape shoots and developing tubercles and their fresh weights were determined.

The best strains chosen as candidate mycoherbicides were also tested for specificity on plants belonging to Solanaceae, Apiaceae, Fabaceae and Asteraceae.

## Results and discussion

Most of the 53 isolates tested using plastic-bag assays caused no or very slight symptoms. Among them, only nine proved to be highly virulent causing severe damage, as early as 3–4 days after inoculation. In some cases, the pathogens rapidly caused browning of tubercles, with necrosis and rot. In a few cases, a thick layer of mycelia was produced, covering tubercles and stopping their growth. Symptoms were never observed on tomato roots. The best isolates were used in the pot assays.

In the first experiment, one isolate of *F. oxysporum* (FT2) reduced the number of emerged broomrape shoots by about 70% compared to the control. It also reduced the fresh and dry weight of emerged broomrapes. A strain of *F. sambucinum* was also quite active, causing around 60% reduction of the emerged shoots. *Fusarium camptoceras* (CT1) caused more than 50% reduction of the emerged shoots, whereas *Fusarium* sp. (BPo2) caused no reduction.

In the second experiment, the best results were obtained when FT2 was applied twice. Also noteworthy were the effects caused by an isolate of *F. proliferatum* (BT1), *F. chlamydosporum* (BPo3) and another of *F. oxysporum* (ACb6), all able to reduce the number of emerged shoots by more than 60%. Some of these species are also reported as pathogens of *O. ramosa* for the first time.

In the third experiment, FT2 and one isolate of *F. solani* (ET4) caused a strong reduction of the number of subterranean emerging shoots, as well as of the fresh weight of subterranean and aerial broomrape parts.

The two best isolates obtained, FT2 and ET4, were able to reduce the number of developing broomrape shoots and allowed the tomato to produce a larger and healthier root system, compared to the untreated control. Moreover, these species proved to be specific, causing no disease to plant species other than *O. ramosa*.

Most of the effective isolates belong to the genus *Fusarium*, which includes many common soilborne and seedborne pathogens. Considering that most of the damage caused by parasitic plants is produced before plant emergence, it is fundamental to have pathogens available that can grow through the soil and survive in it for a long time. Because the fungi could need time to move down and to colonize the host rhizosphere, this problem could be partially overcome by introducing spores or chopped mycelia by drip irrigation systems, allowing tubercle attachment at a very early stage, and preventing the development of the parasite. Experiments are in progress to evaluate this potential.

Studies are also in progress to evaluate the ability of the promising *Fusarium* strains to cause disease on other *Orobanchae* species, such as *O. crenata* (the only other species present in Italy), or *O. cumana* and *O. aegyptiaca*, two other very dangerous and damaging parasitic species. In fact a broader spectrum of action would render the mycoherbicide more suitable also from a commercial point of view.

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Boari, A. & M. Vurro, 2004. *Biol. Control* 30, 212–219.

# *Ranunculus acris* control in dairy pasture using *Sclerotinia sclerotiorum*

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## Introduction

*Ranunculus acris* (giant buttercup) is a widespread and persistent weed of dairy pastures in New Zealand, estimated to have cost the dairy industry NZ\$156 million in lost milk solids revenue in the 2001–02 milking season despite the use of chemical herbicides (Bourdôt et al., 2003). An alternative, biological control, approach using *Sclerotinia sclerotiorum* appeared promising when the fungus, applied as mycelium-infested wheat particles to axils of young leaves on three-month-old potted plants, caused 75% mortality (Green et al., 1993). Field experiments subsequently confirmed this potential, when, applied as a slurry to basal leaf axils of individual flowering plants at 40 ml plant<sup>-1</sup>, the fungus resulted in 57% reduction in plant dry mass (Cornwallis et al., 1999). The fungus was also effective when broadcast as mycelium-infested wheat granules at 600 kg ha<sup>-1</sup> in autumn and spring, reducing the ground cover of the buttercup by 46 and 63% respectively (Harvey & Bourdôt, 2001), and at 500 kg ha<sup>-1</sup> in spring, giving 63 and 13% mortality in two different pastures (Verkaaik et al., 2004).

For both commercial and practical on-farm handling reasons, a weed control product applied at such high rates as 500–600 kg ha<sup>-1</sup> would be unacceptable. To determine the extent to which application rate could be reduced without compromising efficacy, the current experiment manipulated both granule size (diameter) and application rate (kg ha<sup>-1</sup>).

## Materials and methods

*Experimental details.* Four experiments were established in *R. acris*-infested pasture, two on each of two dairy farms (Sites 1 and 2) in the Golden Bay dairying region of New Zealand; one at each site treated on 14–15 October 2003 and the other on 18–19 November 2003. A split plot design with two replicate blocks was used, with the mainplot treatment factor being wounding of the buttercup with a roller prior to application of 12 subplot treatments. The latter were a factorial combination of three granule sizes and three application rates (25, 50, 100 kg ha<sup>-1</sup>), a mix of the three granule sizes at 500 kg ha<sup>-1</sup>, the mix oven-killed as a control, and an untreated control. The three granule size classes (small, 1.0–1.5 mm; medium, 1.5–2.0 mm; large, 2–3 mm) were produced by sieving a mycelium-on-wheat formulation of *S. sclerotiorum*, WH2, prepared using isolate S36 as described by Verkaaik et al. (2004). The treatments were applied to five 2-m plots using a modified fertilizer spreader during the first ¼ of a 28-d grazing cycle. The percentage ground covered by *R. acris* was measured using point analysis (50 points plot<sup>-1</sup>) at time of treatment and 1, 3, and 6 months after treatment.

*Statistical analysis.* A “Before-After, Control-Impact” (BACI) calculation was employed (Green, 1979), in which the impact of *S. sclerotiorum* was assessed by comparing the changes in cover on treated and non-treated control plots between the assessments made before and after the treatment “impact”. The value  $(A_t/B_t)/(A_c/B_c) \times 100$  was calculated for each plot that was treated in October or November at both sites. Here *B* and *A* are *R. acris* ground cover at the time of treatment (before impact) and either 1, 3, or 6 months after treatment (after impact) respectively, and subscripts *t* and *c* denote treated and control plots respectively. The BACI value defined in this way is the cover on the treated plot expressed as a percentage of the cover on the control plot, adjusted for the ratio of treated:control plots prior to treatment. Here the control was the mean of “untreated” and “oven-killed” since their ground covers were similar. The BACI values were log<sub>10</sub>-transformed and statistically analysed using ANOVA. The back-transformed means of the log<sub>10</sub>-BACI values were subtracted from 100 to express the effects as % reductions in *R. acris* cover (Table 1).

## Results and discussion

There was no evidence for any effects from applying the *S. sclerotiorum* in October; weather records suggest that conditions were too cold and that heavy rainfall may have washed the granules from the leaves before infection could begin (data not presented).

With the November application there was a strong dose response at Site 2 and only a weak response at Site 1 (Table 1). At Site 2, the ground cover of *R. acris* reduced with increasing dose from 25 to 100 kg ha<sup>-1</sup> assessed 1 and 3 months after treatment ( $P < 0.01$ ) (Table 1, Figure 1). This reduction was smaller but remained significant ( $P < 0.05$ ) 6 months after treatment. There was no evidence for any effect of either wounding or granule size. By comparison, at Site 1 there was evidence for reductions in ground cover only at 100 kg ha<sup>-1</sup> at the medium granule size, and at 500 kg ha<sup>-1</sup>. This difference in response between the sites implies either that the *R. acris* at Site 1 is genetically more tolerant of *S. sclerotiorum* and/or that environmental conditions for disease were less favourable at this site.

These data reveal that reductions in the cover of *R. acris* of 50–60% are achievable using this granule formulation of *S. sclerotiorum* at 50 kg ha<sup>-1</sup>, a rate that is likely to be commercially viable. Such reductions in cover are similar to those commonly achieved with chemical herbicides (Bourdôt & Hurrell, 1990; Sanders et al., 1994).

Table 1. Percentage reduction in % ground cover of *R. acris* in dairy pasture 1, 3 and 6 months after treatment (MAT) with *S. sclerotiorum* in Nov. 2003, averaged over  $\pm$  wounding. Values are significantly different from zero (**bold type**) when greater than LSE (least significant reduction).

Treatment (kg ha <sup>-1</sup> , granule size)	Site 1 (MAT)			Site 2 (MAT)		
	1	3	6	1	3	6
25, small	36	33	39	-21	10	-
						147
25, medium	-29	-25	1	24	43	2
25, large	18	20	6	<b>58</b>	40	17
50, small	-20	23	-2	<b>66</b>	<b>61</b>	<b>56</b>
50, medium	7	21	11	45	44	10
50, large	-9	5	-21	<b>51</b>	<b>59</b>	11
100, small	4	-5	-32	<b>80</b>	<b>68</b>	38
100, medium	<b>52</b>	<b>40</b>	<b>56</b>	<b>78</b>	<b>79</b>	33
100, large	10	-8	-12	<b>66</b>	<b>65</b>	28
500, mixed	<b>55</b>	<b>52</b>	30	46	36	-34
LSE (5%)	38	40	43	48	52	47
Mean reduction	12	16	8	49	51	1

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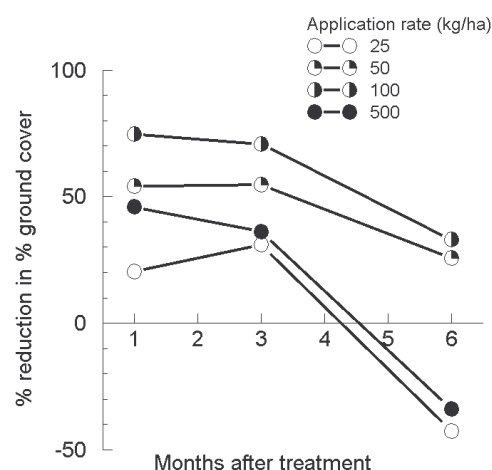


Figure 1. Response of *R. acris* to increasing application rate of granules containing *S. sclerotiorum* applied in November 2003 at Site 2. Means (symbols) are averages over granule sizes and  $\pm$  wounding.

# First survey for biocontrol agents against the native but invasive *Euphorbia esula* in Saône Valley (France)

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## Introduction

*Euphorbia esula* L. subsp. *esula*, leafy spurge, is a native of Eurasia and is commonly distributed in wild and cultivated areas. It has been reported as a problem in the Saône Valley since 1887. Farmers and practitioners noticed an unusual spread in the pastures from 1990s. Currently, it infests 3,500 ha of flooded grassland and could hamper the management of meadows, affecting plant and bird diversity. For 5 years, chemicals were tested to manage this weed but remained noneffective. The opportunity of using biological control on leafy spurge is now being studied as this Eurasian weed has been successfully controlled in North America by its natural enemies (Lajeunesse et al., 1999). This is the first study reporting leafy spurge invasion over large areas in Eurasia. In 2004 the main objective was to ascertain the presence of insects already known as good candidates for controlling the weed. This paper reports these results.

## Materials and methods

A 4-month survey over three highly infested wetlands was conducted in Arbigny, Pont de Vaux and Vésines (Ain). At each site, a 100-m<sup>2</sup> plot was selected and swept 50 times with a sweep net once a week from May to August 2004. Insects were captured and mounted for identification. Experimental plots were fenced to prevent grazing. Additional collecting was made with yellow sticky traps in prairies invaded by leafy spurge to estimate the peak population of *Aphthona* sp. (Coleoptera: Chrysomelidae).

## Results and discussion

The survey resulted in a collection of insects and a plant pathogen (Table 1). Two insects, *Aphthona violacea* and *A. venustula*, are of particular interest. These two *Aphthona* species were collected in the same locality with a peak population in mid July for *A. violacea* resulting both from yellow sticky traps and sweeping net data. In mesic sites, *A. violacea* adults are reported on *E. palustris* and *E. villosa* in France (Doguet, 1994), and on *E. lucida* in Hungary (Nowierski et al., 2002), from April to October. However, our sweep net collection was undertaken on *E. esula*, which has not been recorded before as a host. This aspect will be studied in depth in 2005. *Aphthona venustula* has a wider range of hosts in the *Euphorbia* genus, including *E. esula* (Doguet, 1994) with adults occurring from April to July. It is considered a candidate for biocontrol in the U.S. (Gassmann, 1996) but has not been yet introduced. Damage is induced by external root-feeding larvae in the spring and by leaf-feeding adults in the summer. Other natural enemies collected in the Saône Valley that may have a substantial impact on *E. esula* at the larval stage include *Oberea erythrocephala* (root feeder), *Hyles euphorbiae* (leaf feeder), and *Chamaesphecia* sp. (root feeder), all specific to *Euphorbia* species (Rees et al., 1996). As *E. esula* has a vegetative reproduction, larval root feeders may have a major role to play in reducing populations in the Saône Valley. Despite the failure of these introduced insects to achieve any control in North America (Gassmann & Schroeder, 1995), we think they are valuable candidates in France. Measured efficiency evaluated in an introduced range may not be valid in the native range, as *O. erythrocephala*, *Chamaesphecia* sp. (possibly *C. tenthrediniformis*) and *H. euphorbiae* are well adapted to their climate and host plants. We collected large numbers of adults of *O. erythrocephala* in



2004, and intend to evaluate larval impact in 2005. Collecting *Phyllotreta* sp. and *Longitarsus* sp. on *E. esula* was accidental. In addition, an anamorphic orange rust, identified as *Aecidium euphorbiae*, that induced severe damage to *E. esula*, was collected throughout the summer period. The life cycle is not clearly understood, but heteromacrocyclic development with Fabaceae as an alternate host is proposed (M. Abbasi, pers. comm.). This rust is found throughout Eurasia on *E. esula* and is being examined as a potential biocontrol agent within the U.S. (T.L. Widmer, pers. comm.). A future study will examine biological control through the use of insects and plant pathogens as a possible management strategy. Particular attention will be dedicated to the *Aphthona* spp. having oligophagous activity on the *E. esula* complex, and to root-feeding insects. Insect colonies from selected species will be maintained, and damage activity on *E. esula* evaluated. In addition, we intend to compare the genetic status of the *E. esula* strain in the Saône Valley to Eurasian and North American strains, in order to state whether or not the recently noticed invasiveness is due to an invasion induced by a non-native strain recently introduced or by cultural practices and climatic conditions locally.

Table 1. Natural enemies collected from *Euphorbia esula* in the Saône valley in 2004.

Order	Family	Latin name	Host plants*	Presence	Sites	Date of captures
Col.	Chrysomelidae	<i>Aphthona violacea</i>	<i>Euphorbia</i> sp.	++	a,b	May–Aug.
		<i>A. venustula</i>	<i>Euphorbia</i> sp.	+	b	July
		<i>Longitarsus brunneus</i>	<i>Thalictrum</i> sp.	n/a	a	July
		<i>L. luridus</i>	<i>Ranunculus</i> sp.	n/a	a,c	July–Aug.
		<i>L. reichei</i>	<i>Plantago</i> sp.	n/a	b,c	n/a
		<i>L. rubiginosus</i>	<i>Calystegia</i> sp.	n/a	a	n/a
		<i>Phyllotreta atra</i>	Brassicaceae	n/a	a	n/a
		<i>P. aerea</i>	Brassicaceae	n/a	c	n/a
		<i>P. vittula</i>	Polyphagous	n/a	c	n/a
		<i>P. cruciferae</i>	Polyphagous	n/a	a	n/a
			Cerambycidae	<i>Oberea erythrocephala</i>	<i>Euphorbia</i> sp.	+++
Lep.	Sphingidae	<i>Hyles euphorbiae</i>	<i>Euphorbia</i> sp.	++	a,b,c	June–Sept.
	Sesiidae	<i>Chamaesphecia</i> sp.	<i>Euphorbia</i> sp.	+	c	June
Ured.	—	<i>Aecidium euphorbiae</i>	<i>Euphorbia</i> sp.	++	c	May–June

\* Data collected from Doguet (1994) and Rees et al. (1996). Order: Col., Coleoptera; Lep., Lepidoptera; Ured., Uredinales (Fungi). Presence: +, rare; ++, common; +++, abundant. Collection sites: a, Vésines; b, Pt de Vaux; c, Arbigny. n/a, not available.

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# Controlling *Azolla filiculoides* using *Artemisia dracunculus* and *A. vulgaris* leaf extracts

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## Introduction

*Azolla filiculoides* Lamarck (Azollaceae) is a floating aquatic fern native to South America. It is a weed in many African countries, including Ghana and South Africa (Hill, 1998) and is causing increasing problems in waterways in the UK. Mechanical methods of control have not proved effective and chemical control, although effective, can have negative environmental effects. Biological control using the weevil *Stenopelmus rufinusus* has also been attempted (Hill, 1998; Hill & Cilliers, 1999).

Extracts from a variety of *Artemisia* species including *A. annua* (Duke et al., 1987) and *A. vulgaris* (Inderjit & Foy, 1999) have shown potential for weed control, and artemisinin, extracted from *A. annua*, has been researched extensively (Duke et al., 1987; Chen & Leather, 1990; Lydon et al., 1997). Furthermore, extracts from *A. annua* have inhibited growth of the water weed *Lemna minor* (Stiles et al., 1994). In this study we assess the effect of leaf extracts from *Artemisia dracunculus* (tarragon), available commercially in the UK, and *A. vulgaris* (mugwort), a native UK plant, on *Azolla* growth.

## Materials and methods

Freeze-dried *Artemisia dracunculus* (from a commercial supplier) and *A. vulgaris* (collected locally) were extracted sequentially with methyl chloride, ethanol, and distilled water following Lydon et al. (1997). The extracts were dried under vacuum and resuspended in methanol (methyl chloride proved toxic to *Azolla*), ethanol and water respectively to produce a concentration equivalent to 1 g dw leaf material ml<sup>-1</sup>. In subsequent experiments the extracts were all resuspended in water.

The *Azolla* used in these experiments were from a stock population at the University of Reading, originating from a local garden centre. Preliminary experiments (Prelim 1, 2, Table 1) were carried out in a CT cabinet (21°C, 18 hr light), with fronds grown in glass dishes with 18 ml nutrient solution and 0.2 ml extract solution for five days (against controls with the same solvent), after which the nutrient solution and extract was replaced and growth measured after a further five days. Subsequent experiments were carried out in a glasshouse (about 22 °C with 18 hr supplementary lighting). To investigate the effect of the abstract on an established *Azolla* population, 3 g fw *Azolla* was placed in equal volume of water and 69 g Levingtons' Multipurpose compost in plastic containers (12 cm diameter x 12 cm depth) on 6 October 2004 and it was allowed to cover the surface of the water. On 20 October each container was sprayed with 1.5 ml of extract (Table 1) resuspended in water. Two further sprayings took place at one-week intervals and the containers harvested two weeks after the third spraying and fresh and dry weight measurements taken. The effect of the extracts on a growing *Azolla* population was measured by placing 2 g fresh weight *Azolla* into the same containers with equivalent water and 46 g Levingtons' multipurpose compost. Plants were sprayed with extracts resuspended in water (Table 1) at weekly intervals (0.6, 0.8, 1 ml of extracts respectively) starting three days later and harvested two weeks after the third spraying. In all experiments there were 10 replicates of treatments.

## Results and discussion

The preliminary experiments indicated that water, ethanol and methyl chloride extracts of *Artemisia vulgaris* and *A. dracunculus* were effective in reducing growth of *Azolla*, when



suspended in their solvents (Prelim 1, Table 1), with ethanol and methyl chloride extracts of *A. dracuncululus* killing all *Azolla* within 10 days. Water resuspensions were less effective, but the ethanol extract of *A. dracuncululus* produced a 63% reduction in *Azolla* biomass in 10 days (Prelim 2, Table 1). There was no significant difference in the starting weights of *Azolla* in the greenhouse experiments (data not shown). The ethanol extract of *A. vulgaris* produced a 78% reduction in fresh weight of the established population and 64% reduction in the growing *Azolla* population. In the established population experiment, the *A. dracuncululus* ethanol and methyl chloride extract produced a 41% and 13% reduction in weight of *Azolla* compared to the control. In the growing population experiment some of the extracts produced an increase in *Azolla* growth compared to the control.

Table 1. Effect of *Artemisia* extracts on *Azolla* growth

Extract	Prelim 1	Prelim 2	Established exp.		Growth exp.	
	% fresh wt.	% fresh wt.	Fresh wt, g	Dry wt, g	Fresh wt, g	Dry wt, g
Control	—	—	14.7±0.5	0.85±0.03	6.4±0.3	0.36±0.01
A.v. water	– 30%	—	—	—	—	—
A.v. eth	– 87%	ns	3.2±0.3*	0.23±0.02*	2.3±0.3*	0.11±0.01*
A.v. mc	ns	ns	13.6±0.5	0.83±0.03	8.0±0.5*	0.41±0.01*
A.d. water	– 13%	—	—	—	—	—
A.d. eth	– 100%	– 63%	8.7±0.6*	0.52±0.03*	7.0±0.4	0.38±0.01*
A.d. mc	– 100%	ns	12.8±0.5*	0.78±0.03	5.8±0.2	0.42±0.01*

A.v.: *Artemisia vulgaris*; A.d.: *Artemisia dracuncululus*; water, eth, mc: water, ethanol, methyl chloride extracts respectively. —, treatment not carried out. For CT experiments Prelim 1 (extracts resuspended in solvents) and Prelim 2 (extracts resuspended in water), percentage reduction in fresh weight after 10 days compared to controls is given when significantly ( $P < 0.05$ ) different to controls, ns,  $P > 0.05$ . For the established and growth experiment, mean ± SE given, \*,  $P < 0.05$  compared to control, otherwise  $P > 0.05$ .

Ethanol and methyl chloride extracts resuspended in ethanol and methanol of both *Artemisia* species was effective against *Azolla*. However, it would be unrealistic to apply these extracts in this formulation in the field. Aqueous resuspensions were less effective, but the ethanol extract of *A. vulgaris* was consistently effective against *Azolla* in the greenhouse experiments. There is no evidence from the literature that *A. dracuncululus* and *A. vulgaris* contain artemisinin, [this, and similar compounds would be extracted in the methyl chloride fraction (Lydon et al., 1997)] and thus other compounds are probably responsible for the effect on *Azolla*. Likewise, Lydon et al. (1997) found that the aqueous extract of *A. annua*, which did not contain artemisinin, reduced seedling growth and survival, as can leachates from *A. vulgaris* (Inderjit & Foy, 1999). *Artemisia vulgaris* is widely available as a weed and ruderal plant in the UK and we recommend that further studies be carried out on the effectiveness of this potential control method of *Azolla*.

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# Insect choice in weed biocontrol: the case of *Gastrophysa viridula* on *Rumex obtusifolius*

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## Introduction

The choices that confront insects used for weed biocontrol in the field have received little attention. Even though a monophagous or narrowly oligophagous insect species may be chosen on the basis of laboratory or small-scale pot tests, the choice that this insect makes in the field may be influenced by many untested factors, for example the age of the plant, previous insect damage or fungal infection to the plant. Some of these factors could be particularly important when more than one biocontrol agent is to be used, or the biocontrol agent is attempting to control an indigenous weed which already has herbivores or pathogens attacking it (Hatcher & Paul, 2001). We investigated the effects of leaf size, prior infection by the rust fungus *Uromyces rumicis* (Schum.) Wint. and prior insect feeding damage on the choice made by *Gastrophysa viridula* Degeer (Coleoptera: Chrysomelidae) feeding on *Rumex obtusifolius* L. (Polygonaceae) in the laboratory and field. Both this beetle and fungus have been investigated as potential control agents for *R. obtusifolius* (e.g. Keary & Hatcher, 2004; Zaller, 2004).

## Materials and methods

Laboratory experiments were carried out in glasshouses and CE cabinets, at 20/14°C day/night and 16 h photoperiod. *Rumex obtusifolius* plants were grown in 9 cm diameter pots of loam/sand/peat mixture and grown to leaf two stage, when they were divided into treatments. One no-choice experiment (one plant per pot) and three choice experiments (two plants per pot) were carried out (see results for details). Infection of plants with uredospores of *Uromyces rumicis* followed Hatcher et al. (1994a), although a standardised spore concentration of 22.7 mg L<sup>-1</sup> was used. *Gastrophysa viridula* were taken from a stock population maintained at the University of Reading. For herbivory at leaf two stage, a male and gravid female adult were placed on each plant and bagged and allowed to feed for 24 h. For herbivory at the four leaf stage, the beetles were left on the plants for 48 h. Subsequently, leaf area was measured using digital leaf area measurement software, on images recorded using a digital camera.

The field experiments used plants reared as above and were carried out in the late summer. In the inoculation experiment, plants were inoculated at the leaf three stage, and transferred to a field plot at the leaf five stage. The field plot consisted of two square grids of 64 pots dug into permanent pasture, with pot spacing of 50 cm [following the design of Hatcher et al. (1994b)]. Untreated plants were placed in the first grid, and 32 treated + 32 untreated plants were placed in the second grid in a checkerboard arrangement. Two days later one gravid female *G. viridula* was introduced into the intersection of each four plants and allowed to feed and lay eggs for three days, before leaf area eaten and eggs laid were recorded. This experiment was repeated using plants grazed by *G. viridula* at the early leaf four stage instead of rust infection, and placed into the grids at the leaf six stage.

## Results and discussion

**Laboratory experiments.** When *R. obtusifolius* was grown alone in pots there was a significant difference in the leaf area consumed on the different treatments (control, 6.5 cm<sup>2</sup> ± 0.8; grazed at leaf two stage, 6.0 ± 0.5 cm<sup>2</sup>; infected at leaf two stage, 9.3 ± 1.0 cm<sup>2</sup>, ANOVA <sub>2</sub>, <sup>105</sup>  $P < 0.001$ ), with beetles consuming more on the infected plants. The amount of leaf area

available to the beetle had no effect on the amount of grazing observed. When grown in pairs (one plant infected at leaf two stage and one plant uninfected, or one plant grazed at leaf two stage and one plant ungrazed), the infected plants were grazed to the same extent as their uninfected partners (infected,  $4.5 \pm 0.8 \text{ cm}^2$ ; control,  $5.4 \pm 0.8 \text{ cm}^2$ ;  $P > 0.05$ ), but previously grazed plants were grazed less than the undamaged control plants (grazed,  $2.2 \pm 0.2 \text{ cm}^2$ ; control,  $7.1 \pm 0.6 \text{ cm}^2$ ;  $P < 0.001$ ). When one of the pair was infected the total amount of grazing across both plants was affected by which plant the beetle chose to graze more heavily (total area eaten when: inoculated plant grazed more,  $8.4 \pm 1.1 \text{ cm}^2$ ; control plant grazed more  $11.5 \pm 1.1 \text{ cm}^2$ ,  $P < 0.05$ ), but there was no such effect with previously grazed plants. Relative plant size had no effect on amount of grazing when the beetles were given the choice between two untreated plants of differing size.

**Field experiments.** The amount of grazing on previously infected plants was not significantly different to that on untreated plants in the same plot, but there was less grazing per plant in the mixed than in uninfected plots (area grazed per plant: control plot,  $9.8 \pm 0.9 \text{ cm}^2$ ; mixed plot  $5.1 \pm 1.5 \text{ cm}^2$ ;  $P < 0.001$ ). Within the mixed plot more eggs were laid on infected plants (760) than the uninfected (572,  $\chi^2 P < 0.001$ ), and more eggs were laid in total on the uninfected plot (1615) than the mixed plot (1332,  $\chi^2 P < 0.001$ ).

There was no difference in the quantity of material consumed from the mixed grazed/ungrazed plot and the control plot, but more eggs were laid on the mixed plot (1662) than the control plot (1189,  $\chi^2 P < 0.001$ ). In the mixed plot more eggs were laid on the previously grazed plants (895) than the controls (767,  $\chi^2 P < 0.001$ ).

The contradictions between the laboratory and field experiments (particularly for the beetles' preference for grazed or ungrazed plants) suggests that the beetles may respond to different cues in the laboratory and field. In the laboratory, beetles had relatively simple choices to make between small closely-spaced plants. However, in the field, the beetles are not only able to choose which leaf to feed on, but can move between many plants in search of better food or egg-laying sites. Furthermore, in the field, the plants are much further apart than in the lab, making host location more difficult and increasing the energy cost of time to feeding taken in locating a new host.

The implications of these experiments for *R. obtusifolius* control are mixed. Neither prior insect grazing nor rust infection has any negative effect on subsequent beetle grazing, in comparison with the controls, but a mixed infected/uninfected population of plants resulted in lower levels of damage than was seen on an undamaged monoculture. In this mixed population, the beetles' ability to select the best plants for egg laying is also diminished, possibly reducing the likelihood of effective continued control. These results could have implications for combined insect–fungus weed biocontrol strategies and demonstrates that simple laboratory choice tests are poor at mimicking field situations.

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# Post-dispersal seed predation in rotational fallows

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## Introduction

Post-dispersal seed predation is potentially an important source of seed loss for arable weeds (Westerman et al. 2003a). The most important predator groups of weed seeds are invertebrates (e.g. ground beetles) and vertebrates (e.g. rodents) (Watson et al. 2003, Westerman et al. 2003b). The factors affecting the intensity of seed predation are poorly known. Here, the effect of vegetation structure on the seed predation by vertebrates and invertebrates were studied. The importance of vertebrate predation might be assumed to be more intensive in the dense vegetation.

## Materials and methods

Seed predation of *Chenopodium album* and *Stellaria media* was studied by monitoring removal of seeds from 'seed cards' which were made by gluing seeds with repositionable glue to sand paper (5 cm x 10 cm) (see Westerman et al. 2003b). The number of seeds per card varied between species (30 seeds of *C. album* and 50 seeds of *S. media*). To study the relative importance of predation of invertebrates and vertebrates, the following exclusions were conducted by placing metal wire mesh cages over seed cards: no exclusion (access of all predators), cage with 11 mm mesh size (access of invertebrate predators, exclusion of vertebrates), cage with 2 mm mesh size (exclusion of all predators).

The experiment was established in a fallow experiment, which had different combinations of the following treatments (four replicates of each): duration of the fallow (1 or 2 years), establishment method (undersown or not undersown), green or stubble, seed mixture (*Trifolium pratense* - *Festuca pratensis* or *Agrostis capillaris* - *Festuca ovina*). The size of each experimental plot was 0.3 hectares (44 m x 66 m). One seed card of each combination of exclusions (no exclusion, 2 mm cage or 11 mm cage) and weed species (*C. album* or *S. media*) was placed in each experimental plot. The distance between seed cards was five metres. The consumption of seeds was studied during five two-week exposure periods between 8<sup>th</sup> June and 17<sup>th</sup> August in southern Finland in 2004. The data were analysed by applying ANOVA. The percentage values were arcsine-transformed before the analyses.

## Results and discussion

The consumption rate of seeds was found to differ between exclusions ( $F_{2,889}=60.32$ ,  $P<0.001$ ) and fallow types ( $F_{7,889}=2.15$ ,  $P=0.0360$ ), but not between species ( $F_{1,889}=2.23$ ,  $P=0.1355$ ). An interaction between fallow type and exclusion ( $F_{14,889}=0.91$ ,  $P=0.5445$ ) or between fallow type and species ( $F_{7,889}=1.58$ ,  $P=0.1367$ ) was not detected. The comparison within fallow types (Fig. 1) revealed the predation intensity of seeds of *C. album* by vertebrates and invertebrates did not to differ in any kind of fallows. In contrast, the predation of seeds of *S. media* by vertebrates was higher than predation by invertebrates ( $P<0.05$ ) in 1- and 2-year *F. ovina* – *A. capillaris*, 1-year *T. pratense* – *F. pratensis*, as well as undersown 1-year *T. pratense* – *F. pratensis* –fallows.

The only clear temporal pattern detected in the predation activity during the growth season was a higher consumption rate of the seeds during the last exposure period. The same pattern was found in both species and all exclusions. The predation of seeds of *S. media* by vertebrates was higher than predation by invertebrates ( $P<0.001$ ) during all exposure periods except the first one (22<sup>nd</sup> June). The predation of *C. album* seeds by vertebrates exceeded the

predation by invertebrates ( $P < 0.05$ ) only during third (20<sup>th</sup> July) and in the last (17<sup>th</sup> August) exposure period.

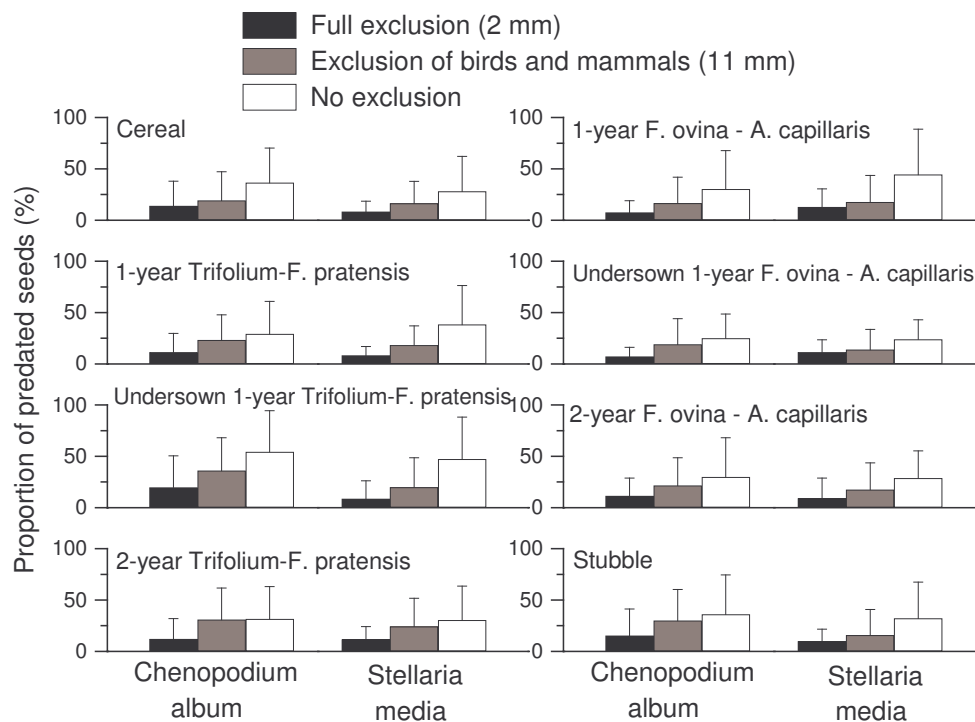


Figure 1. The predation of seeds of *Chenopodium album* and *Stellaria media* by different predator groups in different types of fallows.

The evidence on the impact of vegetation structure on the intensity of seed predation by different predator groups remained weak. The predation of *S. media* seeds by vertebrates was higher than by invertebrates in some fallow types. However, only in one of those fallow types (undersown 1-year-old *T. pratense* - *F. pratensis* fallow) can the vegetation be regarded as dense.

In the present study, the intensity of seed predation was lower and the temporal pattern was opposite compared with previous studies, where the seed predation has usually been most intensive in the early growth season (Watson et al. 2003, Westerman et al. 2003b). The increase in the intensity of predation by vertebrates towards the end of growth season in the present study was probably due to an increase in the population of rodents during the summer. The lower level of predation intensity may indicate that post-dispersal seed predation is less important in weed population dynamics in northern compared with temperate regions.

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# The impact of sown grass margin strips on weed occurrence in arable crop edges

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## Introduction

An appreciation that changes in farming practices are having deleterious effects on previously common flora and fauna of farmland has led to a number of initiatives that seek to combine profitable production with the maintenance of biodiversity. As a result of changes such as increased fertiliser use, silage grass cutting, more winter sowing of crops and more efficient weed and pest control, there have been significant reductions in the abundance and geographic range of common flora and fauna of farmland (Fuller et al., 1995; Wilson & King, 2003; Thorbek & Bilde, 2004). Agri-environment support schemes have been initiated across most European countries as a means of both financially supporting farmers and encouraging more environmentally sound land management (Kleijn & Sutherland, 2003). A number of prescriptions are based on the management of field margins (Marshall & Moonen, 2002), aimed at benefiting elements of both fauna and flora. Manipulation of non-crop habitat to benefit biodiversity is attractive, if impacts on adjacent commercial cropping are minimal. We hypothesised that sown grass margins at the edges of arable fields a) reduced the abundance and diversity of the weed flora in the adjacent crop edge, b) had no significant impact on the weed flora of the field centre and c) enhanced the occurrence of rarer arable weed species. These hypotheses were tested as part of a larger evaluation of the biodiversity impacts of sown grass margin in southern England (Marshall et al., submitted).

## Materials and methods

Twenty-one arable fields where 6-m wide grass margins had been sown at least 3 years previously under UK Countryside Stewardship Scheme contracts were selected and compared with 21 matched non-scheme fields. Paired fields were located close together in similar landscapes, with a similar field boundary structure and arable crop. The 21 pairs were grouped *a priori* into three landscapes based on field size: small (average = 4-ha fields) with much semi-natural habitat, intermediate (8–10 ha), and open (> 10 ha). Flora were assessed in 5-m<sup>2</sup> quadrats in the crop centre and in the pre-existing boundary (*sensu* Marshall & Moonen, 2002) usually beside hedges. Percentage ground cover of higher plant species was assessed by eye between June and July 2003 in ten 5-m<sup>2</sup> quadrats from the two locations and also from three quadrats each in the 6-m margin (where present) and the crop edge. Data were subjected to analysis with generalised linear models (GLM) using the Poisson distribution with a log. link function, and also with simple analyses of variance. Multivariate analyses of the species and land use data were made using the Canoco program.

## Results and discussion

A total of 275 different plant species were identified from the four sampled locations in the boundary, the 6-m strip, the crop edge and the crop centre across the 42 field sites. Species richness, total cover, cover of monocotyledons and dicotyledons in the 6-m strips were unaffected by landscape type, indicating that there was little bias in types of seed mixture used to establish them. Analysis of the species richness of the flora of field boundaries and crop centres, using the full dataset of ten quadrats per site, indicated significantly greater

biodiversity in the boundary of margins adjacent to sown 6-m margin strips (Table 1), but no difference between landscapes.

Table 1. Plant species richness (mean number of species per 50-m<sup>2</sup> sample) in field boundaries and arable crops with and without sown 6-m margin strips. (Superscript letters <sup>a,b,c</sup> indicate overall differences between means at P≤0.05)

	Field boundary	Crop centre
With 6-m margin	33.2 <sup>a</sup>	10.2 <sup>c</sup>
Control (no margin)	27.7 <sup>b</sup>	11.3 <sup>c</sup>

Weed diversity in the crop close to the field edge averaged 13.8 species (7.1 per 15 m<sup>2</sup> in the field centre) and did not differ between margin types (6-m margin vs. control) or landscapes. However, weed cover was statistically significantly lower adjacent to sown 6-m margins (28%), compared with field edges with arable crops close to the boundary (37%). Crop cover did not vary amongst treatments, but was lower than in the field centres (73% : 82%). A comparison of weed cover in crop edges and crop centres (Table 2) confirmed that field edges are weedier than centres and smaller fields tend to be weedier than larger arable fields. There appeared to be no effect of 6-m margin strips on the weed flora of large fields. In small fields with 6-m margins, weed cover was significantly lower, both at the crop edge and field centre, compared with paired fields without margin strips. This may reflect differences in the management and attitudes of farmers in smaller landscapes, while 6-m margins have little impact on weed flora of larger fields.

Table 2. Weed cover (%) in the crop edge or field centre of fields with and without sown 6-m wide grass margins in three landscapes (data averaged over three 5-m<sup>2</sup> quadrats per treatment).

Landscape	Small		Intermediate		Open	
	Edge	Centre	Edge	Centre	Edge	Centre
With 6-m margin	21.1	12.3	30.4	2.4	32.4	7.0
Control field	54.5	23.1	29.6	12.8	26.7	6.9

An evaluation of less common plant species indicated no Red List species in the inventories, though a nationally scarce species, *Euphorbia platyphyllos*, was recorded from one site with a 6-m margin. Using 23 species listed by Wilson & King (2003) and showing significant declines in the UK (Preston et al., 2002), there was no evidence overall that 6-m margins affected rare weed occurrence. The study indicates that 6-m margin strips have no adverse agronomic effects on weed cover; but, from a conservation viewpoint, allow greater perennial plant species diversity to be maintained in non-crop field boundaries.

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# Seasonal variation of seed predation by ground beetles (Coleoptera:Carabidae)

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## Introduction

Post-dispersal predation is an important mortality factor for herbaceous seeds. In temperate regions, the most important invertebrate predators are ground beetles (Carabidae, Coleoptera) (Brust & House, 1988; Honek et al., 2003; Harrison & Regnier, 2003). Carabid species prefer different seed species, but seasonal variation in their preferences is not known. We investigated this variation in *Harpalus affinis* (Schrank) and *H. distinguendus* (Duftschmid), closely related and similar beetle species that occur in arable fields throughout the season and feed on weed seeds dispersed at different periods (Honek & Jarosik, 2000; Honek & Kocian, 2003). We hypothesised that seasonal changes in carabid physiology (dormancy, breeding, teneral development) may be associated with variation in their feeding preferences.

## Materials and methods

Preference of each species were established on five dates. Adults were collected in the field using pitfall traps, brought to the laboratory and stored for 3-5 days at 5-7 °C, then sorted into groups of 10 by selecting individuals randomly from the population for use in the experiment. Their preferences for seed of 28 species of dicotyledoneous herbs of arable fields and surrounding areas were studied in Petri dishes (250 mm in diameter, 50 mm height), each containing a 2 cm layer of sieved soil dug from a depth of >0.5 m to prevent contamination from the soil seedbank. Seeds were exposed in tin trays of 25 mm diameter filled with white soft plasticine. Thirty seeds of each species were stuck on the plasticine surface of a tray. The trays were placed on the ground so that the plasticine surface was level with the soil and accessible to beetles. The trays were exposed in two concentric rings, the outer consisting of 19 trays, the inner of 9 trays. The Petri dishes were placed at 25-27 °C and natural photoperiod. The beetles were supplied with water on a piece of cotton and relative humidity inside the vials was 100 %. For each species and date the experiments were made in five replicates and run for five days. Consumed seeds were counted daily and trays where seed was exhausted were replaced.

## Results

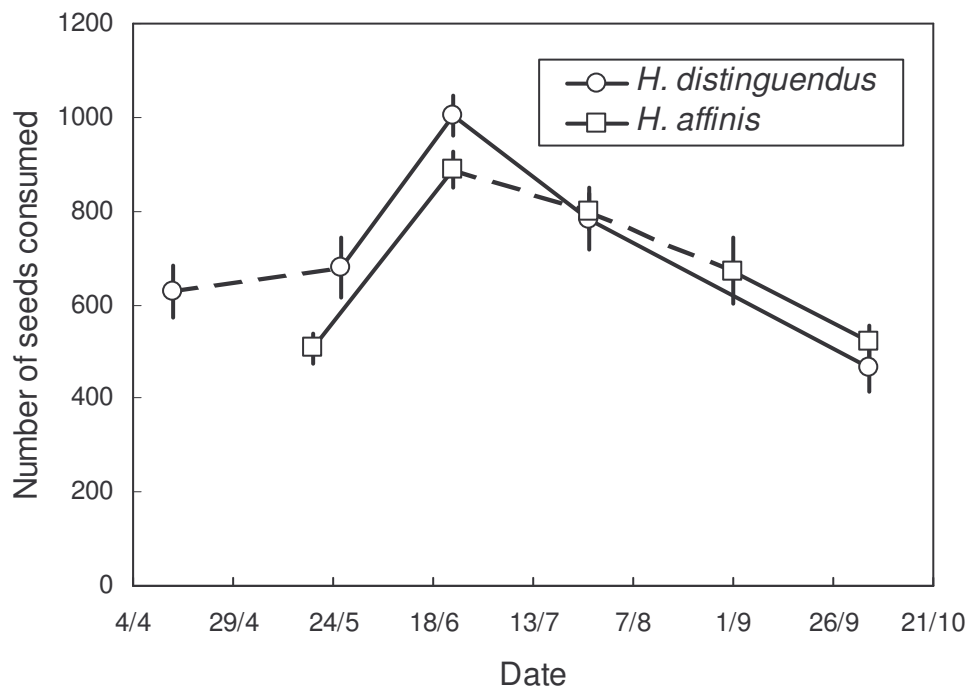
Total seed consumption significantly varied with the season and the course of variation was similar in both species (Fig. 1). In the spring, consumption was only 57 - 68 % of the maximum reached in late June. After this peak, consumption decreased slowly, so that in early October it was again 46 - 59 % of the maximum. Consumption on successive dates differed significantly. By contrast, the consumption of *H. affinis* and *H. distinguendus* on the same dates were similar.

The preferences of both *H. affinis* and *H. distinguendus* were essentially identical and varied little with the season. Seasonal variation in consumption of particular seed was greater in preferred species (*Cirsium arvense*, *Tripleurospermum inodorum*, *Taraxacum officinale* and *Viola arvensis*), while the poorly consumed seed was rejected at any time.

## Discussion

In both beetle species, consumption (quantity of seed consumed) varied with the course of the season, but the preferences (the rank of a particular seed species in the order of consumption) remained similar. The preference is thus determined by innate constraints while consumption varies with physiological state of the beetles (dormancy, reproduction) that changes with the course of the season. This variation is important in estimating ground beetle effect on seed populations.

Figure 1. Average number ( $\pm$ SE, n=5) of consumed seed (all seed species together) per replicate (10 individuals fed a mixture of seed for 5 days) plotted against the date of the experiment



## Acknowledgements

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# Biological control of *Solanum viarum* in the USA: current status and perspectives

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## Introduction

*Solanum viarum* (Solanaceae), known by the common name tropical soda apple, is an invasive prickly shrub in southeastern USA. It is native to Brazil, Argentina, Paraguay, and Uruguay. First detected in 1988 in South Florida (Coile, 1993; Mullahey & Colvin, 1993) it has already invaded more than 400,000 ha of grasslands, improved pastures and conservation areas in 11 states (Alabama, Arkansas, Florida, Georgia, Louisiana, Mississippi, North Carolina, Pennsylvania, South Carolina, Tennessee, and Texas) (Medal et al., 2003). The potential range of *S. viarum* in the USA may extend even further based on studies of the effects of temperature and photoperiod conducted by Patterson (1996). Estimates of annual production loss to Florida cattle ranchers due to *S. viarum* infestations in pastures were US \$ 11 million in 1993 (Mullahey, unpublished data). This figure does not include vegetable crop losses caused from pathogens transmitted by insect vectors from *S. viarum* infested areas. Currently, recommended control tactics for *S. viarum* in pastures are based on herbicide applications combined with mechanical (mowing) practices. These control tactics provide a temporary solution, and can cost as much as \$188 ha<sup>-1</sup> for dense infestations of *S. viarum*. A biological control project against *S. viarum* was initiated in 1997. After 3 years of intensive host specificity testing, the South American beetle *Gratiana boliviana* (Chrysomelidae) was approved and a field release permit was issued by USDA-APHIS-PPQ in May 2003, and its release in Florida began in summer 2003.

## Materials and methods

Field releases of the leaf beetle *Gratiana boliviana* for biocontrol of *Solanum viarum* have been made in 14 locations in 11 Florida counties (Alachua, Collier, De Soto, Hendry, Hernando, Lake, Martin, Okeechobee, Polk, St. Lucie, and Sumter) since the summer of 2003. A total of approximately 8,500 beetles (larvae and adults) have been released in Florida: the number of beetles released at each location varied from 100 to 1,000 depending on beetle availability. Twenty marked *S. viarum* plants, within 100 m of the initial release site, were examined every 4 to 8 weeks during the growing season. Evaluation of the feeding effects of the beetles on the *S. viarum* plants (% defoliation, plant height, plant diameter, number of fruits per plant), and changes in the weed target populations, beetles number, and native plant populations are currently underway at three of the release sites.

## Results and discussion

The estimated (visual) defoliation at the release site in Polk county during 2004 increased on average from 45% (5 May) to 86% (5 October), and was directly associated with the increase in number of adults and immature beetles observed on the *S. viarum* plants during the same period, except in August and October when the number of adult and immature beetles decreased. The number of beetles (adults and larvae) decreased slightly from 6 July to 19 August, this can be attributed to the beetles' dispersal to *S. viarum* plants as far as 700 m away from the initial release site. Dispersal of the beetles seemed to be associated with the availability of *S. viarum* plants. At the end of September 2003 (1 month after the release of approximately 1,000 beetles), at least 40 beetles were observed on *S. viarum* plants 50 m away from the initial release site. Most of the beetles remained at relatively short distances

from the release site, when they had a large number of plants with abundant foliage. As the beetle population started increasing in number, the defoliation of *S. viarum* also increased, and 50% and 60% of the 20 marked plants were completely defoliated at the middle of August and beginning of October 2004, respectively. The number of adult beetles recorded in October on marked *S. viarum* plants continued to decrease which can be attributed to the lack of foliage, or initiation of diapause as a response to changing environmental conditions. A large number of *S. viarum* plants in this release site have been replaced by other plant species, including bahiagrass, *Paspalum notatum*; *Rubus* sp.; dayflower, *Commelina diffusa*; Caesar weed, *Urena lobata*; air-potato, *Dioscorea bulbifera*; roadside flatsedge, *Cyperus sphaecelatus*; oak *Quercus* sp.; and other grasses and broadleaf plants. The beetles keep showing good dispersion ability. On 5 October 2004, larvae and adult beetles were found on *S. viarum* plants 800 m away from the initial release. Additional beetle releases will be made next spring to summer 2005 in different locations in Florida. Post-release monitoring of the beetles and *S. viarum* plants will continue during 2005 in at least three of the release sites. The follow-up studies will include observations on possible effects, if any, on non-target plant species growing in the proximity of the release area, and on the regeneration of native plant species and/or improved pastures that have been displaced by *S. viarum* plants. At the end of 2004, the beetle *Gratiana boliviana* has been established at all the release sites in Florida, and it is causing from 60 to 100% defoliation on *S. viarum* plants at the three release sites monitored.

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# Evolution of life cycle in invasive plants and implications for biocontrol

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## Introduction

Invasion ecology has received considerable attention during the past decades (e.g. Alpert et al., 2000). This is mainly a consequence of the increased awareness of the major threats posed by invasions to biodiversity, ecosystem integrity, agriculture and human health (Mack et al., 2000). The main factors thought to underlay invasion success are: (i) *ecological processes* such as human-altered environments (disturbance, fertilization regime) or the release from biotic constraints (specialized natural enemies, competitively superior neighbours, soil pathogens); and (ii) *evolutionary processes* such as post-invasion evolution of increased competitive ability due to altered selection in the new range (EICA hypothesis). Hybridization can also increase genetic variation, and lead to transgressive (extreme) phenotypes that may be more fit in the novel environment (cf. Müller-Schärer et al., 2004). The rate of adaptive evolution is determined by two components: selection and heritable variation. Therefore, to understand adaptive evolution in invasive plants, we must investigate how selection pressures on phenotypes differ between the native and the introduced range, and how the invasion process affects the amount of genetic variation expressed in a population. Here we explore potential evolutionary trajectories of plant life cycle in the new range, and present hypotheses about how these changes might influence the efficacy of biological control. The most prominent change experienced by introduced plants in terms of natural enemies is a shift in the composition toward an assemblage that is dominated by generalists, and this shift is expected to incur altered selection on traits in invasive plants (cf. Müller-Schärer et al., 2004).

## Evolution of life cycle in the new range and ecological consequences

Observations of several plant species (e.g. *Cynoglossum officinale*, *Senecio jacobaea*, *Oenothera* spp., *Digitalis purpurea*, *Verbascum thapsus*, *Centaurea stoebe* and *Tripleurospermum perforatum*) indicate a change from a prevalent monocarpic habit (plants die after flowering) in the native range of temperate Eurasia to a polycarpic habit (flowering several times) associated with the invasion process into North America (Müller-Schärer & Steinger, 2004, and references therein). We propose that these observations can be explained by the release from specialized natural enemies that can kill plants that overwinter after seed set, such as endophagous (mining and galling) root-feeding herbivores. Little is yet known on the inter-relationship between the life-cycle habit, ploidal level and breeding system, and their associations with the invasion success. Apomixis and autogamy tend to occur in populations at the edge of the species' ranges, and is found largely in populations whose other populations are self-incompatible. Furthermore, apomicts are also often polyploids. A possible explanation is that polyploids tend to be larger than diploids and selection may have favoured polyploids in harsher environments at the edge of the ranges of diploids, from which they have arisen, and where asexual reproduction and selfing is also favoured. Phenotypic consequences of polyploids as compared to their related diploids are manifold, ranging from increased levels of secondary plant compounds up to longer lifespan (e.g. annuals versus perennials), breakdown of self-incompatibility (or induced apomixes) and altered flower morphology that may promote assortative mating within ploidal level (Levin, 2002). Interestingly, chromosome doubling has been found to be often associated with plant invasions (Myers & Bazely, 2003).



### ***Centaurea maculosa* as a research model**

We are presently exploring the role of adaptive evolution in invasions with the European knapweed *C. maculosa* which has become invasive in North America, where it presently covers an area larger than Switzerland and causes reductions in hay production of up to 70%. In North America, *C. maculosa* is a predominantly short-lived polycarpic tetraploid, native to eastern Europe. By contrast, the most abundant and widely distributed *C. maculosa* in western Europe is a diploid biennial (Müller, 1989; Ochsmann, 2001). These diploid populations are also the main source of subsequent introductions of specialized root feeders into North America to control *C. maculosa* (Müller-Schärer & Schroeder, 1993). Our hypothesis is that polyploids have a polycarpic life cycle and may be autogamous, whereas diploids are monocarpic and allogamous. The potential for high seed production early in life, together with the polycarpic life cycle might have favoured the observed spread of the tetraploid plants in North America. A polycarpic habit may, thus, have been negatively selected by specialist herbivores in the native range, but positively selected in the introduced range where the specialist herbivores are absent. Following this argument, invasive populations should be highly susceptible to introduced biocontrol herbivores and their release could reverse the presumed evolutionary shift towards a polycarpic life cycle in the introduced range (Figure 1).

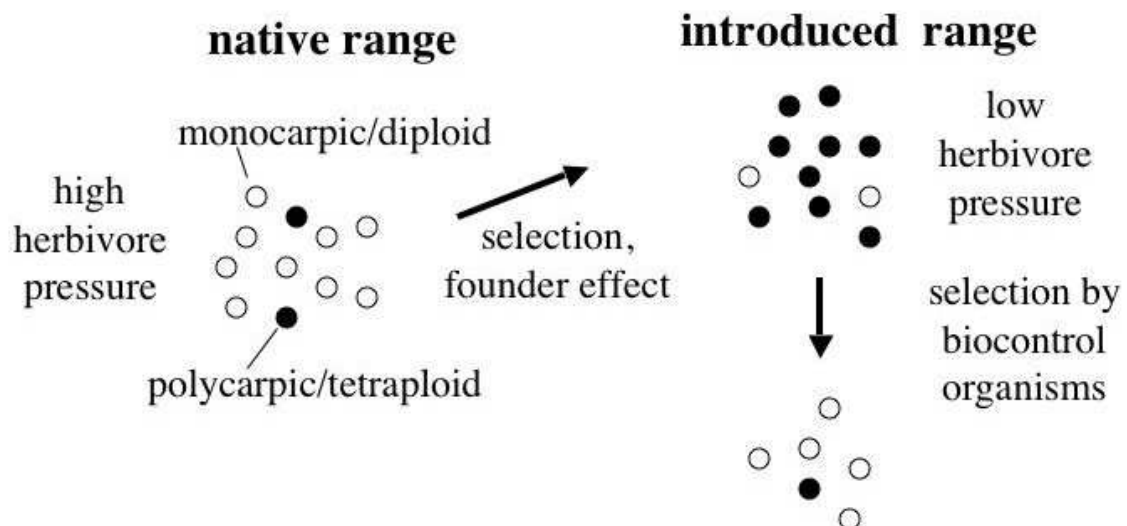


Figure 1. Hypothesized selection by the presence and absence of specialized insect herbivores in the native and introduced range, respectively, and by introduced biocontrol agents.

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# Some effects of imazethapyr on the population dynamics of migratory plant parasitic nematodes

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## Introduction

Some herbicides have been found to promote nematode multiplication, while others were toxic to nematodes and reduced crop damage. Diallylate, cycloate, triallate, metolachlor, benazolin had a stimulating effect on the hatch of larvae from the cysts of *Heterodera schachtii* and on the population of *Pratylenchus neglectus*, but these decreased when chloridazon, lenacil, ethofumesate and fenmedifam were applied (Kraus and Sikora, 1983; Benaszak, 1997; Kornobis, 2000). By contrast, Perry and Beane (1989) found that cycloate prevented the hatch of *Globodera rostochiensis* and *H. schachtii*. Glyphosate had a negligible effect on the free-living nematodes (Dmawska and Kozkowska, 1986), but according to Saly and Ragula (1984) it resulted in an increased number. Our objectives were to study the effect of imazethapyr on the densities of some migratory plant parasitic nematode species naturally occurring in the rhizosphere of the soyabean (*Glycine max* L.) and in the following crops, in the region of Kostinbrod, Bulgaria.

## Materials and methods

During 1998-2000 field trials were conducted on an alluvial soil naturally infected by ectoparasitic *Pratylenchus pratensis* and endoparasitic *Tylenchorhynchus dubius*, *Helicotylenchus digonicus* and *Paratylenchus* spp. In 1998, imazethapyr (Pivot 100 EC) was applied at the 2-3 trifoliolate stage of soyabean. The trial included 3 treatments in 4 replicates with a plot size of 10 m<sup>2</sup>: 1, Untreated control; 2, imazethapyr 0.1 kg a.i. ha<sup>-1</sup>; 3, imazethapyr 0.2 kg a.i. ha<sup>-1</sup>. After soyabean, the plots were left for soil sampling, until sown with oats (*Avena sativa* L.) in 1999 and with maize (*Zea mais* L.) in 2000. For determination of the nematode density, soil samples were collected at 30, 60, 90, 390, 420, 450, 750, 780 and 810 days after treatment (DAT). Ten cores for each sample were taken from 30 cm depth diagonally over the plots. The nematodes were extracted from 200 cm<sup>3</sup> soil samples in 4 replicates by a centrifugal flotation method (Jenkins, 1964) and counted under a microscope. The determination was made by the method of Paramonov (1964) and the data were statistically analyzed using ANOVA.

## Results and discussion

Our data showed significant decrease in the population dynamics of endoparasitic species *P. pratensis* in the plots treated with imazethapyr (Table 1). The average number of larvae in the plots 30, 60 and 90 DAT with imazethapyr 0.1 and 0.2 kg a.i. ha<sup>-1</sup> was 16-61 % and 23-66 % lower than the control. The differences between the treatments were statistically significant. The population densities of ectoparasitic species laying eggs outside the plant tissues showed some response, but there were no significant differences between the treatments and the control. The observed treatment effect on the population dynamics of the endoparasitic *P. pratensis* persisted significantly for one year only. During the next two years, when oat and maize were sown, *P. pratensis* density returned to the level of the untreated control (Table 2). The densities in the former control plots and those treated with both doses of imazethapyr were not significantly different. Similar results were obtained in a field experiment with bean (*Phaseolus vulgaris* L.) treated pre-emergence with imazethapyr at rates of 0.08 and 0.16 kg a.i. ha<sup>-1</sup> (Trifonova and Peneva, 2003). The results showed that imazethapyr affected mainly the endoparasitic *P. pratensis* laying eggs inside the plant roots, confirming data from Weischer & Muller (1985). As an imidazolinone herbicide, imazethapyr reduces the levels of

isoleucine, leucine and valine through the inhibition of acetohydroxyacid synthase (AHAS). This causes a disruption in protein synthesis, which leads to interference in DNA synthesis and cell growth of the susceptible weeds (Stidham, Singh, 1991). Imazethapyr has an inhibiting effect on the root length and the stem weight of some soyabean cultivars (Taregian et al., 2001). In this way, affecting soyabean roots, imazethapyr makes these hosts less suitable for *P. pratensis*. Weisskopf et al. (1988) concluded that the herbicides were able to affect nematode population dynamics for one year, but these returned to normal in subsequent years, in a similar fashion to our results.

Table 1. *P. pratensis* and ectoparasitic plant nematodes population dynamics in 1998

kg a.i. ha <sup>-1</sup> imazethapyr	<i>P. pratensis</i>			Ectoparasitic nematodes		
	30	60	90	30	60	90
	Mean ( SE )	Mean ( SE )	Mean ( SE )	Mean ( SE )	Mean ( SE )	Mean ( SE )
0	50.25 (0.85)	46.50 (6.29)	59.75 (0.85)	21.25 ( 1.38 )	20.75 ( 1.25 )	22.50 ( 1.55 )
0.1	42.25 (1.25)	33.50 (1.71)	23.25 (0.48)	21.50 ( 0.96 )	22.50 ( 1.04 )	20.00 ( 1.22 )
0.2	38.75 (1.49)	30.00 (0.82)	20.25 (0.85)	21.00 ( 1.29 )	22.25 ( 0.85 )	20.25 ( 0.85 )
p-value	<0.001*	0.031*	<0.001*	0.960	0.480	0.339

Table 2. *P. pratensis* population dynamics in 1999 and 2000

kg a.i. ha <sup>-1</sup> imazethapyr	DAT (1999)			DAT (2000)		
	390	420	450	750	780	810
	Mean ( SE )	Mean ( SE )	Mean ( SE )	Mean ( SE )	Mean ( SE )	Mean ( SE )
0	65.00 ( 4.43 )	59.00 ( 4.43 )	64.00 ( 3.16 )	69.00 ( 2.65 )	71.00 ( 1.29 )	63.00 ( 2.08 )
0.1	58.75 ( 2.69 )	59.25 ( 1.34 )	66.25 ( 5.86 )	72.75 ( 4.27 )	76.25 ( 3.12 )	75.75 ( 1.31 )
0.2	69.25 ( 4.78 )	51.00 ( 3.87 )	55.50 ( 6.40 )	63.50 ( 2.87 )	72.25 ( 4.29 )	64.25 ( 2.53 )
p-value	0.240	0.298	0.365	0.199	0.497	0.003*

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# Spatial patterns of plant species richness in relation to distance from crop field margins in the Rolling Pampa (Argentina)

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## Introduction

Crop field margins contribute to maintain biodiversity in agroecosystems. Typically, however, vegetation in fencerows is intensively managed to control potential weed invasion and agricultural pests. Both differences in management practices and biotic environments between the cropped area of fields and its margins could produce steep gradients in plant species richness towards the centre of the field. Furthermore, existing differences among field crops in their agricultural management, ecophysiology and competitive ability may determine different patterns in the species richness of their corresponding weed communities. Thus, alternate crops established within the same growing season may represent different ‘ecological filters’ for the weed flora occurring in fencerows and local seed banks. In this study, we tested the hypothesis that patterns of species richness decline with distance from field margins and will differ between narrow- and broad-leaved crops grown during a given field season (spring or summer).

## Materials and methods

Vegetation surveys were carried out in wheat ( $n = 26$ ) and pea ( $n = 13$ ) fields during spring 2003, and in maize ( $n = 13$ ) and soybean ( $n = 12$ ) fields during summer 2004 in the Rolling Pampa, northern Buenos Aires province, Argentina. In each field, all vascular plant species were recorded at several sampling stations established along a transect running from the wire-fencerow (0 m) towards the field interior. Sampling stations were represented by a series of 25 m-long transects parallel to the fencerow, located at increasing distances according to a power series of 2, from 1 to 128 m away from the field margin. In addition, a complete phytosociological survey was performed on a plot of approximately 0.25 ha located at the centre of the field. Species richness of the fencerow and the field interior were separately compared between the two spring and the two summer crops using Student *t* tests for unpaired samples. Means and their respective standard errors are presented.

## Results and discussion

In all studied crops plant richness abruptly declined over the first 8 m from the fencerow, but stabilised at greater distances (Fig. 1). Richness patterns differed more markedly between spring crops (pea and wheat) than between summer crops (maize and soybean). Although fencerow richness did not differ between pea ( $23.7 \pm 2.05$ ) and wheat ( $21.5 \pm 8.05$ ) crop fields, weed richness at 128 m from the fencerow was significantly greater in pea ( $10.5 \pm 1.23$ ) than in wheat ( $1.8 \pm 0.35$ ) fields ( $P < 0.001$ ). Fencerow richness did not differ ( $P = 0.336$ ) between maize ( $18.6 \pm 1.11$ ) and soybean ( $16.8 \pm 1.57$ ) fields, whereas no differences were observed at 128 m from the fencerow (soybean:  $5.2 \pm 0.89$ ; maize:  $4.6 \pm 0.58$ ,  $P = 0.612$ ). Species richness of the inner field, which represents the average crop environment, was greater in pea ( $20.4 \pm 1.57$ ) than in wheat ( $4.5 \pm 0.62$ ) fields ( $P < 0.001$ ), whereas no differences ( $P = 0.588$ ) were found in the inner field richness between soybean ( $8.5 \pm 0.98$ ) and maize ( $9.2 \pm 0.90$ ) crops. The differences

observed in weed richness within inner fields agree with previous observations for the study region (Suárez et al. 2001, Poggio et al. 2004).

For all four crops, the occurrence of species present in the fencerow was reduced as distance increased towards the inner field, while there was no pattern for species absent in the fencerow. The greatest increase in richness for species occurring exclusively in the inner field was found in pea crops. On average, seven weed species were exclusively present in the inner field (at 128 m from fencerows) for pea crops, while only one new weed was found inside wheat crops. Thus, the observed distance increase in weed richness of pea crops mainly resulted from the addition of species exclusive to the inner field environment. This pattern would contribute to maintain the richness of the local weed flora invading cool season crops.

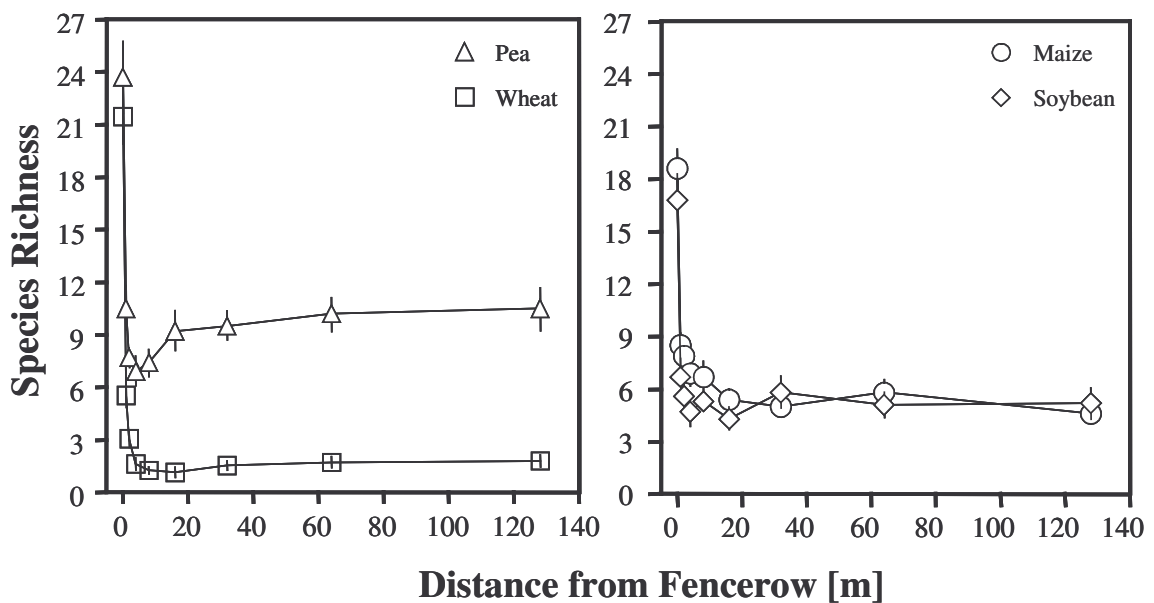


Figure 1. Species richness as a function of distance from the field margin in spring-sown pea and wheat crops (a), and in summer-sown maize and soybean crops (b).

Overall, these differences in plant richness with distance between crops support our initial hypothesis and are consistent with the idea that not only crop attributes, but also associated management of field margins, may strongly affect patterns of weed diversity in agroecosystems. So far, research on the distributional patterns of weed species richness across arable field margins has been rather sparse (Marshall 1989; Wilson & Aebischer 1995), with most work restricted to weed communities of winter cereals or to a particular botanical group of weeds (e.g. dicotyledons, Wilson & Aebischer 1995). Our results may be more widely applicable, since all vascular plants present in the most important field crops for the study region were included in the analyses. The reported spatial variation in plant richness patterns suggests source-sink relationships (Pulliam 1988) between the flora of field margins and that of the interior of arable fields, which are affected by the different crops grown.

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# Plant species richness assessment in Pampean agro-ecosystems

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## Introduction

The importance of species richness in agro-ecosystems has promoted much controversy. Nonetheless, it is widely accepted, on one hand, that richer plant communities enhance the availability of refuges for different life forms, and, on the other hand, that low diversity increases susceptibility to invasion, fungal disease spread and insect community alteration. Land use intensification may threaten the species richness of arable fields and their surroundings, for instance, by growing few crop species with narrow genetic backgrounds and by applying standardised strategies for weed control. Therefore, the impact of weed management practices on species richness of arable plant communities deserve to be investigated, not only to understand the dynamics of weed communities, but also to detect and even preserve endangered weed species contributing to agro-ecosystem functioning (Grime 2002). However, differences among methods to assess species richness at different spatial scales have received some criticism (Hillebrand & Blenckner, 2001), because of the difficulties in data comparison and interpretation. Therefore, this work has the objectives of studying weed species richness in different cropping systems, and, based on this, the discussion of the best methodologies for studies of plant diversity in agro-ecosystems.

## Materials and methods

Surveys were done during post-flowering of pea, wheat, maize and soybean crops located in the central Rolling Pampa, Argentina (34° to 36° S lat. and 58° to 62° W long). The whole surface of fields ranging from 50 to 80 ha was considered the sampling area. Surveys were restricted to field areas which had homogeneous crop cover belonging to the same soil type, and field margins were avoided. Surveys were performed by two or more trained persons who walked across each field during at least 30 min. (approx. 0.5 ha), recording all species observed until no more new species were found. Information concerning crop management and performance (i.e. tillage system, fertilization, weed control and grain yield) was requested from the farmers. Data were summarised by calculating alpha and gamma diversity (Whittaker, 1975). Alpha or local diversity is the number of species in a survey, so called species richness. Gamma or regional diversity is the total number of species occurring in a system. Determining factors were derived from canonical correspondence analysis.

## Results and discussion

Weed species richness differed among crops at both field and community levels (Table 1); differences could be explained by different agronomic variables (Ghersa & León 1999), even though all surveyed fields belonged to the same soil type. Species richness differences between pea and wheat were mainly determined by crop dominance, which include the competitive effect of crop plants and herbicide effects and, to a lower extent, by fertilization and previous crop (Poggio *et al.* 2004). The maize weed community was more diverse when crop yield, a good indicator of soil quality, was greater at both field and community scales (Table 1). Weed species richness of soybean crops varied differently at field and community scales with different levels of crop yield and tillage system. Soybean high yield fields were more diverse than low yield fields, irrespective of tillage system. The opposite patterns were observed at the community scale, as high-yield fields had low and high weed species in conventional and reduced tillage systems, respectively (Table 1). It can be partially concluded that, at field scale, weed community diversity is controlled by both the



crop environment and the physical and chemical properties of soil. Whereas, the observed differences among crops illustrate the extent to which heterogeneity in field crop occupancy may contribute to plant diversity at the landscape scale (Ghersa & León, 1999).

Table 1. Species richness of wheat, pea, maize and soybean crops at both field (average alpha diversity across fields), and community (gamma diversity) scales. Probable determining factors of arable plant diversity are included, according to correlations obtained by canonical correspondence analysis.

	Species Richness		Determining factors
	Field	Community	
Wheat	12.7±0.82	64	Crop dominance (crop yield + herbicide effects), Fertilization (mainly nitrogenous or phosphorous) Previous crop, (Poggio <i>et al.</i> 2004)
Pea	19.8±0.76	91	
Maize			
High yield	22.0±0.85	67	Crop yield (Suárez <i>et al.</i> 2001)
Low yield	15.7±0.70	49	
Soya bean			
Conventional tillage			Tillage system, Sowing date (optimum vs. late), Crop yield (de la Fuente <i>et al.</i> 1999)
High yield	13.8±0.79	45	
Low yield	10.5±0.82	51	
Reduced tillage			
High yield	16.1±0.73	44	
Low yield	14.3±1.50	33	

The proposed methodology successfully overcomes some of the weaknesses of other species diversity studies. Firstly, sampling size was large enough to comprise most of the species able to occur in arable fields, which facilitated the assessment of regional species richness. In comparison, some studies have measured weed diversity using small samples (e.g. 0.25 by 0.25 m quadrats, Doucet *et al.* 1999), which may produce misleading conclusions, or require too many samples to reach acceptable results. Secondly, adequate sampling size also reflects the spatial scale in which ecological processes take place, since the species richness of a local assemblage is determined by ecological filters which operate at different spatial scales (e.g. niche differentiation, habitat heterogeneity, mass effects, *sensu* Shmida & Wilson, 1985). In this sense, we think that the election of small sample sizes is driven by the assumption that species diversity is mainly determined by competitive exclusion. Thirdly, sampling period was selected to ensure that both autumn-winter and spring-summer weed communities were present, that weed chemical control had already been applied, and that crops had achieved maximum ground-cover. These methodological restrictions allow a great number of fields in similar phenological stages to be surveyed. Finally, effects of the surrounding landscape of each sample site were also considered in this methodology, since surveyed fields were randomly selected among landscapes of similar heterogeneity and with the same soil type.

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# Growth and yield of *Coffea arabica* L. in the Soconusco, Chiapas, Mexico as influenced by both soil cover management and biodiversity of concomitant weed-insect associations

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## Introduction

Chiapas is the most important coffee growing region of Mexico. Declining prices on the world coffee markets had a disturbing effect on the social and economic situation in the Soconusco, by far the most productive and extended coffee growing area (76000 ha) of Chiapas (Marroquín *et al.* 2004). Coffee ecosystems with *Coffea arabica* and *C. canephora* are characterized by different cropping intensities, high diversity of soil and climatic conditions and are distributed between 450 and 1800 m a.s.l. (Pohlan 2002a). These eco-physiological conditions and crop management practices contributed to important changes in biodiversity of soil cover and of concomitant weed-insect associations (Pohlan 2002b; Aguilar 2001; Staver 1998; Nestel *et al.* 1994). The coffee berry borer (*Hypothenemus hampei* Ferrari) resurged and is actually the most important pest of coffee. Its biological control has been studied (Barrera 2002; Baker 1999), but the interactions between type of soil cover, this *Scolytidae* and other insects remain relatively unknown.

## Materials and methods

Field experiments were conducted from 2001 to 2004 on the Finca Argovia, (15°07'962'' N, 92°18'177'' W, altitude 622 m a.s.l. and average inclination of 48%) to determine the effects of different soil cover periods with *Canavalia ensiformis* L. and of natural weed community on the growth and yield parameters of coffee (*C. arabica* L.), cv. Caturra (Pohlan 2002a). The experimental block was 120 x 20 m large and arranged as a randomised complete design with 6 treatments in plots of 20 x 20 m, within which 12 random coffee trees samples were recorded. Soil cover and trap analyses were replicated 4 times per plot. Growth and yield parameters of coffee were recorded and the abundance and dominance of natural weed community and of soil cover represented by *C. ensiformis* determined at two-monthly interval for four years. The effects of different soil cover management regimes on the structure of comparative biodiversity of weeds and insects were evaluated between November 2002 and November 2003 and compared with 2 different traditional coffee systems. In this period, the coffee berry borer (CBB / *H. hampei*) and other insects were captured 14 times with ECO-IAPAR - traps at 2 elevations (50 cm and 150 cm).

Table 1: Characteristics of soil cover treatments

Treatment	Soil cover and shade management
a1	Monthly hand cutting of <i>Canavalia</i> vine-shoots and total cut in April
a2	<i>Canavalia</i> cut in February (blossom stage) & mulching between coffee rows
a3	<i>Canavalia</i> cut in February (blossom stage) and mulching under coffee plants
a4	<i>Canavalia</i> cut in April (maturity) and mulching between coffee rows
a5	<i>Canavalia</i> cut in April (maturity) and mulching under coffee plants
a6	Weed slashing by hand every 2 month (conventional) and shading by <i>Inga</i> spp.
a7	Weed slashing by hand every 2 month and shading by forest trees ( <i>Chiche</i> / <i>Aspidosperma</i> spp./ and <i>Paraíso</i> / <i>Melia azederach</i> )
a8	Weed slashing by hand every 2 month & temporary shading by plantain

Canavalia was sown yearly by hand at the end of October in two 40 cm wide rows between coffee rows. *Inga* spp. extended their shadow over the coffee into the treatments a1 to a6. They were pruned every 2 years.

## Results and discussion

Treatments with *C. ensiformis* supported smaller numbers of CBB throughout the year. The 150 cm high traps capture more CBB than those at 50 cm height. In the system with conventional weed management and *Inga* (a6), we found the highest CBB presence. In 2002, between 0,3 and 12 % of coffee berries were damaged by CBB and in 2003 between 1 and 9 percent. Canavalia cultivation after coffee harvest as cover crop presents a very effective alternative for reducing CBB, controlling weed infestation and increasing coffee yield.

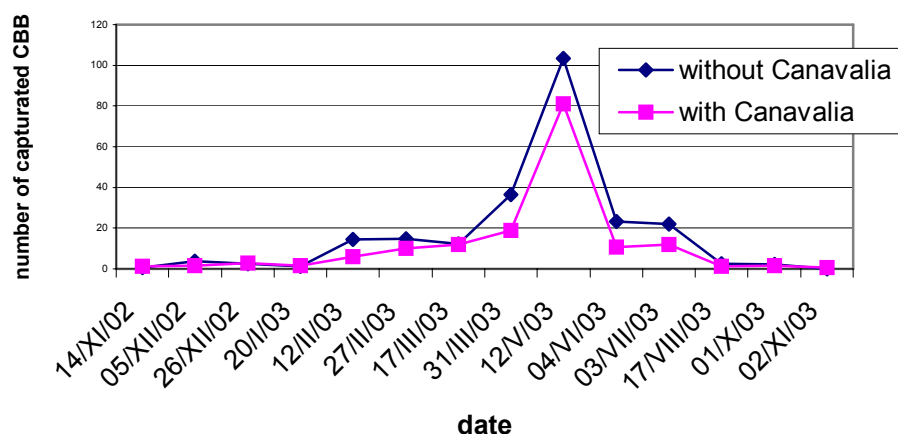


Figure 1. Effects of soil cover on the dynamic of coffee berry borer capture (CBB).

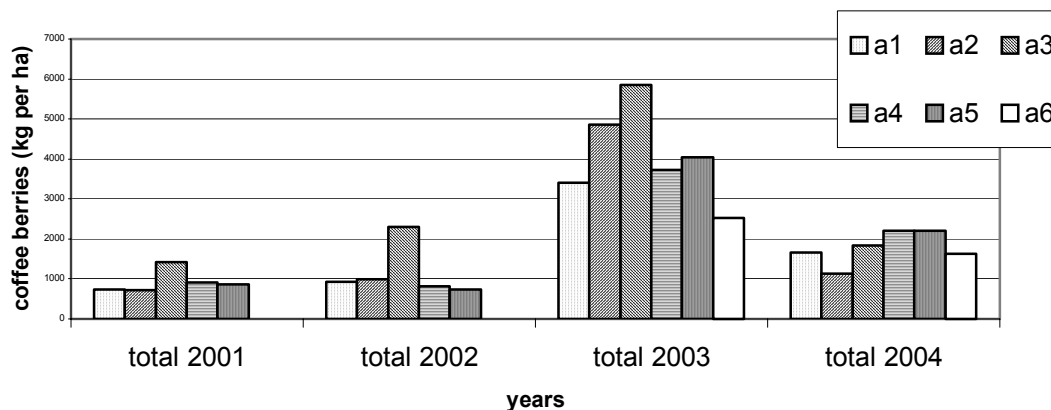


Figure 2. Effects of soil cover management on the yield of coffee berries.

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# Lime reduces the expression of pathogenicity genes of a *Sclerotinia sclerotiorum*-based mycoherbicide

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## Introduction

Giant buttercup (*Ranunculus acris* L.) is a weed in New Zealand dairy pastures that is estimated to cause a loss in national milk solids revenue of \$156 million (Bourdôt et al., 2003). It is difficult to control because of resistance that has developed to the phenoxy herbicides 2,4-D, MCPA and MCPB throughout New Zealand where these chemicals have been repeatedly used (Bourdôt et al., 1990). In addition, cultural practices are considered impractical and/or uneconomic by farmers.

Since its introduction by scientists at AgResearch, Lincoln, Canterbury, a *Sclerotinia sclerotiorum*-based mycoherbicide has shown great potential for the control of giant buttercup in dairy pastures (Cornwallis et al., 1999). However, little is known of the effects of pasture fertilisation and liming on the efficacy of the *S. sclerotiorum*-based mycoherbicide. Experiments were, therefore, conducted to investigate the effect of lime on pathogenicity and expression of key pathogenicity genes of *S. sclerotiorum* isolate S36. An overview of the methods and results is presented.

## Materials and methods

In the first of two experiments, detached *R. acris* leaves were treated with lime at 10 t ha<sup>-1</sup> and/or urea at 50 kg ha<sup>-1</sup> before, simultaneously with, and after inoculation with *Sclerotinia sclerotiorum* S36 (Table 1). A 2-day-old mycelial plug was applied to the leaves, which were held within plastic trays covered with plastic bags and incubated at 20°C under a 12 h photoperiod. The experimental design was a randomised block with five replicates. The percentage leaf area infected was calculated and analysed by analysis of variance.

Table 1. Timing of application of urea, lime and *S. sclerotiorum* on to leaves of *R. acris* in experiment 1.

Treatment number	48 h prior inoculation	24 h prior inoculation	Day 0 Inoculation with <i>S. sclerotiorum</i>	24 h after inoculation	48 h after inoculation
1			Distilled water		
2			Urea		
3			Lime	Urea	
4			Lime		Urea
5			Lime		
6		Lime			
7	Lime	Distilled water			

In the second experiment on the effect of lime on gene expression, leaves were misted with distilled water, inoculated and incubated as described above. The experimental design was a randomised block with three replicates. Twenty-two hours after inoculation (HAI), leaves were treated to run-off with lime (pH 12.5), water (pH 7) or sodium hydroxide (pH 12.45). A single 1-cm sample of infected tissue was taken from the outermost growing margin of the single lesion starting at 24 HAI then every 6 h for a total of 48 h. The infected tissue sample was transferred into a sterile 1.7 ml tube, snap frozen in liquid nitrogen and stored at -80°C until required. Uninfected plant tissue was used as the control. RNA was extracted using Trizol<sup>®</sup> (Invitrogen) and northern hybridisation was done using the ECL Direct Nucleic Acid labelling System (Amersham) as per manufacturer's instructions.

## Results and discussion

In experiment 1, lime applied just before inoculation resulted in 20% less infection by 8 days after inoculation (DAI) compared to the water treatment (Table 2). Lime combined with urea applied either 24 or 48 HAI resulted in 39 and 32% less infection respectively compared to the water treatment 6 DAI. Urea applied at 50 kg ha<sup>-1</sup> did not reduce infection. It was hypothesized that the high pH of the lime contributed to the reduced pathogenicity as it is well established that pathogenicity genes are optimally induced at a pH range of 4–5 (Poussereau et al., 2001). Therefore, experiment 2 was conducted to investigate the effect of lime on the expression of polygalacturonase genes *pg1*, *pg2* and *pg3*, and the protease gene *acp1*. In experiment 2, expression of *pg1/pg2/pg3* and *acp1* was lower under the lime treatment compared the water treatment, while expression was stronger under the NaOH treatment compared to the water treatment.

Table 2. Effect of timing of application of lime/urea on the pathogenicity of *S. sclerotiorum* isolate S36.

Treatment no.	Treatment	Percentage leaf area infected							
		2 DAI		4 DAI		6 DAI		8 DAI	
1	Water	10	b <sup>1</sup>	56	a	88	a	100	a
2	Urea	6	b	38	abc	70	ab	91	ab
3	Lime/Urea 24 HAI	0	c	24	bc	54	b	90	ab
4	Lime/Urea 48 HAI	0	c	24	bc	60	b	91	ab
5	Lime	0	c	19	c	50	b	80	b
6	Lime 24 h prior inoculation	5	bc	44	ab	86	a	100	a
7	Lime 48 h prior inoculation	17	a	54	a	88	a	100	a
	LSD	5		20		25		14	

<sup>1</sup>Values followed by the same letters are not significantly different ( $P < 0.05$ ) as determined by Fisher's LSD

Oxalic acid is an important pathogenicity factor in *S. sclerotiorum* as it decreases the environmental pH to approximately 5 (Bateman & Beer, 1965). Key pathogenicity genes *pg1/pg2/pg3* and *acp1* are induced under low pH conditions (Poussereau et al., 2001) with subsequent secretion of pathogenicity enzymes, such as polygalacturonase (PG) and proteases (Noyes & Hancock, 1980). These results suggest that reduced pathogenicity under the lime treatment may be attributed to a combination of high pH and Ca<sup>2+</sup> liberated from lime. The high pH may have contributed to lowered expression of *pg1/pg2/pg3* and *acp1*, while Ca<sup>2+</sup> may have inhibited the activity of the enzymes, specifically PG, which has been shown to be partially inhibited by 2 mM of calcium (Cabanne & Doneche, 2001). As NaOH had a similar pH to that of the lime treatment, gene expression was predicted to be similar under both treatments. However, gene expression under NaOH was greater than the lime treatment and the water treatment implicating Ca<sup>2+</sup> rather than pH. Further research is planned to investigate the effects of lime, calcium and sodium hydroxide on the production of oxalic acid, polygalacturonase and protease. Results from this research indicate that farm fertiliser management may affect the efficacy of the *S. sclerotiorum*-based mycoherbicide for controlling giant buttercup.

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# Controlling and utilising weeds in apple orchards

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## Introduction

The Hungarian climate is especially dry in the middle of summer and there is less possibility to irrigate orchards than in western Europe. Although mulching is labor intensive and can promote microbial diseases if the soil is covered for a long time, its controlling effect on weeds can be long-lasting. Hay, sawdust, straw, compost, wool dust, manure, coarse bark and black polypropylene can be used as mulching materials for apple orchards (Rifai et al., 2002). In these situations, some soil covering techniques that also protect the soil against the compacting effect of heavy machinery and against the soil losses mainly on hillsides (Papp & Tamás, 1979) and on sandy soils are required. Grass species as living mulch can provide a useful covering without reducing the soil moisture content (Breil & Klöcher, 1981). The use of cover crops, especially of grasses as catch crops in orchards, is important because of the role in regulating soil water content (Parente et al., 1999.). Competition among the covering agents or mixtures and the orchard trees should be avoided. The common method of tilling the whole surface by disking or particularly by rotary hoeing during the season seems to be good for weed control, but it increases the evaporation and energy demands (Garics, 1995).

## Materials and methods

Thirteen plant mixtures were tested as soil covers in apple orchards on a sandy loam soil against a control of naturally occurring weeds. Some parcels were sown in autumn, others in spring. The fruit plantation was established in 1990 and the cover structures were sown in 1992. Soil was prepared for sowing by rotary tiller and seedbed preparator. All the mixtures and species were sown by special grass sowing machine and then they were ring rolled. The tree row distance is 4.5 m, the cover strip is 2 m wide. Date of sowing was April and October. In the case of small 28 m<sup>2</sup> (14 m \* 2 m) plots, sowing was carried out by hand. The experiment was carried out in two stages. The second phase was concentrated only on the successful plots.

Soil moisture and compaction were measured. Measurements were carried out using a special instrument connected to a PSION data recorder in each plot. Percentage plant cover of all plant species was assessed. In the second phase, only the soil moisture and coverage were monitored, with moisture analysed by traditional soil sampling and weighing the wet and air-dried samples. Data were analysed using the SPSS 9.0 package and Tukey's tests.

## Results and discussion

The relative ratio of grass-type plants was lower in the control plots than on the other treatments. The cover percentage of monocotyledonous plants in the treatments was stable in the first 3 years of this study, except in control plots, where significant difference can be found among the years. Permanent cutting depressed the ratio of harmful water prodigal annuals and had let the grasses occupy the free places among the cut-resistant weeds (i.e. *Taraxacum officinale* L.). Under the Hungarian arid climate, the grasses die off during the summer period, covering the surface and reducing evapotranspiration. This situation causes less water consumption than vegetated surfaces, where weeds continue to transpire and use soil moisture. The situation is similar to the case of whole surface tilling methods, when the

evaporation is increased by the loosening effect on soil and the evaporation-reducing cover is destroyed.

All the spring-sown plots had more weed components, especially *Taraxacum officinale* L. and *Plantago major* L, but the Hortobágy plots supported mainly just *Achillea millefolium* L.

#### Soil compaction

In the first period of our experiment (1995 - 1997), the control treatment was one of the best, because of its adaptation to the local climate and soil and the cheapness of the method. Hortobágy mixture, which was the other promising mixture, seems to be worthwhile to combine with some other species or with the local weed flora. The weedy plots with their deeper root system could give a good structure to the soil which seemed to reduce compaction. A similar effect could be recognised in case of the *Poa pratensis* or *Festuca rubra*, where the dense rhizome carpet could give efficient soil protection.

#### Soil moisture

Plant mulches can reduce moisture losses in orchards, but they can be successful only with drought-tolerant local species that can suppress the water prodigal annuals, such as *Amaranthus retroflexus* L.. In extremely dry periods, Szarvas I., which is a special lawn mixture, based on high percentage of *Lolium perenne*, could not cover the soil and allowed water loss via evaporation. The *Festuca rubra* and *Trifolium repens* mixture could protect the soil because their root system is situated in shallow levels without deep rooted dicotyledonous weeds. After the autumn rains, Szarvas I. and the weedy control allowed the water infiltration to deep soil horizons, facilitated by their deeper root system. The soil moisture content under these covers was almost stable even in dry or wet periods, unlike the other mulch seed mixtures.

#### Cover percentage

It was expected that the succession will change plant communities to a locally-adapted mixture, dominated by rhizomatous grasses (e.g. *Festuca rubra*) (Petrányi, 1980.). The first years showed such a tendency, with increasing grass coverage in weedy plots and decreasing amount of fibrous rooted grasses (i.e. *Lolium perenne*) observed. However, a stable mixture has not been achieved, with continuing changes after 6 years. It is expected that these will continue and a weed-and-grass mixture will develop that can protect the soil without competing for soil moisture.

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# Field-scale interactions between weeds and *Aphis fabae* Scop. (homoptera: aphididae) in conventional and low-input sugarbeet.

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## Introduction

Low-input cropping systems are often related to higher weed species diversity and density. Crop pest species have been reported to decrease in fields with a higher vegetational diversity caused by a reduction of crop appearance and nutritional quality (Pimentel, 1961). For *Aphis fabae* Scop. the decision to form a colony and the possibilities to develop large colonies depends on the nitrate level in the plant phloem (Kennedy *et al.*, 1950). If host nutritional quality becomes low, this affects aphid fecundity and they start to develop more winged forms for dispersal. It can therefore be expected that competition with weeds has a negative impact on crop nutrition and susceptibility to aphids. In fields with a higher weed density there is also a higher diversity of possible alternative host plants for aphids.

This study was therefore designed to establish differences in *Aphis fabae* Scop.– crop - weed interactions in two cropping systems differing in tillage technique, fertilization level and weed and pest management. The experimental hypothesis was that, due to their polymorphism aphids are able to optimise resource use through changing infestation dynamics as a response to differences in crop-weed competition in differently managed cropping systems.

## Materials and methods

The study was carried out at the Interdepartmental Centre for Agro-environmental Research 'E. Avanzi' of the University of Pisa in a long-term field scale trial started in 1993 (average field size 6000 m<sup>2</sup>). The sugarbeet crop was included in a 6-year rotation (sugarbeet, durum wheat, grain sorghum, sunflower, durum wheat and set-aside) managed according to a conventional (CS) or a low-input system (LIS) differing in tillage technique, fertilisation level, pest, weed and set-aside management.

Data sampling was carried out in 2001 and 2002 in two adjacent fields at three times: three weeks after crop emergence, two weeks after post-emergence herbicide application and one month later. Data on weed species and sugarbeet density and aphids infestation were collected in 35 fixed plots of 1 x 1 m per field arranged in a regular grid. In CS in 2002, a total of 40 plots were sampled because this field was longer. Aphid infestation was estimated applying various abundance and damage classes described in the literature (Banks, 1954; [www.ipm.ucdavis.edu](http://www.ipm.ucdavis.edu)), and weed presence was determined by density at the first two sampling dates and by the Braun-Blanquet method at the third sampling date.

For measurement of aphid response to crop – weed interaction, an index expressing relative density of sugarbeet with respect to total vegetation density (Sugarbeet/vTot) was used. Data were analysed with nonparametric correlation tests and spatial data analysis was performed with semivariogram analysis and with cross-correlation to test the interpolation model for spatial representation.

## Results and discussion

Sugarbeet density was not different between the two cropping systems and varied around 6 plants m<sup>-2</sup> in both years. Weed species richness varied between 1.6 and 8.1 species m<sup>-2</sup>, was higher in the LIS than in the CS and decreased during the crop growing period. Weed density decreased after the post-emergence herbicide treatment in both systems although density levels were always higher in LIS than in CS. Dominant weed species were similar in both years, but species composition differed more between the two systems in 2002 than in 2001.

Data showed a positive correlation ( $P \leq 0.01$ ) between number of winged forms and total vegetation density during sugarbeet emergence in 2001. In 2002 this correlation could not be verified because sugarbeet was sown earlier and aphid arrival did not coincide with sugarbeet emergence. After herbicide spraying in 2001, there were more dispersal forms in the LIS, especially in those areas where Sugarbeet/vTot was low and competition was high (Fig. 1a). At the same time, in the CS, there were bigger colonies than in the LIS, especially in those areas where Sugarbeet/vTot was high (Fig 1b). Since the development of dispersal forms indicates lower nutritional crop quality, these results indicate that the general crop quality was higher in CS than in LIS, probably due to the higher N fertilization level. The differences within the fields were likely due to weed competition with the crop, which negatively affected crop nutritional quality and thus resulted in the development of more dispersal forms. After post-emergence weed control in 2002 weed composition in CS was dominated by wild beet (3.9 plants  $m^{-2}$ ), whereas in LIS weed species composition was dominated by *Ammi majus* (2.5 plants  $m^{-2}$ ), *Equisetum arvense* (4.6 plants  $m^{-2}$ ) and *Polygonum aviculare* (3.7 plants  $m^{-2}$ ). Aphid infestation severity on sugarbeet was higher ( $P \leq 0.05$ ) in CS than in LIS. In CS there was a correlation ( $P \leq 0.05$ ) between colony size on sugarbeet and on wild beet and between colony number on sugarbeet and on wild beet ( $r = 0.39$  and  $0.34$  respectively; d.f. = 38). It can be hypothesized that aphids are attracted by the inflorescence of wild beet and that they colonize sugarbeet from wild beet.

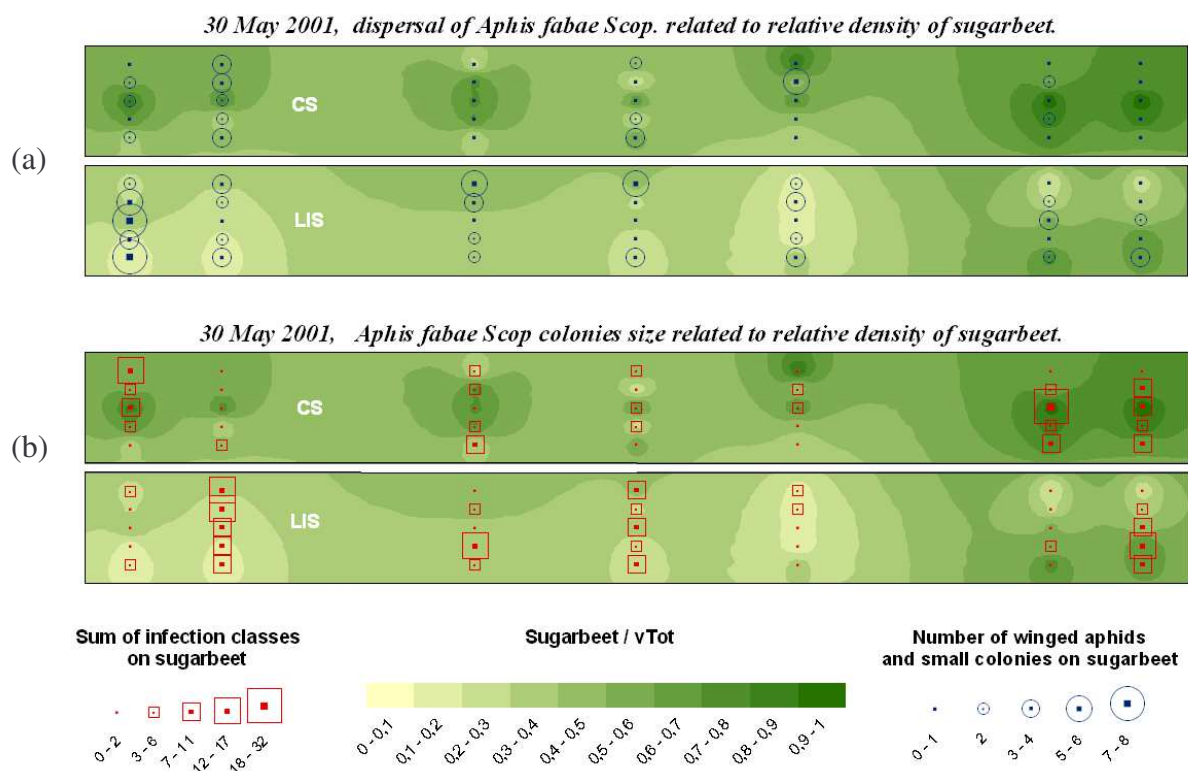


Figure 1. Relationship between (a) winged forms after post-emergence weed control and sugarbeet relative density and (b) colony size and sugarbeet relative density.

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# Classical biological control of European exotic environmental weeds: opportunities and constraints

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## Introduction

Classical biological control remains the only tool available for permanent ecological and economic management of targeted invasive exotic species. The release of specialist natural enemies onto exotic invasive species that have escaped these through introduction into non-native regions aims to restore an ecological equilibrium. The enemy release hypothesis describes how escape from natural enemies can act to increase invasiveness in exotic species. This is evidenced by a 60% success rate against targets using classical biological control programs around the world. The biological control approach engenders some concern however because of historically crude, unscientific, pest management activities and evaluations of threats that released natural enemies will unnaturally antagonize non-target species where released. Direct negative effects are now entirely predictable, however, through recognized processes of risk assessment. This means biological control remains an ecologically sound approach to invasive species management through a precautionary approach for deliberately introducing beneficial exotic species.

137 species of classical biological control agent have been released against exotic insect pests in Europe since 1901 (Greathead & Greathead, 1992). Classical biological control of weeds has also been practiced in some Eastern European countries against annual ragweed (*Ambrosia artemisiifolia* L.; Julien & Griffith, 1998), however to date no exotic biocontrol agents have been released in Western Europe against an exotic weed. This dearth has occurred in the face of increasing numbers of exotic invasive plants taking over National Parks, forests, amenity areas and islands in this region (Clement & Foster, 1994; Muller, 2005; <http://www.ceh.ac.uk/epidemie/>). As an example, amongst the 217 potentially noxious exotic plants listed in France, already 60 are already causing damage to ecosystems. In this paper we review why classical biological control of exotic plants remains largely untested in Europe. We also review all widespread exotic plants in Europe for their occurrence and impacts and potential as targets for classical biological control from both an ecological and economic perspective. Finally we review the regulatory framework under which such releases could be made within the EU and its constituent countries to predict whether this approach can have a useful role to manage invasive exotic weeds in Europe in the future.

## Materials and methods

We reviewed the scientific literature on alien plant species across Western Europe to generate a list of species of widespread concern and significant impacts in both Mediterranean and temperate climates and terrestrial and aquatic ecosystems. This list of over 200 exotic species was then assessed for biological control potential based on a) degree of historical success of biocontrol against these targets, ecological homologues and related species, b) taxonomic isolation of these weeds from European native flora as a measure of risk of non-target damage, c) likelihood of suitable natural enemies being available as potential agents and d) likelihood for conflicts of interest. We used this information to develop a priority list of possible biological control weed targets in Europe.

The International and European legislation relating to biological control in particular and the introduction of beneficial exotic organisms in general were reviewed. The current

legislative framework for classical biological control activities in Europe is described and constraints this may pose for biological control releases of arthropods or plant pathogens were evaluated.

## Results and discussion

Our review identified 19 alien plant species (Table 1) with significant and 12 additional species with slight potential as targets for classical biological control in Europe.

Table 1. Exotic invasive plants in Western Europe prioritised as potential biocontrol targets.

Species	Life form <sup>a</sup>	Country of origin	EU climate distribution	Genus native to Europe	Past or current biological control publications
<i>Buddleja davidii</i>	Ph	China	Temperate	No	Yes
<i>Fallopia japonica</i>	Ge	Japan	Temperate	No	Yes
<i>Accacia dealbata</i>	Ph	Australia	Med	No	Yes
<i>Azolla filiculoides</i>	Hy	N America	Temp/Med	No	Yes
<i>Ailanthus altissima</i>	Ph	China	Temp/Med	No	Yes
<i>Impatiens glandulifera</i>	He	India	Temperate	Yes	No
<i>Rhododendron ponticum</i>	Ph	S Europe	Temp/Med	Yes	Yes
<i>Robinia pseudoacacia</i>	Ph	N America	Temperate	No	No
<i>Senecio inaequidens</i>	He	S Africa	Temp/Med	Yes	Yes
<i>Ambrosia artemisiifolia</i>	Th	C America	Temp/Med	Yes	Yes
<i>Carpobrotus edulis</i>	Ch	S Africa	Temp/Med	No	No
<i>Heracleum mantegazzianum</i>	He	W Asia	Temperate	Yes	Yes
<i>Solanum elaeagnifolium</i>	He	S America	Tem/Med	Yes	Yes
<i>Baccharis halimifolia</i>	Ph	N America	Med	No	Yes
<i>Hydrocotyle ranunculoides</i>	Hy	N America	Temp/Med	Yes	No
<i>Ludwigia peploides</i>	He	S America	Temp/Med	Yes	Yes
<i>Crassula helmsii</i>	Hy	Australasia	Temperate	Yes	No
<i>Elodea canadensis</i>	Hy	N America	Temperate	No	No
<i>Myriophyllum aquaticum</i>	Hy	S America	Temp/Med	Yes	No

<sup>a</sup> Ph, phanerophyte; Ge, geophyte; Hy, hydrophyte; He, hemicryptophyte; Th, therophyte; Ch, chamaephyte. NB: spp. arranged (by lines) in groups of decreasing priority, but of equal priority within a group.

EU legislation for the importation and release of exotic organisms for biological control relates to EU Directives that put into action components of two relevant International Conventions to which the EU is party; the Convention on Biological Diversity and the International Plant Protection Convention. These directives currently allow for the importation and release of exotic organisms under the code of conduct for such activities in the IPPC. Macro-organisms can be released following each country administering the code, but the decision for release by one country should include discussions with other affected countries in the EU. The responsibility for any damage that ensues remains with the organization that made the release. Micro-organism releases currently appear to require the same regulatory clearance as agrochemicals: a pathway completely alien to the use of classical biological control agents.

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# The potential for managing weeds to minimise impacts on yields and enhance the ecological diversity of arable fields – a winter wheat case study.

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## Introduction

Bird, invertebrate and plant numbers have declined in arable ecosystems over the last 30 years, as evidence of a decline in biodiversity. Weed species differ in their value for the arable ecosystem and in their impact on the crop. Selective removal of some weed species whilst retaining others, may deliver biodiversity value without jeopardising crop production. Two trials were established, in autumn 2003, to compare and target usage of different herbicides to selectively leave *Tripleurospermum inodorum* L. as a food species for birds and invertebrates in winter wheat. Trials were sited at ADAS Boxworth (Eastern England, chalky clay loam) and Rothamsted Research (Central England, silty clay loam). Whilst aiming to retain *T. inodorum* in a background of other broad-leaved weeds, the Boxworth trial focussed on selective removal of volunteer *Brassica napus* ssp. *oleifera* (DC) Metzg., whilst the Rothamsted experiment aimed to remove *Stellaria media* L.

## Materials and methods

At Boxworth, winter wheat cv. Malacca was sown on 3 October 2003 at approx.310 seeds/m<sup>2</sup>. Target weed species were hand broadcast on half of the 48 plots (4 blocks x 12 treatments) measuring 3 x 14.4 m, each sown with *T. inodorum* at 3850 seeds/m<sup>2</sup> and *B. napus* at 200 seeds/m<sup>2</sup>. Clodinafop-propargyl was applied to all plots to minimise grass weeds. However, herbicide-resistant *Alopecurus myosuroides* Huds. was a problem and necessitated an overall application of propoxycarbazone-sodium @ 42 g ai/ha on 2 March 2004. Six experimental herbicide treatments comprised untreated, amidosulfuron, two doses of florasulam (not currently approved in sequence with propoxycarbazone-sodium) or single doses of pendimethalin (applied pre-emergence) or mecoprop-p (Table 1). At Rothamsted, winter wheat cv. Consort was sown on 29 September 2003 at approx.350 seeds/m<sup>2</sup>, with target weed species broadcast on all but the 4 weed-free plots of the 32 plots (4 blocks x 8 treatments) measuring 3 x 10 m, each sown with *T. inodorum* at 2000 seeds/m<sup>2</sup> and *S. media* at 1000 seeds/m<sup>2</sup>. The herbicide treatments at Rothamsted comprised full weed-free control (flupyr-sulfuron+pendimethalin), or clodinafop-propargyl followed by either of two doses of fluroxypyr, carfentrazone-ethyl+mecoprop-p, or mecoprop-p or untreated in spring (Table 1).

Effects of herbicides on *T. inodorum* seed production, weed numbers and weed biomass were assessed. Winter (Dec-Jan) wheat and weed counts were followed by a spring biomass assessment on all plots and a summer biomass on all plots at Rothamsted, but only on selected plots at Boxworth. These were followed by summer weed counts and targeted individual *T. inodorum* plant biomasses. At Rothamsted, *T. inodorum* capitula weights, numbers and seeds per capitula were assessed against plant dry weights and weeds/m<sup>2</sup>. Crop yields were measured.

## Results and discussion

At Boxworth, populations of 156/m<sup>2</sup> *T. inodorum* and 112/m<sup>2</sup> *B. napus* plants were established. The propoxycarbazone-sodium treatment retained *T. inodorum* and some other desirable broad-leaved species, but there was less than full control of *A. myosuroides*. The spray killed the volunteer *B. napus* and *T. inodorum* vigour may have been reduced, as numbers declined through the spring on otherwise untreated plots. Crop dry weights were not

significantly affected by the treatments, but harvest yields showed differences. *T. inodorum* survived all treatments and varied in dry weight between sown treatments from 0.03 g to 0.62 g/plant with the sown untreated giving a total dry weight of almost 36 g/m<sup>2</sup>. Yields reflected *T. inodorum* numbers. Florasulam-treated plots had least *T. inodorum* and gave higher yields than untreated. Previous use of propoxycarbazone-sodium and speed of weed control may have affected yields.

Table 1. Wheat yield data from Boxworth (plot combine harvested on 31/8/04) and Rothamsted (hand harvested 25-26/8/04) –weeds sown except where shown below.

Treatment (All rates are given in g ai/ha)	Yield t/ha (At 85% moisture)		% yield loss compared to most effective treatment	
	Boxworth	Roth-sted	Boxworth	Roth-sted
Untreated	10.07	6.82	5.8	18.9
flupyr-sulfuron + pendimethalin @ 10 + 120 g – no weeds sown		8.40*		0.0
amidosulfuron @ 20 g	10.28		3.8	
pendimethalin @ 1320 g	10.29		3.7	
mecoprop-p @ 600 - 690 g	10.37	7.80*	3.0	7.2
mecoprop-p @ 172.5g		7.26		13.6
florasulam @ 7.5 g	10.69*		0	
florasulam @ 3.75 g	10.51*		1.7	
fluroxypyr @ 400 g		7.69*		8.4
fluroxypyr @ 100 g		7.06		16.0
carfentrazone + mecoprop @ 15 +600 g		7.55		10.1
carfentrazone + mecoprop @ 4 + 150 g		7.20		14.3
<b>S.E.D</b>	<b>0.170</b>	<b>0.397</b>		

\* = Significant at 5%

At Rothamsted, autumn populations of *T. inodorum* and *S. media* were 103 and 129 plants/m<sup>2</sup>, respectively. *T. inodorum* survived all the treatments, but herbicides affected weed size/biomass as well as numbers. *T. inodorum* capitula/seed numbers were proportional to biomass and weed number. Maximum dose fluroxypyr @ 400g ai/ha, reduced the *T. inodorum* biomass in mid-summer by nearly 90%. However, individual plant weights at harvest were reduced by a more modest 60% (to 0.35g dry weight). Fluroxypyr controlled *S. media* and other weeds, but permitted the survival of some *T. inodorum*, despite the severe reduction in vigour, resulting in 108 to 266 capitula/m<sup>2</sup> depending on dose applied. Mecoprop alone and in mixture with carfentrazone resulted in the production of 231 to 256 capitula/m<sup>2</sup> but at the lower rates were not effective on *S. media*. Yields were significantly lower than weed-free treatments, especially where low rates of were applied. Even the 7.2% yield loss from 690g ai/ha mecoprop for wheat at £60/T in 2004 would have cost £38/ha (£27/ha net after herbicide application) and such costs for biodiversity benefits may have to be only on part of a farm to be economically acceptable.

These trials showed potential to selectively manage *T. inodorum*, for example, with high rate of carfentrazone + mecoprop in the presence of *S. media*. Many other treatments showed potential to ensure some *T. inodorum* survival, with associated seed production and other resources for invertebrates and with restricted growth of other broad-leaved weeds. Grass weeds complicated weed management. We conclude there is some potential for managing weeds that contribute to biodiversity in wheat, through using ‘appropriate’ doses of herbicide, but it challenges economic sustainability and makes weed control decision-making more complex.



# Functional diversity within the UK arable weed flora

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## Introduction

Plants adapted to different habitats will have distinct functions within the arable eco-system, in terms of supporting higher trophic groups. Eco-system function will be determined by plant strategy mediated by life history and growth traits. It is desirable to maximize functional diversity at the farm scale by promoting a diversity of habitats in space and time. This will involve judicious weed management within a range of crops, as well as managed field margins and set-aside. This approach raises a number of scientific challenges. Firstly, arable plants need to be grouped in terms of their eco-physiological traits. The effect of management filters, such as rotation, on the population dynamics and productivity of different functional groups can then be predicted using simulation models (Booth and Swanton 2002). Finally, the potential benefit to higher trophic groups of different arable plant communities needs to be quantified (Marshall et al. 2003). This project screened 20 annual weed species and two crops for a range of eco-physiological traits. The results of an initial multivariate analysis clustering the species in functional groups are presented.

## Materials and methods

A combination of field and pot experiments were used to parameterise the eco-physiological profiles of a range of annual weeds and crops. Plants were grown in small plots in the field with a crop of winter wheat and sampled at regular intervals to plot functions of growth characteristics (height, leaf: stem ratio, specific leaf area, reproductive partitioning, time of flowering and leaf senescence) against photo-thermal time. Seedlings of each species were also grown in pots and sampled at much shorter intervals to measure relative growth rate (RGR) in the exponential phase. This experiment was repeated four times, twice in the autumn and twice in the spring, to parameterise a simple model of the response of RGR to the temperature and light. A separate pot experiment was done to measure the rate of CO<sub>2</sub> assimilation of the 24 species at four temperatures. The aim was to combine information from all of the experiments to fully parameterise the INTERCOM model of crop / weed competition (Kropff and Spitters 1992).

## Results and discussion

Because the matrix of plant species by eco-physiological traits will not be complete until the end of 2005, an initial multivariate analysis is presented here using the traits for which information is available for all the species (seed size, autumn RGR, spring RGR, maximum height, time of first flowering, time of leaf senescence). Four groups were identified representing four distinct plant strategies for growth and reproduction (Figure 1). Group I was composed of tall species with a large seed, relatively low seedling growth rate and late flowering (*Avena fatua*, *Anisantha sterilis*, *Galium aparine*, *Triticum aestivum*). Species in Group II were of a similar height and late flowering but had a much smaller seed and high seedling growth rate (*Alopecurus myosuroides*, *Tripleurospermum inodorum*, *Papaver rhoeas*, *Lolium multiflorum*, *Brassica napus* and *Sinapis arvensis*). Group III included the classic ruderal species – short, small seed, high seedling growth rate and early flowering (*Stellaria media*, *Veronica persica*, *Lamium purpureum*, *Senecio vulgaris*, *Poa annua*,

*Capsella bursa-pastoris*). Finally, Group IV was composed of short species with a small seed but with a low seedling growth rate and late flowering. These were the more stress tolerant ruderals (*Viola arvensis*, *Myosotis arvensis*, *Geranium molle*). This group also contained the obligate spring germinating species (*Chenopodium album*, *Polygonum aviculare* and *Fallopia arvensis*) which will probably be separated out in the final analysis. A number of trade-offs were identified between traits. For example, seed size was negatively correlated with seedling growth rate (Storkey 2004).

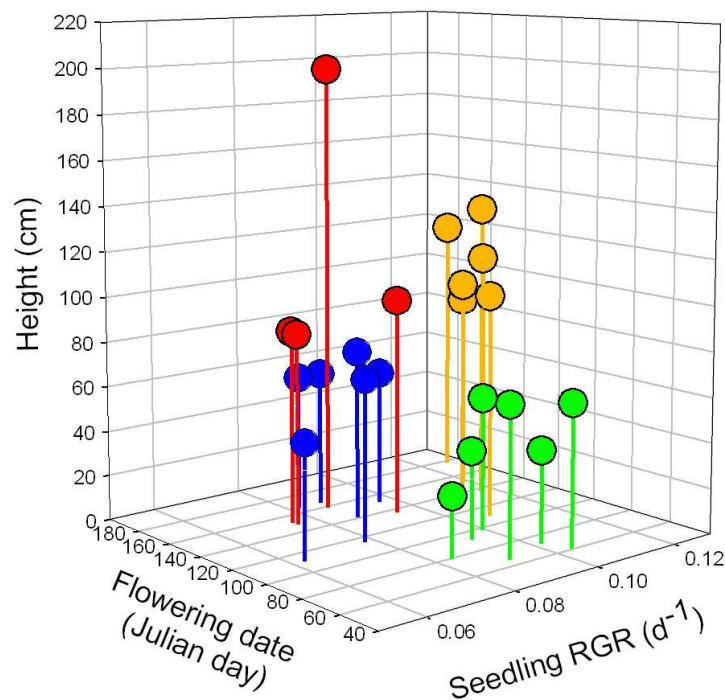


Figure 1. Summary of multivariate analysis based on six eco-physiological traits but illustrated using three. ● Group I, ● Group II, ● Group III, ● Group IV. See text for explanation.

The contrasting eco-physiology of the different groups will result in different responses to management filters such as fertilizers and rotation. Of particular importance are the phenological characteristics, such as flowering time, which will determine the windows of opportunity for regeneration for different groups within any particular farm management regime. The effect of these different filters on the indigenous weed community can be analysed using simulation models of competition and population dynamics.

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# ***Tamarix* (saltcedar) species, their natural enemies and allelopathic effects in the Eastern Mediterranean region of Turkey**

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## **Introduction**

The genus *Tamarix* comprises about 54 species of shrubs worldwide (Baum, 1978), including several weedy species in riverbeds, riverbanks, water bodies and also in some cultivated areas and pastures. Five species of this genus were reported in the flora of Turkey (Baum, 1967). It has been reported that saltcedar reduces the livestock stocking capacity by displacing forage grasses, by using ground water or irrigation water that otherwise could be available to grow forage or crop plants, by increasing soil salinity, and by increasing the incidence of fires (DeLoach et al., 2000). There are several species of natural enemies that attack saltcedars (*Tamarix ramosissima* Ledeb. and *Tamarix gallica* L.). The most promising ones are *Psectrosema* spp., *Hypophyes pallidulus*, *Corimalia tamarisci*, *Coniatus tamarisci*, *Stylosomus* prob. *tamaricis* and *Agdistis tamaricis* (Sobhian et al., 1998). Large populations of *Aceria tamaricis* were found on *Tamarix smyrnensis* Bunge in Efes near İzmir in Turkey in 1995. This was previously reported as an eriophyid mite on this weed species. Also, some similar gall symptoms associated to *A. tamaricis* was noticed on *Tamarix gallica* L. (De Lillo & Sobhian, 1996). The objectives of this study were to identify the weedy *Tamarix* species and their distribution in the Eastern Mediterranean region of Turkey, their natural enemies with potential for its biological control, and to study the allelopathic effects of *Tamarix* spp.

## **Materials and methods**

Surveys were conducted in the Eastern Mediterranean Region of Turkey during 2002–2003. At 10 km radius of Adana a quadrat sample was taken and 1000 m<sup>2</sup> of area was marked where *Tamarix* species were found (Uygur, 1985). We stopped at 136 points in total. Sixty-six samples were collected to diagnose *Tamarix* species. Identification was done according to Baum (1967). The population density parameters were calculated (Odum, 1971). Also, in every site in these areas, a maximum of 10 plants were examined for the presence of any insects and pathogens; 242 plants in 2002 and 270 in 2003 were examined. Insect sampling were done according to Karaca et al. (2002).

Furthermore, *Liocleonus clathratus* (Coleoptera: Curculionidae), which was brought from Antalya, was used in a host specificity test in the screenhouse (dimensions 210 cm x 320 cm x 596 cm). Subsequently, 100 adults of *L. clathratus* were released on 3-year old *Tamarix smyrnensis* (two different origins), *Tamarix tetrandra* Boiss & Heldr. *Tamarix aphylla* (L.) Karst., *Myricaria germanica* (L.) Desvoux, *Populus alba* L., *Passiflora caerulea* L., *Limonium angustifolium* (Tausch.) Turril., and *Alhagi pseudalhagi* (Bieb.) Desv. (10 plant species x five replications).

Also, during this study, *T. smyrnensis* leaves were collected in different dates from the field. To prepare the watery extract of *T. smyrnensis*, entire leaves of the plant was collected from their natural habitat and put into water in a plastic container under laboratory conditions. After 1 week, extracts were filtered and kept in the refrigerator for 21 days. The germination tests used these extracts and also distilled water as control on seeds of crops, soybean (*Glycine max*), lettuce (*Lactuca sativa*), wheat (*Triticum vulgare*) and cotton (*Gossypium hirsutum*) and weed species and were conducted according to Uygur (1985) in order to investigate the effect of *T. smyrnensis* extracts on seed germination. Data were evaluated using a *t*-test at *P* = 0.05. Effectiveness of the extracts was shown using Abbott's formula.

## Results and discussion

Four *Tamarix* species were identified in the Eastern Mediterranean region of Turkey. *Tamarix smyrnensis* was found as the most dense and widespread species in this region (Table 1). The percentage cover of *Tamarix* species in the all surveyed area was shown as general coverage and the coverage of *Tamarix* in the location which was found was shown as special coverage in the Table.

Table 1. The determination of saltcedar species (*Tamarix* spp.) according to their frequencies, general coverage, and special coverage proportions (%)

Identified saltcedar species	% Frequency	% Gen. coverage	% Spec. coverage
<i>Tamarix smyrnensis</i> Bunge	28.9	13.3	45.9
<i>Tamarix hampeana</i> Boiss & Heldr	5.8	1.4	23.8
<i>Tamarix tetrandra</i> Pallas ex Bieb	1.9	0.9	45.0
<i>Tamarix parviflora</i> D.C.	1.9	0.8	40.0

The infestation rate of *Corimalia* spp. (Col: Curculionidae), which damages the flower buds of plants, was 68.6% in 2002 and 82.2% in 2003. The infestation rate of *Psectrosema* spp. (Dip: Cecidomyiidae), which forms galls on young green shoots, was 49.2% in 2002 and 53.7% in 2003. Also, the infestation rate of *Parapodia* spp. (Lep: Gelechiidae), which forms galls on side branches, was 35.1% in 2002 and 34.1% in 2003. The infestation rate of *Eriophyidae* (Acarina) that forms galls on flowers and axial buds was 4.5% in 2002 and 2.2% in 2003. According to these observations, *Corimalia* spp. was found to be the most widespread species on *Tamarix* spp. It was also determined that *Liocleonus clathratus* did not adapt to the test plants' habitat in the screen house. No gall formation was found on the root crown of any test plant. According to the Abbot formula, the water extract of *T. smyrnensis* reduced seed germination by 22.6% (extract:  $65.1 \pm 6.3\%$ , control:  $84.1 \pm 4.6\%$ ) in soybean; 30.2% (extract:  $67.1 \pm 11.4\%$ , control:  $96.1 \pm 0.8\%$ ) in lettuce; 12.1% (extract:  $86.3 \pm 3.9\%$ , control:  $98.1 \pm 0.6\%$ ) in wheat; but did not affect the seed gemination of cotton (extract:  $72.4 \pm 6.4\%$ , control:  $73.3 \pm 2.2\%$ ). Furthermore, the extract of *T. smyrnensis* reduced the seed germination of weeds by 60.1% (extract:  $30.0 \pm 7.9\%$ , control:  $75.2 \pm 1.1\%$ ) for *Amaranthus retroflexus* L.; 31.6% (extract:  $57.0 \pm 6.6\%$ , control:  $83.3 \pm 3.1\%$ ) for *Lolium perenne* L.; 41.3% (extract:  $25.1 \pm 7.0\%$ , control:  $42.9 \pm 3.6\%$ ) for *Portulaca oleracea* L.; 82.2% (extract:  $5.4 \pm 1.2\%$ , control:  $30.4 \pm 7.5\%$ ) for *Silybum marianum* (L.) Gaertn.; but did not affect the seed germination of *Avena sterilis* L. (extract:  $57.4 \pm 8.1\%$ , control:  $76.1 \pm 5.4\%$ ).

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# Tillage effects on the diversity of the surface soil seed bank in a 4-year winter crop rotation

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## Introduction

Recently, European Community agricultural policy has strongly encouraged conservation tillage practices in order to decrease soil loss. In this context, information for weed management is needed, as the effect of cultivation on the weed flora and weed seeds and propagules in the soil vary with depth and type of tillage (Froud-Williams et al., 1984; Grundy et al., 1999). The main objective of this study was to compare the surface soil seed banks formed after eight years (two complete rotations) of winter cropping under different tillage systems in a Mediterranean climate.

## Materials and methods

A field survey was carried out on 12 July 2001 to assess the richness, abundance and spatial distribution of the viable seeds in the soil in three 30x90 m<sup>2</sup> plots under winter crop rotation (pea-wheat-wheat-barley) established in 1993 and located at Torre Marimon (Caldes de Montbui, Barcelona). Three tillage systems were compared: mouldboard plough (conventional tillage, CT, 25 cm depth), chisel (minimum tillage, MT, 15 cm depth) and no tillage (NT). The survey area was divided into two blocks because variation could be expected due to slope-associated effects (8% to 10% E-W). Thirty-six samples of soil were taken in a 3x3 m grid within each experimental plot (block x tillage system) using cylinders 10 cm deep and 4.5 cm in diameter. Apparently viable seeds were identified and counted in 150 g sub-samples processed following Carretero's method (1977). Analyses of variance testing the tillage and block effects on the amount of seeds were done for the two most abundant weed species using the GLM procedure of SAS (1999). The raw data were used to calculate the Shannon-Weaver diversity indices of the surface soil seed bank of each tillage system, as well as a measure of evenness (Margalef, 1986).

## Results and discussion

The species and numbers of seeds varied with the tillage system that had been practised. In total 769 seeds of 22 weed species were counted in the MT samples, 544 seeds of 23 species in NT, and 296 seeds of 16 species in CT. Percent distribution of the seeds of 18 species that achieved abundances of at least 1% are given in Figure 1 (results for 15 species are not shown). The Shannon-Wiener diversity indices obtained were 3.16 for MT, 3.00 for NT, and 2.50 for CT, and the corresponding evenness values were 0.70, 0.66 and 0.55. The NT weed community also showed the highest values of richness, diversity and evenness at the harvest time of the preceding crop rotation phases (Mas & Verdú, 2003), but CT did not have the lowest, which corresponded to MT. Dorado et al. (1999), who evaluate weed seed banks in cereal crops in central Spain, also found larger and more diverse weed seed banks in no tillage plots than in conventional ones.

Only nine species were common to all three tillage systems (Fig. 1). Among them *P. aviculare* and *D. eruroides* are able to compete with the winter crop plants of the rotation. They showed very different field distributions: seeds of *D. eruroides* were significantly ( $P < 0.001$ ) more abundant under CT than under the other tillage systems tested, while those of *P. aviculare* were most abundant under MT. The findings for *P. aviculare* supported the



hypothesis that seedling recruitment could be limited in NT, with respect to MT and CT (Verdú & Mas, 2004).

Table 1. Significance of the sources of variation resulting from the ANOVA, and means obtained for each tillage system considering data for two species.

Species	Source of variation		Mean (seeds 150g soil <sup>-1</sup> )*		
	Block	Tillage system	Conventional	Minimum	No tillage
<i>Diploaxis erucooides</i>	0.5171	<.0001	1.50 A	0.57 B	0.32 B
<i>Polygonum aviculare</i>	0.4905	0.0001	1.43 a	2.21 b	1.01 a

(\*) For each species, means (back-transformed) with different letters were significantly different at  $P = 0.05$  (Tukey's HSD test), based on transformed (square root of value+3/8) data.

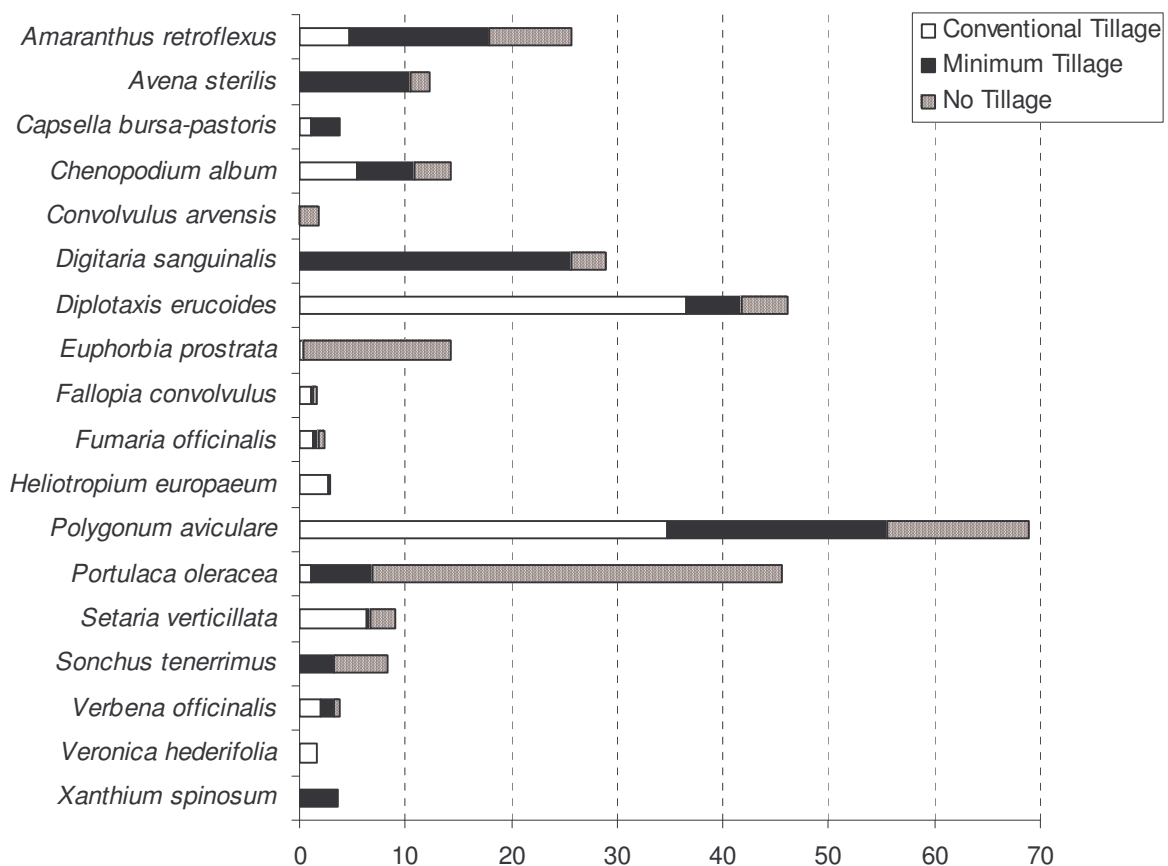


Figure 1. Specific relative contribution to the soil seed bank comparing three tillage systems. Species with a contribution lower than 1% do not appear in this figure.

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# Dominant phenolic compounds in ruderal species of *Reynoutria* and *Impatiens*

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## Introduction

Plant phenolics are biologically active compounds that are produced as secondary metabolites in different parts of plants. Most of these compounds have anticancer, antioxidative, antimicrobial and fungicidal properties. Therefore, we suppose that some plants with phenolic compounds are more persistent and are more competitive among other plants ([www.ars-grin.gov/duke](http://www.ars-grin.gov/duke)).

There are two introduced invasive genera in the Europe that are spreading from ruderal to natural habitats. The first one is *Reynoutria*, which contains three tall perennial species. The second one is *Impatiens*, which contains three annual species (one of them, *I. noli-tangere* is native to Europe, but also grows in ruderal habitats).

Our research is focused on phenolic compounds in different parts of *Reynoutria* and *Impatiens* species. The genera differ in their life histories, but both contain some phenolic compounds, which may be necessary for their invasive strategy.

## Materials and methods

The *Reynoutria* specimens (five from each species: *R. japonica*, *R. sachalinensis*, *R. x bohemica*) were collected in May 2002 in the Český Krumlov district (Czech Republic). Leaves and flowers of *Reynoutria* were dried at laboratory temperature, rhizomes were lyophilized, then the materials were pulverized and extracted with diluted methanol.

Five plants of each *Impatiens* species (*I. noli-tangere*, *I. parviflora*, *I. glandulifera*) were collected in July 2003 in the Č. Budějovice district (Czech Republic). The plants were divided into roots, leaves and flowers and mixed samples from five plants of each part were made. The materials were extracted fresh.

All samples were analyzed using HPLC with a DAD detector on a C18 column (2 x 150 mm, 3 µm), in a water–acetonitrile gradient with the addition of trifluoroacetic acid. Stilbenes (resveratrol and its derivatives) were detected at 315 nm and others phenolic compounds at 220 nm.

## Results and discussion

The different parts of the investigated plants (underground parts, leaves, flowers) contain many biologically active compounds such as stilbenes (resveratrol and its derivatives), quercetin and its derivatives, catechin, epicatechin, naphthoquinones, chlorogenic acid, caftaric acid and derivatives of caffeic acid (Vrchotová et al., 2004 a,b,c). Some of the dominant phenolic compounds are summarized in Table 1. The extract from above ground parts of all investigated plants contained many quercetin derivatives. The dominant compounds from the underground parts of plants are very different.

It is known that underground parts of *Reynoutria japonica* contains large quantities of stilbenes (Vastano et al., 2000). We found that stilbenes are also present in *R. sachalinensis* and *R. x bohemica* (the smallest amounts in *R. sachalinensis*). There are also large quantities of catechin and epicatechin in the rhizomes of the *Reynoutria* species. On the other hand, naphthoquinones are dominant in extracts from roots of the investigated *Impatiens* species.

A high concentration of these phenolic compounds (especially resveratrol and naphthoquinones) is probably important for the invasive properties and high resistance to insects and pathogens (Harborne, 1993) of both *Reynoutria* and *Impatiens* species.

Table 1. Summary of some dominant phenolic compounds in *Reynoutria* and *Impatiens*

	Underground organs	Leaves	Flowers
<i>Reynoutria japonica</i>	stilbenes catechins	quercetin derivates caftaric acid chlorogenic acid catechins	quercetin derivates catechins
<i>Reynoutria sachalinensis</i>	catechins stilbenes	quercetin derivates catechins caftaric acid	quercetin derivates catechins
<i>Reynoutria x bohemica</i>	stilbenes catechins	caftaric acid chlorogenic acid catechins	quercetin derivates catechins
<i>Impatiens noli-tangere</i>	naphthoquinones	caffeic acid derivates quercetin derivates	quercetin derivates quercetin
<i>Impatiens parviflora</i>	naphthoquinones	caffeic acid derivates quercetin derivates	quercetin derivates quercetin
<i>Impatiens glandulifera</i>	naphthoquinones	quercetin derivates	quercetin derivates quercetin

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# Vectoring of *Fusarium tumidum* spores by four insect species for biological control of gorse

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## Introduction

Gorse (*Ulex europaeus* L.) is a serious weed in New Zealand pastures and forest plantations adversely affecting agriculture and indigenous biota (Froude, 2002). A fungus, *Fusarium tumidum* Sherb., which occurs naturally in New Zealand, has some potential as a mycoherbicide against gorse (Morin et al., 2000). However, formulation of an effective mycoherbicide has met with considerable difficulties. An alternative method of disseminating spores of the pathogen to control the weed may be to use insect species as deliberate vectors. Gillespie & Menzies (1993) reported dissemination of *Fusarium oxysporum* f. sp. *radicis-lycopersici* by adult fungus gnats. Recently, corn earthworm moths, *Helicoverpa zea*, have been shown to transmit sorghum ergot, *Claviceps africana*, from diseased to healthy plants (Prom et al., 2003). As *F. tumidum* produces only macroconidia, insects that naturally carry large fungal spores on their cuticle may have greater capacity to vector the spores of this pathogen. This study aimed to determine the potential of four adult insect species (known to visit gorse plants) to vector *F. tumidum* spores.

## Materials and methods

### Natural *Fusarium* spp. on the cuticle of four insect species

Gorse seed weevil *Exapion ulicis* (Förster), gorse pod moth *Cydia succedana* (Denis & Schiffermüller), gorse thrips *Sericothrips staphylinus* Haliday, and light brown apple moth *Epiphyas postvittana* (Walker) collected from the field, were used for this study. Two techniques (washing and direct plating on agar) were used to remove fungi on the insects' cuticle; these fungi were then cultured on Potato Dextrose Agar. Representative fungal colonies were subcultured on to Potato Carrot Agar and Hay Agar (19–23°C) for identification. The mean fungal spore size from each insect species was determined using the Soft-imaging System programme, analySIS<sup>®</sup>, which measured the vertically projected area of 45–60 spores from each of 15 distinct fungal groups.

### Transmission of *Fusarium tumidum* spores by insect species

Live insects (50–60) of each species (from the field except *E. postvittana* which was laboratory-reared) were placed on cornmeal-glucose agar (CMGA) cultures of *F. tumidum* spores for 24 h and then transferred to sterile containers. At each time point (0, 24, 48 and 72 h after exposure), three replicates of five insects of each species (three of the moths) were washed in 1 ml of potassium phosphate buffer and a 10-fold dilution series prepared. Samples (0.1 ml) were plated on two replicate CMGA plates and incubated for 7 days under near UV and white light at 19–23°C. The number of *F. tumidum* spores present on individual insects of each species was determined. Data for total *F. tumidum* colony counts were log transformed and analysed by analysis of variance. In another experiment, *E. ulicis* and *S. staphylinus* were exposed to the spores for only 1 h and then washed and assessed as previously described. The mean body length of each insect species was measured.

## Results and discussion

Both *C. succedana* and *E. postvittana* were found to carry *Fusarium* spp. naturally. Only *E. postvittana* naturally carried *F. tumidum* spores and also carried the largest fungal spores with

mean vertically projected area of  $123.4 \mu\text{m}^2$ . Other *Fusarium* spp. carried by *E. postvittana* were *F. poae*, *F. moniliforme*, and *F. dimerum*. *Cydia succedana* carried only *F. moniliforme*. Following exposure to the spores, *F. tumidum* spores were recovered from all the insects. Recovery of *F. tumidum* decreased with increasing time after exposure (Figures 1 & 2) but remained generally constant for the first 24 h when insects were exposed for 24 h. Loss of spore viability or dislodgment from the insects may account for this decline. The spore viability was 60%. *Epiphyas postvittana* carried the most *F. tumidum* spores (59 spores insect<sup>-1</sup>) ( $P < 0.01$ ) followed by *C. succedana* and *E. ulicis* with 12 and 6 spores insect<sup>-1</sup>, respectively. *Sericothrips staphylinus* carried the least spores (1 spore insect<sup>-1</sup>), which was recovered from insects after 48 h. There was a strong correlation ( $R^2 = 0.83$ ) between the body length of insect species and log<sub>10</sub> of the number of *F. tumidum* spores carried. *Epiphyas postvittana* was considered to have the greatest potential for vectoring *F. tumidum* spores.

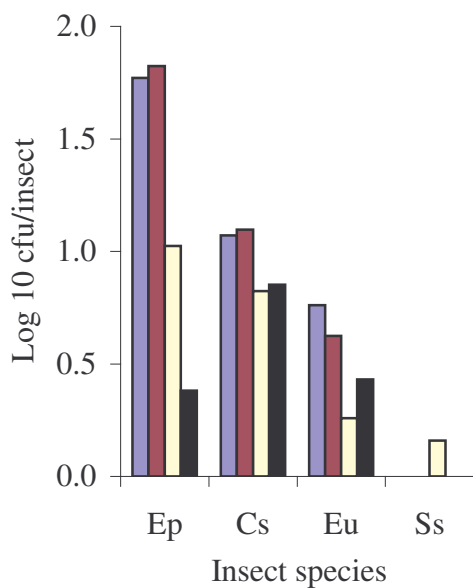


Figure 1. Cfu of *F. tumidum* recovered from insects exposed to *Fusarium* spores for 24 h.

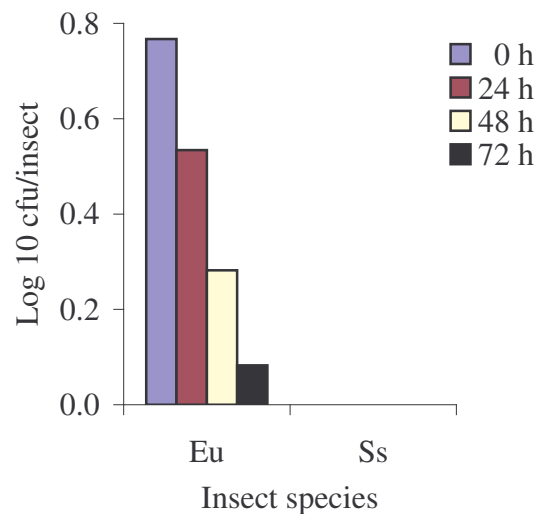


Figure 2. Cfu of *F. tumidum* recovered from *E. ulicis* and *S. staphylinus* exposed to *Fusarium* spores for 1 h.

Ep: *Epiphyas postvittana*; Cs: *Cydia succedana*; Eu: *Exapion ulicis*; Ss: *Sericothrips staphylinus*

The duration of *E. ulicis* and *S. staphylinus* exposure to the spores appeared to have no effect on the number of *F. tumidum* spores carried. *Exapion ulicis* and *S. staphylinus* exposed to the spores for 24 or 1 h carried 6 and no spores insect<sup>-1</sup>, respectively, when washed immediately after exposure (Figures 1 & 2). Pheromone lures will be used to attract the moths to inoculum stations for more effective loading and thereafter infect the weed (“lure-load-infect”). However, more work is needed to develop this concept.

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## The genus *Ascochyta*, a source of toxic metabolites with potential herbicidal activity

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In the last few years, the possibility of using microorganisms as biocontrol agents and also compounds of natural origin, such as fungal metabolites, for weed control, has gained increasing interest since, compared to traditional chemical products, these appear to be more environmentally friendly and safe.

In the last decade, the research carried out by our group has focused on the production of toxins by species of the genus *Ascochyta*, responsible for several diseases, usually appearing as necrotic lesions on leaves and stems, of important crops. We identified a wide range of metabolites having different chemical structures and biological properties. First, cytochalasins A and B were isolated from *A. heteromorpha*, a pathogen of oleander. Several new cytochalasins, some of them with unusual structures, were successively identified from both the liquid and solid cultures of the fungus (Vurro et al., 1997). Later, some other phytotoxins were obtained from other species of *Ascochyta*, such as ascosalitoxin and pinolidoxins, isolated respectively from cultures of *A. pisi* (Evidente et al., 1993a) and *A. pinodes* (Evidente et al., 1993b). Both these fungi are responsible for pea anthracnose.

Considering the interesting phytotoxic metabolites produced by species of the genus *Ascochyta*, and considering that some *Ascochyta* species have been proposed as mycoherbicides for the biological control of very dangerous and widely diffused weeds, i.e. *A. caulina* against *Chenopodium album* (Netland et al., 2001), *A. cypericola* against *Cyperus rotundus* (Upadhyay et al., 1991), and *A. sonchii* against *Sonchus oleraceus* and *Cirsium arvense* (Berestetski & Smolyaninova, 1998), it was of interest to study the metabolites produced by those pathogens, and their possible use as natural herbicides.

From the culture filtrates of *A. caulina* growing in static conditions, three main toxic metabolites were isolated and chemically and biologically characterized: *trans*-4-amino-D-proline (Evidente et al., 2000); ascaulitoxin, a new glucopyranoside of an unusual nonproteogenic *bis*-amino acid (Evidente et al., 1998); and its aglycone (Evidente et al., 2001).

Assayed on detached leaves of the host plant, all the three pure compounds showed strong phytotoxicity, with necrosis resembling those caused by the pathogen. Assayed on other plant species, ascaulitoxin has shown a good activity on a wide array of plant species (Evidente et al., 1998), whereas *trans*-4-aminoproline is ineffective on monocots (Evidente et al., 2000) and the aglycone was more active compared to ascaulitoxin (Evidente et al., 2001).

Many greenhouse and semi-field experiments were carried out on young *C. album* plants by spraying suspensions containing different combinations of conidia, herbicides at reduced rates and an almost pure, highly water-soluble mixture of the three fungal metabolites. The latter was obtained using a simple and quick method of purification based on ion-exchange chromatography.

The application of this mixture caused strong toxic effects, appearing as leaf and stem necrosis and plant death, on *C. album* plants. This effect, not observable if applied to maize or sugar beet plants, was strongly enhanced and with additive or synergistic effects if the mixture was used with the conidia or the herbicides (metribuzin and rimsulfuron). The best



results were achieved with the toxins in combination with a reduced dose of metribuzin (at 1/5 of the labeled rates) causing more than 90% fresh weight reduction (Vurro et al., 2001).

More recently, another new phytotoxic metabolite, named ascosonchine, has been isolated from the culture filtrate of *A. sonchi*. This is a potential leaf mycoherbicide of *S. arvensis*, a perennial herbaceous weed widely distributed in the temperate regions of the world.

Ascasonchine is a new phytotoxic enol tautomer of 4-pyridylpyruvic acid. When assayed on punctured host leaves this caused wide necrotic circular lesions 2 days after application (Evidente, 2004). When assayed on other several weed and crop plants, both monocots and dicots, the toxin proved to have interesting selective herbicidal properties. It was very active, causing severe necrosis on many species such as *Euphorbia helioscopia*, *Salvia officinalis* or *Triticum* sp. However, it proved to be completely ineffective on all the solanaceous species assayed (tomato, eggplant, red pepper, potato) and was slightly active or almost inactive on leguminous (bean and chickpea) and cucurbitaceous (melon and zucchini) plants. Interestingly, ascosonchine proved to have almost only phytotoxic activity, being quite ineffective when assayed on *Geotricum candidum*, *Pseudomonas syringae* and *Lactobacillus plantarum* (Gram – and Gram + bacteria, respectively). No effects were observed in the brine shrimp assay (*Artemia salina* larvae), when tested at concentrations up to  $10^{-4}$  M.

A mixture containing around 1% of ascosonchine, obtained by cationic exchange chromatography, had toxicity similar to the pure toxin when assayed on host leaves, meaning that other metabolites could interact with ascosonchine in causing the toxic effects. Studies are in progress to identify those compounds. The production of the mixture of metabolites would be much easier than the purification of pure ascosonchine, and its higher relative toxicity compared to ascosonchine would make easier its practical use as natural herbicide.

Within an European project, many other strains of *A. sonchi* and other *Ascochyta* species will be considered for the production, purification and evaluation of toxic metabolites as natural herbicides.

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# Soil thermal – moisture regime of a *Triticum durum* – *Zea mays* rotation

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## Introduction

The temperature gradient (grad T) and the moisture gradient (grad W) are the main indicators for “non-isothermal distribution of moisture in soil. This research investigates a comparative analysis of thermo-moisture exchange in the soil profile of a rotational cropping sequence of *Zea mays* and *Triticum durum*. The crops were established on a Pelic Vertisol soil type and subjected to one of two treatments – weed free after herbicide application or weed infested throughout. Both treatments were fertilized with N<sub>7</sub> for the maize and N<sub>3</sub> for the durum wheat. Changes in the thermo-moisture regime of the soil layer monitored were registered in relation to the composition and quantity of weed infestation. The quantity valuation for defining these changes is made by the “moisture” thermo-gradient coefficient  $\delta$ , interconnected with grad W and grad T.

## Materials and methods

Herbicide applications to *durum* wheat consisted of chlorsulfuron (6 g a.i. ha<sup>-1</sup>) + isoproturon (203 g a.i. ha<sup>-1</sup>) and to maize – atrazine (79,2 g a.i. ha<sup>-1</sup>) + alachlor (185 g a.i. ha<sup>-1</sup>). The temperature gradient (grad T) in soil profile is defined by the measured out-going voltage of a battery of thermo-electric transducers put in a metal container at a depth of 0.7 m. Two containers with conductometric transducers located at fixed depths  $\Delta L = 0.1 - 0.2 - 0.3 - 0.4 - 0.5$  m were positioned in both herbicide treated and untreated crops. The electric resistance R measured with conductometric transducers in different soil layers is an information indicator for definition of grad W (Wiebe, 1971, Hancock 1954), according to the equation:

$$\text{grad}W = -\frac{\Delta W}{\Delta L} = -\frac{\lg[(R + \Delta R)/R]}{b \cdot \Delta L}, \text{ where the coefficient } b = 0.45 \text{ has been defined}$$

experimentally for the soil type (Pelic Vertisol). The relationship between grad W and grad T defines the “moisture” thermo-gradient coefficient  $\delta$ , as follows:

$$\delta(W, T) = -(\text{grad}W/\text{grad}T) \text{ (Globus, 1983).}$$

The coefficient  $\delta$  appears as an integral characteristic for effective exchange at various thermal-moisture regimes in the soil profile. If this condition is formally accepted, then, with some exceptions, it could be said that  $\delta$  values differ little from the “absolute” -  $\delta = f(W)_{T=\text{const}}$  and  $\delta = f(T)_{W=\text{const}}$ .

## Results and discussion

Table 1 shows the measured values for grad T, grad W and the calculated values of  $\delta$  at various depths for both crops (in rotation) at different growth stages. The  $\delta$  high values at low and high moisture in the soil profile indicate an almost isothermal regime. This is an indication of the formation layer of resistance which on the one side changes the effectiveness of thermo-moisture exchange and on the other prevents the reverse flow of dissolved mineral salts to the zone of higher temperature, i.e. the opposite zone to which moisture has accumulated. In maize, after growth stage “3-4 leaf”, the heavy weed infestation in the herbicide free (323 nm. m<sup>-2</sup> during “9-10 leaf” compared to 93 nm. m<sup>-2</sup> in herbicide treated crop resulted in differences in the thermogradient coefficient and conforms to the layer of resistance in the experimental profile. The correlation in  $\delta$  values between treatments at the 3-4 leaf growth stage is 1: 4, and at the 9-10 leaf growth stage it increases to 1:10. The maize yield was reduced 6 fold with weed infestation (Table 1).

For durum wheat there were significant differences in  $\delta$  values between treatments, especially in layer 0.2 – 0.3 m from “flowering” phase to “full ripeness” phase (Table 1). Here the level of weed infestation was lower – 23.5 nm. m<sup>-2</sup> in the herbicide treated and 33 nm. m<sup>-2</sup> in the weed infested at “tillering”. The difference in durum growth between treatments was negligible – 5 kg da<sup>-1</sup> (Table 1).

Table 1 Values of the indicators for thermal-moisture regime in maize – durum (in rotation) at various crop growth stages at different levels of weed infestation

Phases of maize development (2001)									
Soil depth [m]	Variants	3-4 <sup>th</sup> leaf grad T=0,28 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]		9-10 <sup>th</sup> leaf grad T=0,39 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]		14 <sup>th</sup> leaf grad T=0,3 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]		Silking grad T=0,28 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]	
		-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>	-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>	-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>	-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>
0,1 - 0,2	+weed inf.	0,26	0,093	0,45	0,115	0,1	0,033	2,44	0,871
	+herb.	0,24	0,086	0,04	0,01	0,1	0,033	1,57	0,561
0,2 - 0,3	+weed inf.	0,12	0,043	1,53	0,392	0,54	0,18	3,13	1,118
	+herb.	0,08	0,028	0,18	0,046	0,34	0,113	3,09	1,104
0,3 - 0,4	+weed inf.	0,86	0,307	0,56	0,144	0,06	0,02	1,86	0,664
	+herb.	0,58	0,207	0,15	0,038	0,06	0,02	0,5	0,178
0,4 - 0,5	+weed inf.	0,18	0,064	1,83	0,469	2,0	0,667	1,88	0,671
	+herb.	0,11	0,039	0,13	0,033	1,56	0,52	0,8	0,286
Yield [kg da <sup>-1</sup> ]: 95,8 (infestation); 615,8 (with herbicides)									
Phases of durum wheat development (2002)									
Soil depth [m]	Variants	Tillering grad T=0,7 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]		Flowering grad T=0,42 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]		Milky ripeness grad T=0,1 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]		Full ripeness grad T=0,1 [°C . 10 <sup>2</sup> m <sup>-1</sup> ]	
		-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>	-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>	-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>	-grad W %.10 m <sup>-1</sup>	- $\delta$ %.°C <sup>-1</sup>
0,1 - 0,2	+weed inf.	0,68	0,097	0,67	0,160	0,65	0,65	0,64	0,64
	+herb.	0,1	0,014	0,2	0,048	0,65	0,65	0,15	0,15
0,2 - 0,3	+weed inf.	0,5	0,071	3,32	0,79	2,34	2,34	1,78	1,78
	+herb.	0,48	0,068	0,32	0,076	0,63	0,63	0,38	0,38
Yield [kg da <sup>-1</sup> ]: 234,25 (infestation); 239,28 (with herbicides)									

The presence of a “moisture” thermo-gradient coefficient in the soil profile  $\delta$  is a factor, influencing the movement of moisture consisting not only of water but also dissolved particles of the major plant nutrients. This movement results in a change of moisture relations and subsequent re-distribution of water and mineral salts in the root layer.

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# Effects of set-aside management on weed flora and productivity of subsequent wheat

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## Introduction

Rotational set-aside represents an opportunity to control weeds within a crop succession (Clarke, 1992, Rodriguez and Mamarot, 1995). For this scope, the efficacy of the required cultural operations must be thoroughly evaluated. Basically, set-aside field can be covered by spontaneous vegetation (undisturbed fallow) or by seeded plants (cover crop), in this instance an N residue is expected from leguminous species. Otherwise plant emergence in fallow can be prevented by cultural or chemical means. The costs of these managements should be compared with a possible benefit on the succeeding crops in terms of low weed infestation and higher yield.

## Materials and methods

The research was conducted from 1993 to 1997 in a hill farm of Bologna University at Ozzano (Bologna) on clay soils. In different years wheat (*Triticum durum* Desf.), sugarbeet (*Beta vulgaris* L.), maize (*Zea mays* L.), and sunflower (*Helianthus annuus* L.) crops were followed by set-aside. In each set-aside these managements were compared: undisturbed fallow, red clover (*Trifolium pratense* L.) cover crop, and bare fallow obtained by chemical treatments or mechanical operations. Experimental plots were 350 or 500 m<sup>2</sup> of area and were arranged according to randomized block designs, with 4 replications. Clover was seeded in early spring and ploughed under in the autumn. Chemical treatments were spring application of simazine+glyphosate, followed by a summer application of glyphosate or dicamba+2,4D+MCPA. Cultural operations consisted of 2-3 harrowing or mowing passes during spring and summer. Set-aside was followed by winter wheat crops in which plots were divided into four subplots. On them the following treatments were compared factorially: with and without herbicide (a spring application of diclofop+ioxynil+bromoxynil), low and high N fertilization levels (75 and 150 kg N/ha with urea). Weed cover in fallow and in the succeeding wheat was monitored by 2-3 Braun Blanquet relevées. Wheat yield was determined. In the last experiment weed seedbank was assessed by seed count in soil samples. They were collected to 25 cm depth in the autumn after set aside and in the chemically untreated, low fertilized plots of the following wheat.

## Results and discussion

On the overall average of the four crops and years (the individual effects of which cannot be separated), fallow management resulted in significantly different weed abundance. The vegetation in undisturbed fallow was denser (153±7% cover) than in seeded set-aside (104±3%). Chemical treatments (93±6% cover) were less effective than cultural means (68±5%) in preventing weed presence, mainly due to the resistance or prolonged emergence of some species. In wheat after undisturbed fallow a heavy weed infestation was observed, which was difficult to check even by effective herbicides (Figure 1a). An abundant infestation was also recorded after the cover crop, but it was adequately controlled by chemicals. Bare fallow, obtained both chemically or mechanically, resulted in a low infestation in the following wheat, which was further reduced by herbicide distribution in the cereal. As a consequence, on average of the four experiments (Figure 1b), wheat yield was significantly

lower when the preceding set-aside was left undisturbed. Satisfactory grain yields were obtained after clover, even without herbicides, testifying that the observed high infestation comprised of less competitive weeds. There was little effect of fallow on wheat nitrogen requirement. Adding fertilizer N raised grain yield ( $4.06 \pm 0.12$  t/ha and  $4.90 \pm 0.14$  t/ha for N1 and N2, respectively), but the increase was not influenced by the preceding leguminous species. Seedbank after undisturbed fallow was significantly greater than after bare or seeded set-aside (Table 1), as reported elsewhere (Connolly and Naylor, 1996) but decreased in the non-herbicide treated plots of the subsequent wheat. However, for wheat grown after clover or herbicide treatment, the seedbank almost doubled, but after cultivated fallow its increase was slight. In conclusion, undisturbed fallow appeared unadvisable because of the intense seed rain that can damage the following crop. However the consequences did not appear long lasting, probably because the seed rain is made up of species that can be easily controlled by conventional herbicides. A preceding leguminous cover crop prior to wheat proved as effective as the prevention of weed emergence in set-aside by chemical or mechanical means, but its higher costs were not compensated by a significant N residue benefit. In conclusion mechanical weed suppression appeared to be the best choice during set-aside, capable of maintaining a low weed seedbank at an acceptable level of costs.

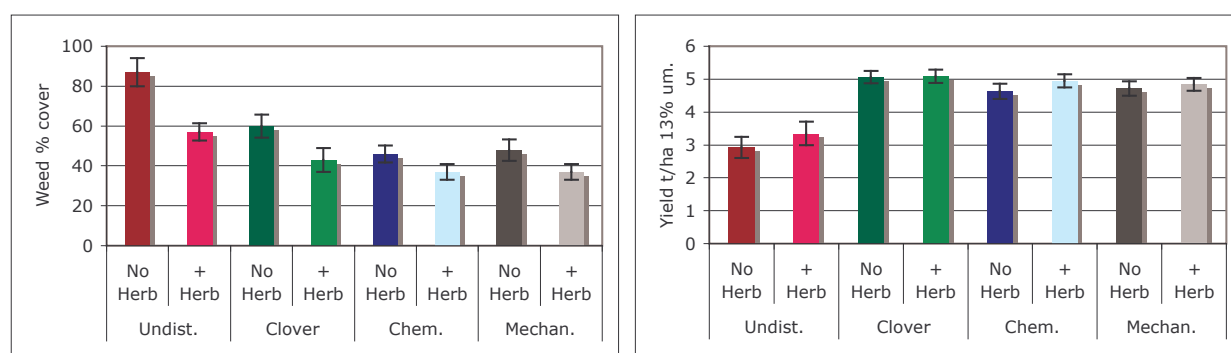


Figure 1. Effects of set-aside management (undisturbed fallow, clover cover crop, chemically and mechanically treated set-aside) on weed infestation and grain yield of the succeeding wheat crop with or without herbicides ( $\pm$  standard errors,  $n=32$ ).

Table 1. Seedbank assessed at the end of fallow and after the succeeding wheat.

Set-aside management	Million seed $ha^{-1}$ ( $\pm$ standard error, $n=4$ )	
	After set aside	After succeeding wheat
Undisturbed fallow	$117 \pm 15$	$102 \pm 17$
Clover cover crop	$45 \pm 15$	$81 \pm 18$
Chemically treated set-aside	$38 \pm 10$	$79 \pm 20$
Mechanical set-aside	$53 \pm 23$	$61 \pm 11$

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# Impact of different soil tillage systems on weed control and the evolution of weed seedbank in a long term field crop trial

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## Introduction

Soil tillage has always been intensified with the developing agricultural machinery. To face the growing problems of intensive farming like erosion, monoculture and others, Integrated Crop Protection systems including reduced soil tillage have been established in the eighties. The Agroscope RAC Changins research station at Nyon, Switzerland started a long-term trial in 1969 to compare ploughing and various no-till systems. Observations of the weed seedbank started in 1989, with the objective to compare the evolution of the weed flora under different soil tillage systems. The analysis of the weed seedbank is an effective tool to the studies of the evolution of weed flora in relation with cultural practices.

## Materials and methods

The trial was started in 1969 to compare conventional ploughing with systems of reduced soil tillage. A crop rotation of winter wheat, winter oil seed rape, winter wheat and maize was grown on a loamy soil and on a clay humic soil. Three ploughless techniques were compared to conventional mouldboard ploughing: deep cultivation with a chisel-plough (20-30 cm depth), shallow cultivation with a cultivator (10-15 cm) and minimum tillage with a rotary harrow (5-7 cm); instead of minimum tillage, direct drilling (cereals) and strip tillage (maize) were occasionally performed.

The development of the weed seedbank was observed from 1989. Soil sampling was made every 2 years in the spring with a cylinder of 5 cm diameter and ~20 cm deep; 50 cores were taken from each treatment plot of 160 m<sup>2</sup> and there was one replicate for each treatment. After sampling, the soil was stored at 4 °C in the darkness. After, the seeds were washed out with water and sieved through a 4 mm and a 0.25 mm sieves. Weed seeds were germinated in sterile soil for 4-5 weeks under controlled conditions (Beuret, 1989). Seedlings were identified and counted.

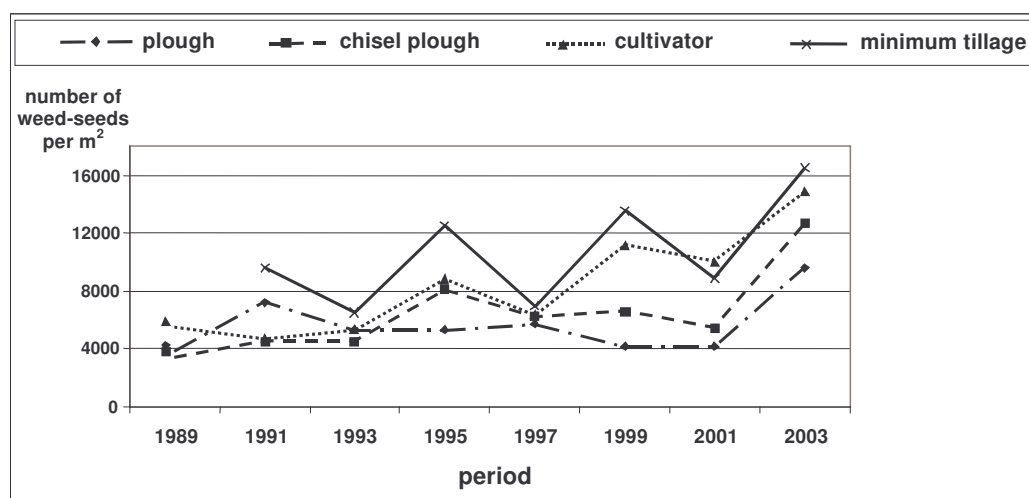


Figure 1. Evolution of the weed seedbank following different soil tillage methods from 1989 to 2003

## Results and discussion

At the time observations were started in 1989, the weed seedbank was the result of the trial started in 1969 on that field. Figure 1 shows a slight increase in the weed seed number/m<sup>2</sup> in the conventional plough system during the observed period. Globally the lower the intensity of soil tillage the higher the number of weed seeds observed. Moreover, the variation during the period in number of weed seeds/m<sup>2</sup> is also wider in the minimum tillage system. These results underline the “herbicide effect” of deep and intensive soil tillage.

More than 50 weed species were identified (Mayor & Maillard, 1995). The main weed species in all plots during the trial period were *Brassica napus/Sinapis arvensis* (~25 % ground cover), *Myosotis arvensis* (~25%), *Capsella bursa pastoris* (~23%), and *Viola arvensis* (10%). *Brassica napus/S. arvensis* and *V. arvensis* disappeared partially in “cultivator” and “minimum tillage” plots in favor of *M. arvensis* (~36%) and *C. bursa pastoris* (~34%). Grass-weeds did not become a problem in minimum tillage (~2%). An increase of perennials in ploughless tillage like bindweed and thistles has been observed but was not measured.

Table 1. Mean number of 34 years of interventions for weed control, their relative costs, and the means of total of passes for soil tillage and weed control during the trial period from 1969 to 2003 in different soil types (Vulliod & Mercier, 2004).

tillage	hoeing	selective herbicide	non-selective herbicide	total weed control	relative costs of weed control	total passes incl. tillage
loamy soil						
<b>plough</b>	0.06	1.5	0.2	1.8	100	6.9
<b>chisel plough</b>	0.06	1.5	0.3	1.9	105	7.2
<b>cultivator</b>	0.06	1.5	0.4	2.0	108	6.7
<b>minimum tillage</b>	0.09	1.6	0.6	2.3	122	4.5
clay humic soil						
<b>plough</b>	0.09	1.6	0.2	1.9	100	6.9
<b>chisel plough</b>	0.09	1.6	0.3	2.0	105	7.1
<b>cultivator</b>	0.09	1.6	0.4	2.0	106	6.7
<b>minimum tillage</b>	0.09	1.6	0.6	2.3	119	4.4

Decreasing the intensity of soil tillage leads (i) to increased use of non-selective herbicides, (ii) to increased costs for weed control, and (iii) to massive reduction of total production costs due to reduced soil tillage (Table 1). Analysis of the weed seedbank during a 14 year period confirms a higher weed seed number when soil tillage is minimised. Nevertheless, weed control methods did always perform correctly, in all plots of the trial, and during the full period of the trial.

The “herbicide effect” of tillage is replaced by the more frequent use of glyphosate. Instead of controlling mainly young stages of annual weeds after conventional ploughing, more perennials and older annuals have to be controlled in ploughless and no-till systems. This additional use of glyphosate bears the risk of introducing resistance.

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# A framework model for the population dynamics of weeds in field vegetable systems

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## Introduction

The current trend in weed management is to understand the population dynamics of weeds and their role in the agro-ecosystem (Cousens & Mortimer, 1995; Buhler, 1999). Stricter approval conditions related to health safety reasons and the increased development costs of herbicides have led the agrochemical companies to concentrate their search on compounds which are effective for a few major crops. This is leaving serious gaps in weed control for growers of more specialist crops. In most horticultural production systems the low weed thresholds necessary to achieve crop quality demands are an additional problem.

Limited options for weed control may lead to the extended use of a particular strategy and this will increase the likelihood of a tolerant weed flora developing. However, gradual changes due to natural variation in the susceptibility of a population to a weed control method may only be detectable after a number of seasons. Modeling approaches combined with experimental verification provide a realistic approach to understanding the complexity of the processes involved. Individual phases of the life cycle of weeds (Figure 1) have been described by a number of mathematical / statistical models produced in recent years (Table 1).

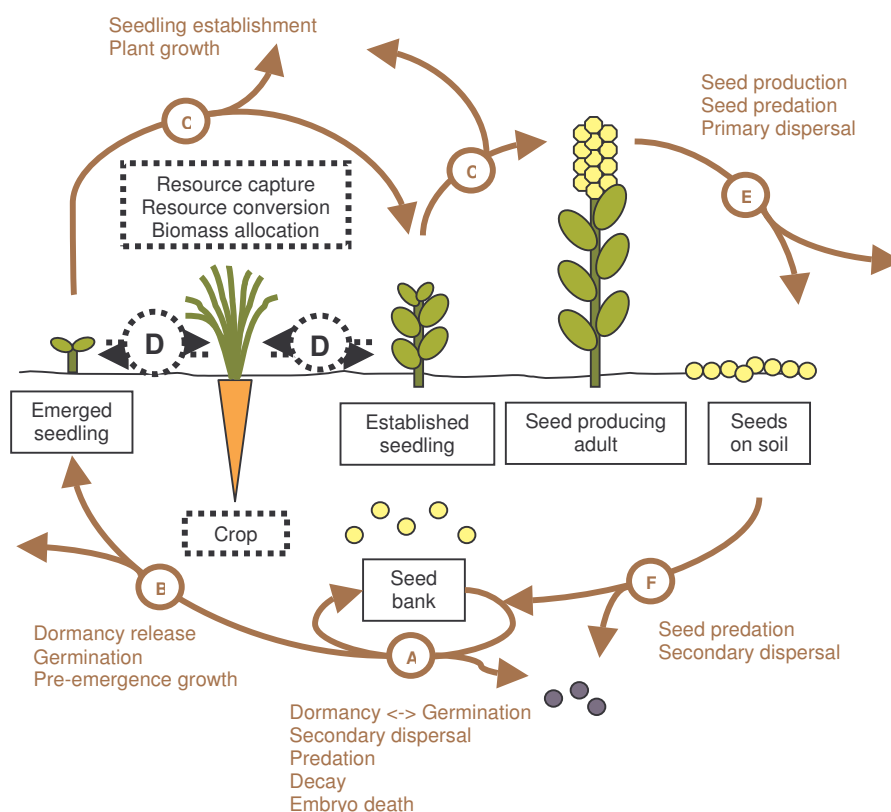


Figure 1. Life cycle diagram for annual weed, capital letters refer to key processes and are further explained in Table 1 (modified after (Gallandt *et al.*, 1999))

Table 1. Current models for the key processes in the life cycle of an annual weed species.

	<b>Key processes</b>	<b>Available (component) models</b>
A.	Seedbank persistence	Vitta <i>et al.</i> , 1989; Grundy <i>et al.</i> , 1999; Colbach <i>et al.</i> , 2000
B.	Emergence	Grundy <i>et al.</i> , 1996; Vleeshouwers, 1997; Grundy & Mead, 2000; Vleesh. & Bouwmeester, 2001; Grundy <i>et al.</i> , 2003
C.	Establishment	Being developed as part of current project
D.	Competition	Acker <i>et al.</i> , 1997; Park <i>et al.</i> , 2001
E.	Plant reproduction	Lutman, 2002; Rasmussen & Holst, 2003
F.	Seed return	Extensive literature review should provide mechanisms and parameters which will then be implemented in simple model.

However, very few studies have attempted to combine these models within a dynamic framework covering the entire life-cycle. Such a framework model would allow us to simulate the outcome of a range of weed control strategies on the long-term composition of the weed seedbank. Further, sensitivity analysis of the framework model will point out the ‘weakest links’ within a weeds lifecycle enabling us to address optimal weed control strategies. Thus, the main aim of this Ph.D studentship is to develop a modeling framework through the integration of published models and additional experimentation where required. Initially the framework will be targeted at two problematic weed species, common chickweed (*Stellaria media*) and mayweed (*Tripleurospermum inodorum*). However, the generic understanding and principles within the framework will be applicable to other weed species as well.

### Materials and methods

- Review existing models and identify their constraints.
- Identify the availability and usefulness of long-term unpublished data collected at Warwick HRI as input for the framework model.
- Identify the life stages in the cycle where information is not sufficient or where models are not available.
- Set up field experiments, to obtain any additional information.
- Implement mathematical algorithms of component models into MATLAB.
- Ensure that MATLAB output and input of component models ‘fit’.
- Define scenarios and management strategies as identified by DEFRA
- Run simulations and perform sensitivity analysis

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# Dependence of weed flora on agrochemical soil properties formed by different manure rates in acid and limed soils

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## Introduction

Liming and application of organic fertilizers, which increase microbiological soil activity, play an important role in soil fertility maintenance (Perucci et al.,1997). The calcium, contained in lime and manure, neutralizes the mobile aluminium that is harmful to plants and decreases soil acidity. Continuous manure application improves other soil agrochemical properties such as phosphorus, potassium and humus content. However, some amount of weed seeds are incorporated into the soil with manure. Data of our investigations shows that 1 kg of cattle manure contains approximately 460 weed seeds (Ciuberkis,1996). Therefore, manure application increases not only weed seed stock in soil but may also change number and composition of weed species (Forcella et al.,1996). Thus, under the influence of antropogenic activity the stock of weed seeds in the soil as well as weed infestation of crop stands may change. It is important to consider it in planning weed control measures, particularly in sustainable or organic agriculture. There is a lack of information on weed flora changes obtained from long-term experiments.

The aim of the investigation was to determine the influence of different soil agrochemical properties formed under long term application of different manure rates in acid and limed soils on weed flora.

## Materials and methods

Two long-term field trials have being carried out since 1949. The experiments are being conducted on sandy loam acid ( $\text{pH}_{\text{KCl}}$  4.0-4.2) soil. They are located in the same field very close to each other. Soil of one field trial was unlimed, and soil of another one was limed by one rate according to hydrolytic soil acidity with pulverized limestone before the establishment of the experiment and later periodically after every 7th year (i.e. at the end of each crop rotation). The increasing rates of manure (20, 40, 80 and  $120 \text{ t ha}^{-1}$ ) were applied in both field experiments. Half rate of manure was incorporated into the winter wheat and the other half rate into the fodder beet. One treatment had no manure application. The size of the initial plot was 8.5 m by 6 m ( $51 \text{ m}^2$ ). Five replicates were used each experiment. The following crop rotation was applied: 1) winter wheat (*Triticum aestivum*), 2) spring barley (*Hordeum vulgare*), 3) spring oat (*Avena sativa*), 4) fodder beet (*Beta vulgaris*), 5) spring barley, 6) first year perennial grasses: red clover (*Trifolium pratense*) + timothy (*Phleum pratense*), 7) second year perennial grasses. Results of the last crop rotation are presented in this paper.

The weed flora was assessed in four quadrates (0.5 m x 0.5 m) per plot at the end of May – early June, i. e. before weeding of fodder beet and at tillering stage of cereals. Correlation analysis was applied to establish the dependence of the weed flora on soil agrochemical characteristics.

## Results and discussion

Liming and long-term usage of manure in acid soil contributed to the improvement of soil agrochemical properties that caused changes in the weed flora. The number of acidophilic weeds (*Spergula arvensis*, *Sclerantus anuus*, *Raphanus raphanistrum*, *Rumex acetosella*) was approximately 10 times less in crops grown in limed soil as compared with that in acid soil.



The number of nitrophilous weeds (*Chenopodium album*, *Capsella bursa-pastoris*, *Tripleurospermum perforatum*, *Stellaria media*) was greater by 40-50 % in crops cultivated on limed soil than on the acid soil only when manure was not applied or used at low rates. The number of these weeds was similar in crops on both soils when higher rates (80-120 t ha<sup>-1</sup>) of manure were applied. Using the average weed number data from the last crop rotation (7 years) applied on acid soil, a very strong negative and significant linear correlation was established between acidophilic weeds number and soil pH, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O and humus content (Table 1). Analogical correlation was also established between these weed numbers and soil agrochemical properties when liming was periodically applied. This means that continuous manure application has a positive influence on the improvement of soil properties and reduction of acidophilic weeds in acid and limed soil as well. Improved soil agrochemical properties influenced nitrophilous weeds in the opposite way.

Table 1. Correlation coefficient between acidophilic, nitrophilous and total weed number on agrochemical soil properties (\* P < 0.05, \*\* P < 0.01, n.s. P > 0.05 )

Soil chemical indicators	Acidophilic weeds	Nitrophilous weeds	Total weed number
	<i>r</i>	<i>r</i>	<i>r</i>
Acid soil			
pH	- 0.91*	0.93*	- 0.92*
P <sub>2</sub> O <sub>5</sub>	- 0.88*	0.95*	- 0.92*
K <sub>2</sub> O	- 0.93*	0.97**	- 0.95*
Humus	- 0.98**	0.98**	- 0.98**
Limed soil			
pH	- 0.97**	0.77*	0.41 n.s.
P <sub>2</sub> O <sub>5</sub>	- 0.99**	0.95*	0.68 n.s.
K <sub>2</sub> O	- 0.99**	0.92*	0.62 n.s.
Humus	- 0.97**	0.97**	0.72 n.s.

A positive significant dependence was found between nitrophilous weeds numbers on soil pH, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O and humus content in acid soil. A significant correlation was not established between these weed numbers and soil pH when liming was applied, because in this case pH<sub>KCl</sub> varied from 5.8 to 6.1. Improvement of other agrochemical properties of limed soil stimulated the spread of nitrophilous weeds. Positive linear correlation between these weed numbers and content of P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O and humus was very strong and significant. Improvement of soil agrochemical properties in acid soil played a positive role in the decrease in total weed numbers, due to reduction of acidophilic weeds. Very high and significant negative correlation were established between total numbers of weeds and the investigated soil properties when liming was not applied. No significant dependence was found for the total number of weeds on agrochemical properties in limed soil

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# **ALOMYSYS: a model of the effect of cropping systems on weed demography. Example of blackgrass (*Alopecurus myosuroides* Huds.)**

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## **Introduction**

Black-grass (*Alopecurus myosuroides* Huds.) is a common annual grass-weed in autumn-sown crops of Western Europe. It has become increasingly resistant to herbicides, but can be controlled by adapting cultural practices. Because of the large range of variations in cultivation techniques and their interactions, models are necessary to develop new cropping systems that reduce weed infestations while limiting herbicide applications. Such models must avoid "black box" relationships and describe the effects of cropping systems by mechanistic functions that distinguish the various underlying biological processes (*e.g.* seed germination), in interaction with environmental conditions (*e.g.* soil structure). A model quantifying the effect of cropping systems on seedbank survival, germination and emergence, in interaction with climate and soil structure, has already been developed (Colbach *et al.*, 2005a,b; Colbach *et al.*, 2005). The objective of the present paper was to complete the annual life-cycle with further sub-models quantifying the effects of cropping systems on tillering, flowering and seed production.

## **Materials and methods**

Fig. 1 describes the evolution from the seedbank to the newly produced seeds in ALOMYSYS (*Alopecurus myosuroides* =f(cropping **system**)). Literature data was used for winter seedling mortality (Barralis, 1970), leaf and tiller emergence model (Chauvel *et al.*, 2002) and seed production after cutting in set-aside (Dalbiès-Dulout & Doré, 2001). Greenhouse studies were carried out to model tillering in optimal conditions. In addition, field trials were set up during several years to quantify the effects of emergence date, nitrogen availability, crop competition and intra-specific competition. These results were used to develop the remaining sub-models, and then added to the already existing "seed bank  $\Leftrightarrow$  seedlings" sub-model.

## **Results and discussion**

The various equations used to build the model are synthesized in Fig. 1. Figure 2 compares for instance combinations of high- (only winter crops) and low-risk rotations (mainly spring crops) with intensive, low-input (little nitrogen, herbicides and tillage) black-grass-minimising management strategies (frequent summer tillage, mouldboard ploughing, high sowing densities, new herbicides). These simulations show that ALOMYSYS can be used to compare existing cropping systems and to evaluate changes in farming practices in order to design new cropping systems that control weeds with the minimum of herbicides while still assuring a satisfactory income to farmers.

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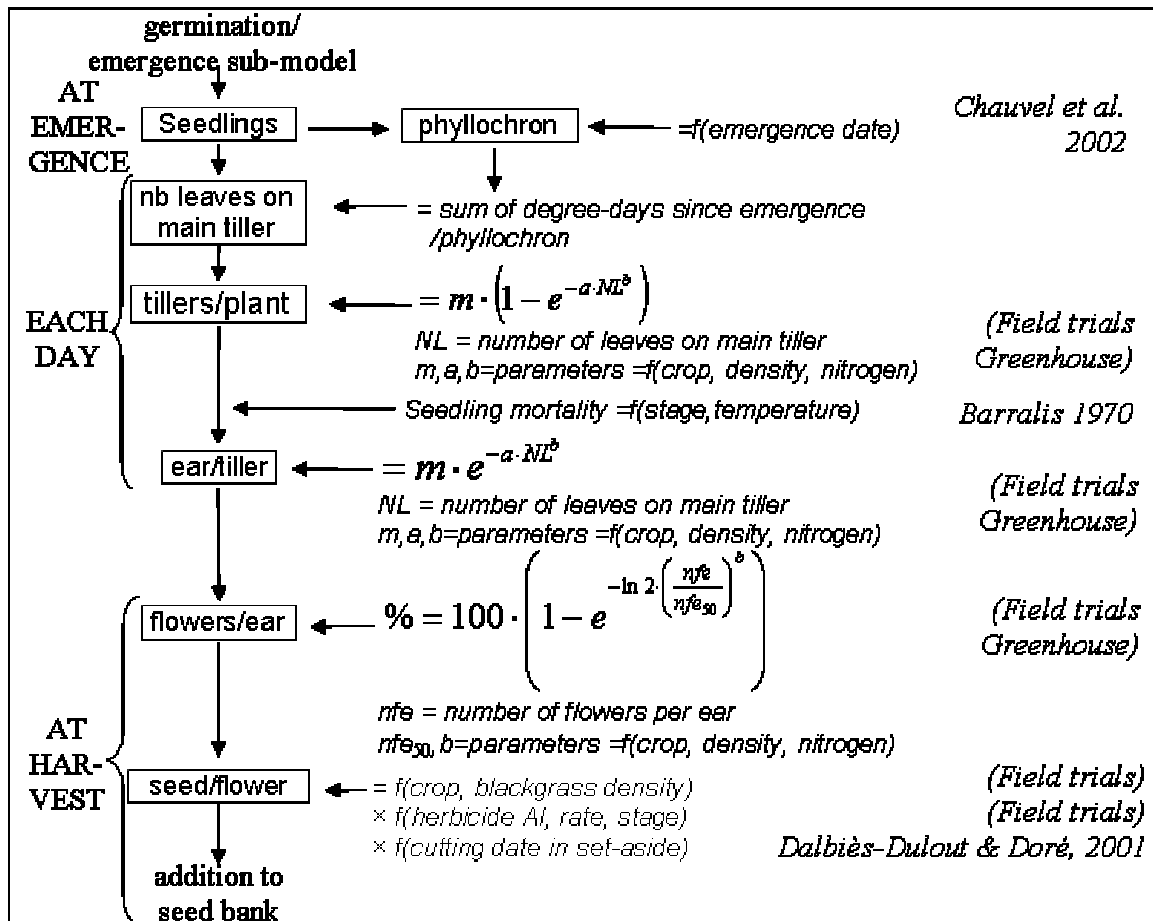


Figure 1: Organization of the seedling → seed production sub-model of ALOMYSYS

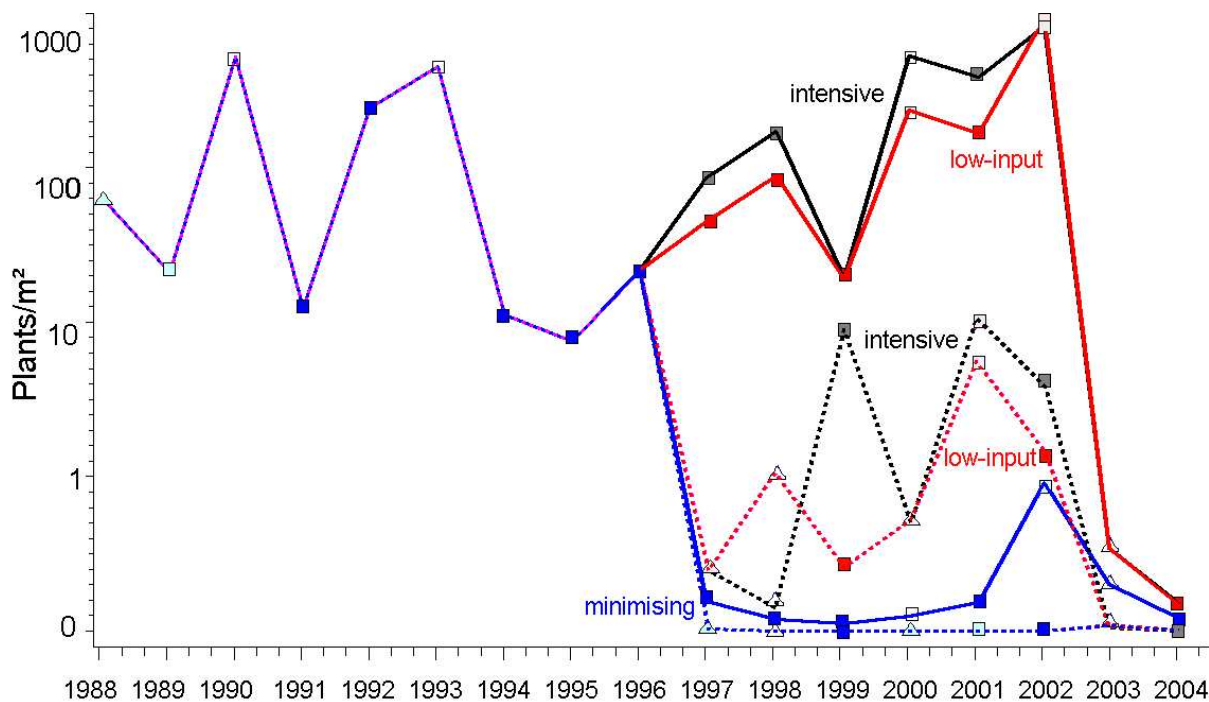


Figure 2. Blackgrass density at harvest simulated with ALOMYSYS. Testing of two rotations (winter crops only —; mostly spring crops ---) and three strategies (intensive, low-input, minimising blackgrass) after an initial risky system (1998-1996). Symbols represent crops (winter wheat ■; winter barley □; winter oilseed rape □; spring barley △; spring pea △).

## Induction of seed germination in *Orobanche aegyptiaca* to various seed stimulants

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### Introduction

The root parasitic weeds, which belong to the genus of *Orobanche*, are serious pests in Greece, especially in arable crops such as tobacco, tomato and faba beans. An essential feature is that *Orobanche* seeds only germinate when exposed to a specific chemical signal, i.e. a germination stimulant, usually exuded by the roots of the host plants (Bouwmeester *et al.*, 2003). It has long been realized that if a germination stimulant is applied to the soil in a suitable form before the host crop is planted, the parasitic seeds would germinate in the absence of a host and the germinated seeds would not survive, since the necessary attachment to the host plant is not possible. This method is commonly referred to as “suicide germination” approach. The natural germination stimulants do not show any ecological side-effects, they are active at very low concentrations, easily decompose in the soil and they show no toxicity to micro-organisms.

### Materials and methods

Germination responses of four populations of *O. aegyptiaca* to two germination stimulants GR<sub>24</sub> and a new natural product (NNP) were investigated at doses of 3ppm and 0.3125 v/v respectively. In addition, response to three growth regulators, IAA, Kinetin and GA<sub>3</sub> were investigated at the following concentrations: IAA 1 mg L<sup>-1</sup>, Kinetin 10 mg L<sup>-1</sup>, GA<sub>3</sub> 25 mg L<sup>-1</sup> (Batchvarova *et al.*, 1999). Firstly, the seeds were surface sterilized by immersion in sodium hypochlorite (3% v/v) for 3 minutes and then rinsed five times for 5 minutes with sterile distilled water before being subjected to the various treatments. The seeds were then spread in Petri dishes on filter paper moistened with 3 ml of water and pre-conditioned at 21°C for 9 days. After this pre-treatment four replicate dishes of each treatment were incubated at 25°C.

### Results and Discussion

The data showed that NNP had similar behaviour to that of GR<sub>24</sub>, as the analysis of variance between the two treatments showed differences not statistically significant (F- value:5.13, *p*: 0.0102). It is noticeable, that in one instance NNP showed increased effectiveness (Fig.1). Germination of *Orobanche* seeds ranged from 43 to 99% after NNP application and from 30 to 99% after GR<sub>24</sub> treatment. Populations showed high variability in their germinability with differences statistically significant (F- value: 79.83, *p*: 0.000). Populations A2 and A3 showed lower values of germinability relative to A1 and A4 (Fig. 1). Germination of populations A1, A3 and A4 were stimulated to a significantly greater extent by treatment with GR<sub>24</sub> whereas that of A2 was significantly greater following treatment with NNP. Treatment with the growth regulators IAA, Kinetin and GA<sub>3</sub> was less effective. Analysis of variance among the growth regulators showed differences not statistically significant (F-value: 4.13 *p*: 0.0522). However, Kinetin appeared to be the better seed stimulant relative to IAA and GA<sub>3</sub>.with mean values of germinability 28, 23 and 28 %, respectively. All treatments referred to above were followed by the use of water as a control treatment and the results obtained indicated zero germinability in all cases. These studies suggest that NNP may have seed stimulant activity comparable with that of the synthetic gibberellin analogue GR<sub>24</sub> of encouragement for field trials.

## Acknowledgements

We would like to thank Prof. Klaus Wegmann, University of Tübingen, Tübingen W. Germany, for providing us with GR<sub>24</sub> and with useful advice for the proper conduction of the experiments.

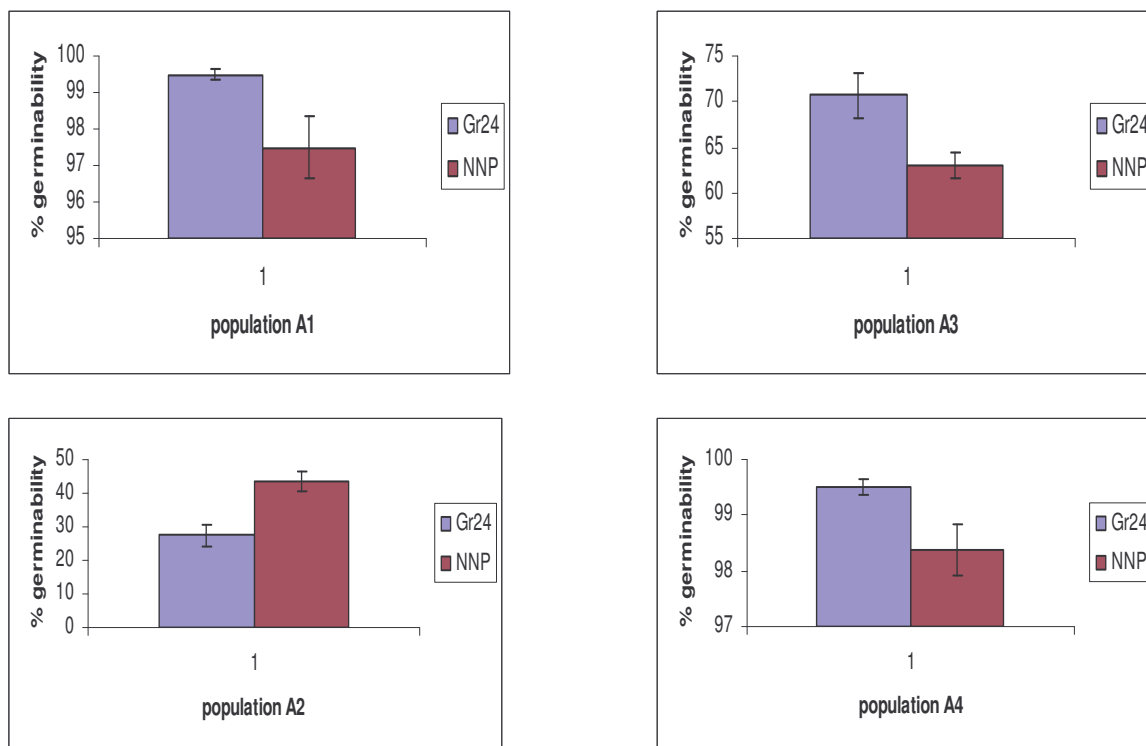


Fig. 1. Effect of the seed germination stimulants (GR<sub>24</sub> and NNP) on germination of four populations (A1, A2, A3, A4) of *Orobanche aegyptiaca*.

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# Relationship between crop rotation, herbicides rotation and soil weed seedbank and number of emerged weed species

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## Introduction

A long-term experiment was established in 1970 on a white luvic soil to investigate the relationship between crop and herbicide rotation on the soil seedbank and seedling emergence. Crop rotation consisted of maize, potatoes and winter wheat with four replications.

## Materials and methods

The weed seedbank in soil was assessed 20 years after initiation of the experiment before sowing of maize. The influence of herbicide treatments in maize and potato crops on the soil weed seedbank are presented for untreated plots of winter wheat crop.

### *Treatments in potato crop*

- 1.Untreated (manual and mechanical weed control)
- 2.Prometryn+alachlor (2+2 l.kg/ha)
- 3.Prometryn+fluazifop-butyl (2+0.375 kg,l/ha)
- 4.Metribuzin+alachlor (1.05+2.1 kg,l/ha)
- 5.Metribuzin+fluazifop-butyl (0.7+0.375 kg,l/ha)

### *Treatments in maize crop*

- 6.Untreated (manual and mechanical weed control) - without herbicides
- 7.Atrazine+cyanazine(1.25+1.25 kg/ha] treatment in rows
- 8.Butylate+2,4-D+dicamba (3.5+0.56+0.1 l/kg/ha)]
- 9.Butylate+atrazine (2.1+1.5 l kg/ha)]

### *Treatments in wheat*

- 1.Untreated
- 2.Fluroxypyr+clorsulfuron (0.175+0.0056 l,kg/ha)

## Results and discussion

The rotational use of herbicides in maize crop influenced the size and composition of the weed seedbank and seedling emergence of *Chenopodium album*.

The size of weed seedbank and number of seedlings emerged was least in the herbicide rotation which included atrazine+butylate in maize and with alachlor+prometryn in potato, but the greatest infestation was obtained following treatment with butylate +2,4-D +dicamba in maize crops, and with prometryn+fluazifop+butyl in potato. (Fig. 1.)

The coefficient of multiple correlations between weed seedbank and seedling emergence of *Chenopodium album* was highly significant.

Treatments with atrazine in maize crops, irrespective of herbicide treatments in potato, reduced the weed seedbank and seedling emergence of *Matricaria inodora*.

Relationships between the soil weed seedbank and *Matricaria inodora* plants, explained through the coefficient of multiple correlations were highly significant.

Infestation with *Amaranthus retroflexus* was greatest for the rotation in which maize was treated with butylate+atrazine, and in potato crop treated with prometryn or metribuzin plus fluazifop-butyl. Conversely, prometryn or metribuzin with alachlor reduced infestation with *Amaranthus retroflexus* to the greatest extent.

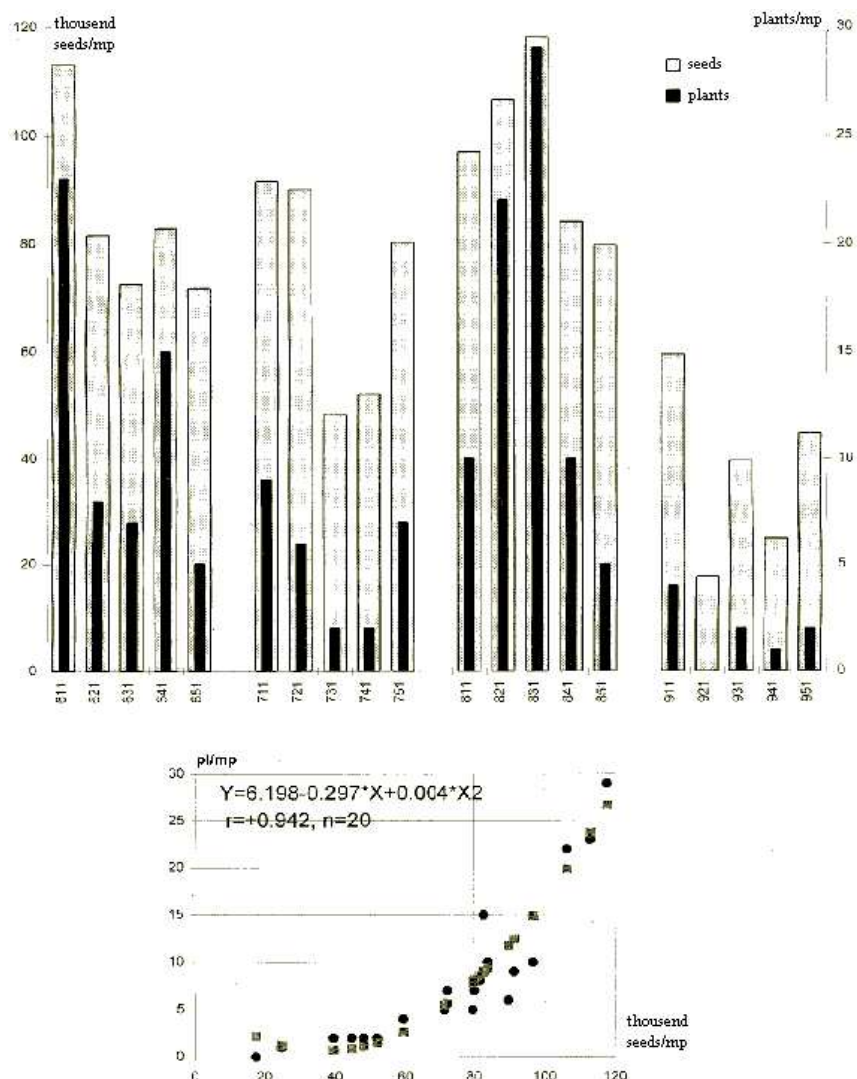


Fig. 1. Effects of treatments on seedbank and seedling emergence of *Chenopodium album*. Correlations between seedbank and number of seedlings. (The first number represents the treatments in maize, the second in potato and the third in winter wheat)

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# Long-term changes in the weed populations resulting from strategies for restricted herbicide use in field vegetable systems

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## Introduction

Applications of herbicides at reduced rates can be very effective because the full recommended doses are calculated to ensure reliable control of specified weeds under a range of conditions. However, if a lower rate is used, less susceptible individuals among the weed species may survive and set seed. Following the EU pesticide review, there is increased reliance on a small and still declining range of products that can be used in field vegetables. This restricted range of herbicides in combination with reduced application rates may compromise efforts to prevent the development of resistance or cause shifts in the weed flora (Gressel, 1995). Under typical cropping situations a combination of cultivation and different active ingredients used as part of a programme will normally help to buffer shifts in the above-ground flora, hence changes may be subtle and difficult to observe in the short-term. In particular, natural variation in herbicide sensitivity makes small responses difficult to detect in the early stages (Collings *et al.*, 2001). Using an artificially restricted herbicide programme to maximise the potential for change to occur, a field trial was designed to monitor and make a descriptive assessment of the weed flora over a number of years. An additional objective was to quantify the relative importance of the potential factors that may be involved in bringing about change.

## Materials and Methods

The field trial was made at Kirton, Lincolnshire, UK over a period of 9 years (1996 to 2004). To maximise the pressure for change treatments consisted of a single herbicide applied at the same rate to a plot in every year. In addition, the experimental plots were subjected to only a single shallow cultivation in the spring of each year. This was to minimise the diluting effect of the seedbank and hence maximise the potential contribution made by the most recently shed seed from weeds that had either survived sub-lethal herbicide applications or had emerged after treatment. Six different herbicides were selected as treatments; propachlor, pendimethalin and linuron (all applied pre-emergence) and ioxynil, bentazone and linuron (all applied post-emergence). All of the herbicides are commonly used in field vegetable production in the UK. Each herbicide was applied at either 1/4, 1/2 or full-recommended rate. Untreated plots were also included to enable baseline changes in the weed flora to be monitored. The weed flora on the plots was recorded for species composition on an approximately monthly basis each year through the growing season. An additional part of the study was to collect seeds from surviving individuals of selected species to determine whether there were detectable differences in germination, resistance and subsequent growth of the progeny.

## Results and discussion

Relative to 1996, a change in the weed flora was recorded over the duration of the study with an increase in the number of species recorded on all plots, reaching a maximum averaged over all treatments in 2002. As the area chosen for the trial had

been previously intensively cropped, it was not surprising that adopting either the zero herbicide regime or one of the single herbicide regimes, would cause this significant increase in the number of species recorded ( $P<0.001$ ). In addition, control plots and those receiving reduced application rates tended to support the more diverse floras ( $P<0.001$ ). Since 2000, the increase in the total number of species recorded appeared to slow down, even decline, suggesting that it may have started to stabilise.

Generally, plots receiving pre-emergence treatments had a greater number of species than those receiving post-emergence treatments (Figure 1). For example, linuron, applied both as a pre and post emergence treatment gave a greater species diversity when applied pre-emergence. Clearly the timing of application can be as important in determining the emerging flora as the choice of active ingredient.

Differences were observed in the composition of the weed flora. For example, *Sinapis arvensis* and *Thlaspi arvensis* were dominant on the plots receiving propachlor and pendimethalin, whilst *Poa annua* was dominant on the plots receiving ioxynil and bentazone. For propachlor, significantly greater species richness ( $P<0.001$ ) and lower species dominance were observed compared with the control. This suggested that the selective nature of the product was removing a dominant species from the flora thus creating a gap. This reinforces that herbicides can be used to modify the weed flora composition as selective management tools (Pywell *et al.*, 1998)

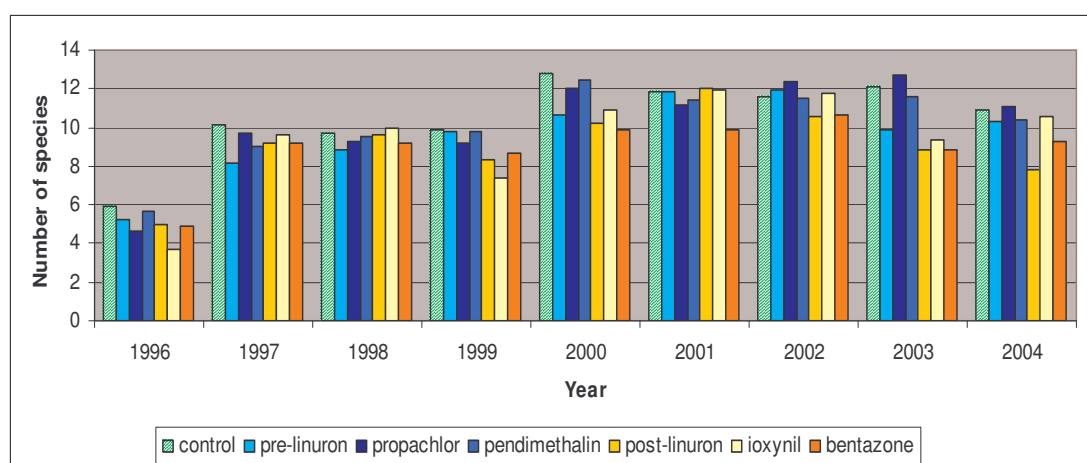


Figure 1. Number of species recorded on plots receiving different single-product herbicide treatments between 1996 and 2004.

The weed flora resulting from the 9-year study represents a complex and significant interaction of season, application rate and product ( $P<0.001$ ). The selective nature of the products and whether they were applied either pre or post emergence, were thought to be the primary drivers of change in the weed flora. However there also appeared to be strong seasonal impacts on herbicide effect ( $P<0.001$ ). The aim of the study is now to subject the complete data set from 1996 to 2004 to a more detailed analysis that will help to define the relative importance of the factors involved.

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# Research concerning the influence of minimum soil tillage systems on weed density and crop yields of soybean, wheat, potato, rape and corn

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## Introduction

The paper presents the influence of a minimum soil tillage system on weeding and crop production of soybean, wheat, potato, rape and corn in the conditions of the Cluj-Napoca area, Romania. The practice of minimum soil tillage systems leads to modifications in weed seed reserves in the soil, both in the amount of weeds and their floral composition (Cardina et al., 1991; Cristian & Ball, 1994; Guş et al., 2003).

## Materials and methods

Experiments were carried out between 2000 and 2004 at the University of Agricultural Sciences and Veterinary Medicine in Cluj-Napoca, on a plane field, with alluvial mollic soil and having a ground water depth of 2-3 m. On the 0-20 cm depth the clay content was 51.1% and the pH was 7.25. The base saturation degree was 96%, humus content was 3.21%, phosphorous 124-148 ppm, potassium 148-172 ppm and total nitrogen 0.143-0.158%. The average multi-annual value for rainfall was 613 mm, and the average air temperature value was 8.2°C.

The experiments included two soil tillage systems (classic and minimum tillage) and 5 different crops: soybean (Perla variety), wheat (Arieşan variety), potato (Sante variety), spring rape (Bolero variety) and corn (PR39D81 hybrid).

*Classic system:* V<sub>1</sub> – plough + disk (2x)  
*Minimum tillage system:* V<sub>2</sub> – chisel + rotary harrow,  
V<sub>3</sub> – paraplow + rotary harrow,  
V<sub>4</sub> – rotary harrow.

The weed control was carried out uniformly in every crop to identify the influence of the soil tillage system. The amount of weed was assessed in 4 replicates, on a 0.25 m<sup>2</sup> area, 2 weeks before harvesting. The crop yields were determined for every crop, treatment and replicate.

## Results and discussion

By continual use of the minimum soil tillage systems for 5 years on the same plots, modifications in floral composition and amount of weeds were observed (Table 1). Thus, in the ploughed treatment (V<sub>1</sub>), the number of weed species decreased from 7 to 11. On the minimum tillage system, the number of weed species increased by an average of 3-5 in the chisel and paraplow treatments and by an average of 6-7 in the rotary harrow treatment.

The amount of weed increased in all minimum soil tillage systems treatments, compared with the classic system (annually ploughed plot). The increase in weed density was different, depending on the tillage system and the ecology of the individual weed species.

The density of annual monocotyledonous weed species was increasing by 15-19% in the treatments worked with chisel and paraplow and by 33% in the treatment worked with the rotary harrow. In the case of annual dicotyledonous species the growth was 5-16% for the treatment worked with chisel, 7-20% for the paraplow and 22-36% for the rotary harrow. The



maximum weed densities were recorded in the case of perennial dicotyledonous weeds (*Convolvulus arvensis* and *Cirsium arvense*): 23-25%, 27-30% and 41-44% for the chisel, paraplow and rotary harrow treatments respectively.

Table 1. The soil tillage systems influence on weed density and floristic composition

Tillage system	Plough + disk (2x)		Chisel + Rotary harrow		Paraplow + Rotary harrow		Rotary harrow	
	no/m <sup>2</sup>	% (W.)	no/m <sup>2</sup>	%	no/m <sup>2</sup>	%	no/m <sup>2</sup>	%
<i>Setaria glauca</i>	0.78	100	0.90	115	0.93	119	1.04	133
<i>Amaranthus retroflexus</i>	2.42	100	2.80	116	2.90	120	3.20	132
<i>Chenopodium album</i>	2.00	100	2.11	105	2.14	107	2.68	134
<i>Galinsoga parviflora</i>	1.88	100	2.01	107	2.23	119	2.30	122
<i>Polygonum convolvulus</i>	1.44	100	1.64	114	1.62	112	1.96	136
<i>Cirsium arvense</i>	0.88	100	1.08	123	1.12	127	1.24	141
<i>Convolvulus arvensis</i>	1.46	100	1.82	125	1.90	130	2.10	144
Weed species number	7-11		9-12		13-16		15-17	

Within the same working system, the weed density also differs because of the crop (Table 2). The minimum soil tillage treatments ensured that crop yields were similar to those obtained in the conventional system, except for the potato and corn crop. The rotary harrow treatment was found to be useful for the rape and wheat crops.

Table 2. The influence of soil tillage systems on weeding and crop yield

Cultivated crop	Weeding Crop yield	Soil tillage system			
		Plough + disk (2x)	Chisel + Rotary harrow	Paraplow + Rotary harrow	Rotary harrow
Soybean	kg/ha (S.U.)	260	270	280	290
	kg/ha	2848	2860	2867	2747
Wheat	kg/ha (S.U.)	670	720	830	870
	kg/ha	3451	3391	3387	3282
Potato	kg/ha (S.U.)	1520	1860	1900	2450
	kg/ha	29428	26317	26853	19521
Rape	kg/ha (S.U.)	530	730	820	860
	kg/ha	1588	1552	1532	1505
Corn	kg/ha (S.U.)	1070	1830	1900	1890
	kg/ha	5857	5704	5737	5395

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# Dispersal of weeds by tillage and harvest in maize

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## Introduction

Crop management can facilitate the formation of spatially asymmetric patterns by dispersing weed seeds further in the driving direction than perpendicular to this. Cultivation caused seeds to travel up to 23 m (Mayer *et al.*, 1998). However, the majority of weed seeds are not dispersed further horizontally than 1m from source (Rew & Cussans, 1997). Still, each pass with cultivation equipment through the field is likely to make the spatial distribution of seeds in the soil less uniform, resulting in the elongated shape of weed patterns. For better understanding of spatial behaviour of weeds, we think it is important to look at dispersal processes on a field wide scale. The aim of this study was to investigate the dispersal by harvester and cultivator in a field cropped with continuous maize and to determine the effect of timing of seed shedding on dispersal.

## Materials and methods

The experiment was performed in 2002, on a 2 ha sandy soil field near Wageningen. The entire field was used in the experiment and was cropped with maize. Plant species used as weed plants were not present in the field before the start of this study. Plots, free of maize plants, were sown with *Sinapis alba* and *Phacelia tanacetifolia*. At time of harvest these species were carrying a large amount of ripe seeds. Just prior to harvest, seeds of *Silybum marianum* and *Borago officinalis* were placed on the soil surface of small plots. Agricultural practices were performed in a fixed pattern, driving in circles, anti-clock-wise. A few weeks after harvest and cultivation of the soil with a cultivator, spatial distribution of plant material and seedlings of the entire field was mapped using 1.5×1.5m contiguous quadrats.

## Results and discussion

Dispersal of *B. officinalis* was similar to that of *S. marianum*, and dispersal of *P. tanacetifolia* was similar to that of *S. alba*. Location of *S. marianum* and *S. alba* before and after harvest is shown in Fig. 1 & 2 respectively. Forage harvester and cultivator were able to transport seeds and plant material in the direction of the field traffic, into the headlands, which appeared to be the limit for dispersal. This is probably caused by turning of machinery and unearthing of the tines before the start of the next pass. Lateral dispersal was minimal. Plants with ripe seeds on them generally dispersed further from their initial source than species that had already dropped seeds prior to harvest. This was also found by Colbach *et al.* (2000). Effect of forage harvester and cultivator will also be determined in separate experiments on clay soil. The consequences of the experimental results for spatio-temporal behaviour of weeds in practice will further be studied with the aid of a model, which will consist of a series of functions describing dispersal by tillage and harvest in continuous maize cropping.

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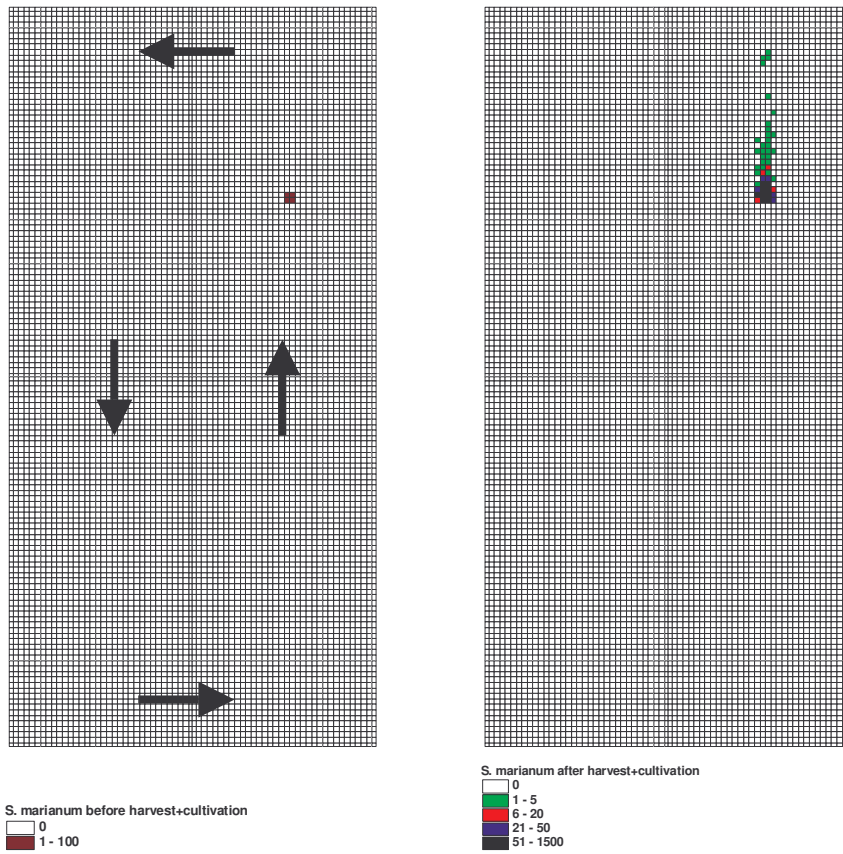


Figure 1: Location of *S. marianum* before and after harvest + cultivation

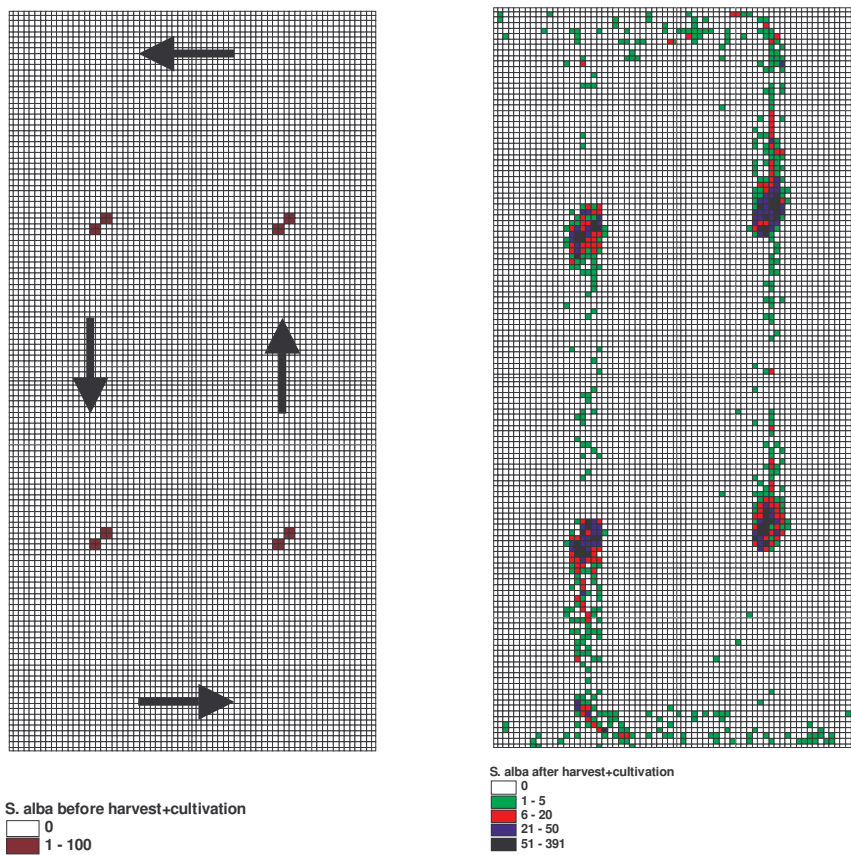


Figure 2: Location of *S. alba* before and after harvest + cultivation

# Field weed population models: a review of approaches and application domains

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## Introduction

Mathematics is widely accepted as one of the purest forms of science. While the mathematical models of physics are accepted as laws of nature, mathematical models of living systems are approximations to nature, simplifications of systems much too complex to grasp fully and in detail. Ecological modelling has been seen as ‘the construction of elaborate diagrams and mystico-mathematical representations of assumed relationships’ (Hedgpeth 1977). Although mathematics can be used as the common language of natural science, wherein thoughts can be expressed objectively and unambiguously, ‘Was sich überhaupt sagen lässt, lässt sich klar sagen’ (Wittgenstein 1918), to the uninitiated mathematical models can be incomprehensible and an obstruction, rather than a pathway, to communication and insight. How can we as scientists put models to best use?

‘Models help us formulate notions ... about the dynamics of the different species that an ecosystem comprises. These models are most useful when they help us to formulate and to test theory ..., and to manage ecosystems in an environmentally friendly manner’ (Gutierrez 1996). Thus we must use models both as thinking tools, helping scientists to form a consistent conception of ecological systems and providing a frame for their research, and as practical tools ultimately causing farmers to take better decisions. From the outset of model development, it is important to realise which aim has priority.

We evaluated models of weed population dynamics based on an analysis of their assumptions, biological rationale, flexibility, documentation, accessibility, demand for parameter estimation and documented validity. We arrived at general recommendations regarding which modelling approach should be applied in order to address different application domains.

## Materials and methods

The development of weed population dynamics models has not been excessively prolific, maybe due to the problem of validating such long-term processes. Thus we aim at reviewing all models of this kind, expecting to find 50 models in total, only 10-15 of these being very complex. We will identify which questions the models address, the structure of model, what kind of life cycle aspects are accounted for, which factors are assumed to have an influence on population dynamics (intrinsic species characteristics, weather and climatic factors, management), and whether any validation has been attempted.

## Results and discussion

Natural populations are regulated via factors operating in a density-dependent manner (Gutierrez 1996). For weed populations these factors are competition for common resources within and between species, but also weed control actions if these are applied by the farmer in response to weed density. Density-dependent relations are thus at the heart of any model of weed population dynamics. Positive feedback, readily seen in the explosive proliferation of weeds in badly managed fields, is the force underlying the necessity for weed control. When

positive feedback mechanisms are coupled with density-dependence in models, chaotic behaviour may arise (Berryman and Millstein 1989) and indeed weed population dynamics have been claimed inherently chaotic and thus in principle unpredictable (Firbank 1989; Gonzalez-Andujar and Hughes 2000). In the field, however, environmental variation caused by weather and agricultural practice are the main causes for the variability of weed population dynamics, rather than chaos (Berryman and Millstein 1989; Freckleton and Watkinson 2002). Thus weed population dynamics *are* predictable – within bounds.

General recommendations for developing ecological models:

(1) The questions that the model should address must be clearly formulated, before an appropriate modelling approach can be chosen and modelling itself begun. Beware of the temptation to just get the modelling going as a starting point.

(2) For a model to have scientific merit, its mathematics and rationale should be published, and its implementation made publicly available on Internet or by request. The model coding itself should be open for scientific review. Demanding code secrecy, in style of commercial software, is incompatible with scientific ethics and progress.

(3) Turning the model into a black box, either through sloppy implementation or documentation, should be avoided. The modeller is likely to lose track of the model's inner workings, and the scientific community is likely to lose interest all together.

(4) The model should be based on well-established biological relationships and sound rationale, rather than inventing all model components and equations anew.

Specific recommendations for developing weed population dynamics models:

(5) 'Agronomists look for differences, biologists for relationships' (G. Nachman, pers. cit.). To aid model synthesis of experimental results, weed scientist should give more emphasis to uncover general biological relationships than tabulate effects of agronomical treatments specific to site and year.

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# Incidence of weed infestations in cropped and uncropped arable fields subject to mechanical and chemical weed control measures

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## Introduction

Harsh economic situations have necessitated Polish farmers to leave some areas of agricultural land uncropped. These areas are colonised immediately by arable weeds, the majority of which develop ripe seeds which are shed on to the soil. The soil seedbank is a main source of weed infestation (Kropff *et al.* 1996, Pawlowski 1966, Wesolowski 1984). These seeds may remain viable and pose a threat to subsequent crop yields.

## Material and methods

Soil infestation by weed seeds was investigated in cropped (winter wheat) and un-cropped fields (ley fallow following winter wheat in the 1995/6-1997/8 period). The research was conducted on loess soil; forecrop of winter wheat as well as fallow was potato. Both cropped and un-cropped fields underwent the same agronomic practices and treated with the same weed control measures. Experimental treatments were: untreated –A, harrowing at GS 10-11 Zadoks *et al.* (1974) – B, at GS 25-26 C, herbicide thifensulfuron-metyl + chlorsulfuron, (40.92+4.09 g ha<sup>-1</sup>), 7 days after spring harrowing – D, and combination of these BC, BD, CD BCD. Soil samples were taken on the day of wheat harvest (27, 22 and 22 of July in 1996, '97 and '98 respectively). Five soil samples from each plot (20 m<sup>2</sup>) were taken and thoroughly mixed; 1/5 of that (by weight) was partitioned for analysis; washed and sieved, the seeds extracted (by hand with using of forceps) and the number of viable seeds determined.

## Results and discussion

Table 1. Density of weed seeds (no m<sup>-2</sup>) and species in 0-1 cm soil layer and some traits of winter wheat (three years mean)

Treatment (applied on crop and fallow)	Weed seeds m <sup>-2</sup>			Weed plant m <sup>-2</sup>		Winter wheat m <sup>-2</sup>	
	cropped field and –species no	fallow field and –species no	seed ratio: fallow to cropped	fallow	winter wheat	seedling density at GS 10-11	ears density at harvest
A	13243.5 – 11	119476.4 – 17	9.0	178.7	103.0	466.3	565.9
B	9972.4 – 13	108097.4 – 15	10.8	165.2	139.4	471.8	589.0
C	9660.6 – 14	132520.8 – 16	13.7	129.6	69.3	459.1	611.1
BC	12944.9 – 11	92127.0 – 14	7.1	142.7	76.8	439.4	571.3
D	13084.2 – 10	39591.0 – 12	3.0	130.0	48.4	481.9	610.5
BD	12779.0 – 12	36804.3 – 11	2.9	114.1	49.4	489.6	594.1
CD	8797.9 – 12	33175.0 – 11	3.8	99.8	43.6	476.6	684.6
BCD	7152.5 – 11	37136.1 – 12	5.2	143.0	28.7	454.4	619.1
Average	10954.4 –17 *	74866.0– 25*	6.8	137.9	69.8	467.4	605.7

species number in all objects as total (not average)

The soil seedbank is a potential source of weed infestation in cultivated crops (Kropff *et al.* 1996, Pawlowski 1966, Wesolowski 1984). Agronomic practices play an important role in limiting that source. From the different weed control measures, presented in Table 1, harrowing supplemented with a herbicide – CD and BCD as applied separately B and C markedly diminished seedbank size on cropped field. Setting arable land aside, even in the

short-term increased the number of weed seeds in the soil substantially from almost three (BD) to almost fourteen times (C) relative to cropped field and also increased weed species diversity (about 8 species in total). Conversely, the presence of a crop even without any weed control treatment (A) resulted in a nine-fold reduction in the size of the seed bank; and was even more effective if subject to harrowing B and C.

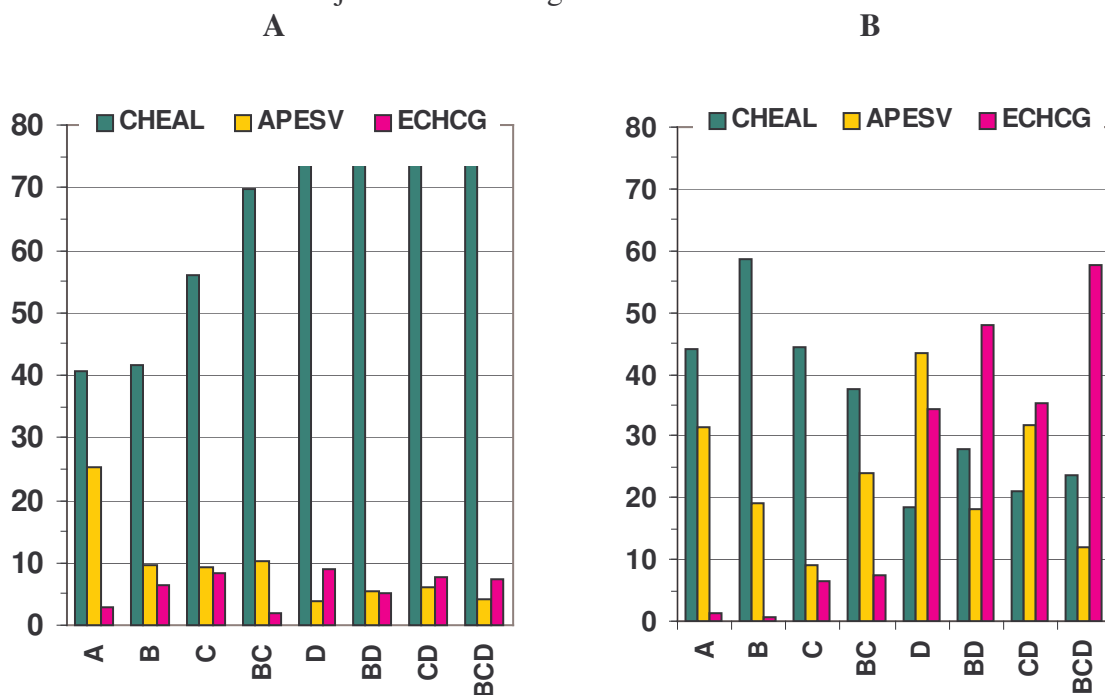


Fig. 1. The contribution (%) of dominant species in the soil seedbank under field cropped with winter wheat- A and uncropped (fallow) B

The arable weed species exclusively occupied the cropped and uncropped field (Jedrussczak et al. 2004). The structure of their seeds by species in the soil bank was rather different on analysed places. The greatest seed density belonged to *Chenopodium album* L. (CHEAL) amongst 17 species of cropped field (Fig. 1). *Apera spica-venti* (L.) P. Beauv. (APESV) and *Echinochloa crus-galli* (L.) P. Beauv. (ECHCG) were the next most frequently occurring but at much lesser densities than *C. album*. Under fallow (with 25 species) seeds *C. album* dominated mainly on harrowed plots and oscillating about 20% in other treatments. From the D (herbicide treatment) the most important frequently occurring was *E. crus-galli* and *A. spica-venti*. Within the weed vegetation, *C. album* constituted 0.1% to 38% in winter wheat and 7% to 60% in fallow (Jedrussczak et al. 2004 a, b). This and the other species referred to are abundant under loess soil in the central-east part of the region where the investigation was conducted. Thus a considerable weed problem will occur on reversion to arable cropping following a period of fallow.

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# Studies on Weed Population Growth During the Vegetation Period of Spring Forms of *Triticum durum* and *Triticum aestivum*

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## Introduction

The demand for the grain of durum wheat is increasing in Poland. An attempt to introduce the cultivation spring of forms of *Triticum durum* to the soil-climatic conditions of the Lublin area showed, that they gave a 40% lower yield as compared to *Triticum aestivum*. Therefore, a hypothesis was put forward that one of the reasons for lower yields was that it was less competitive with weeds.

## Material and Methods

The field experiment was established in 2002-2003 on loess soil (pH 5.5-5.6) at Czesławice Experimental Station, near Nałęczów in central-eastern Poland. The experiment consisted of three replicates of a split-plot randomized block design. Each replicate included two breed lines: LGR 1359/8, LGR 899/17A of *Triticum durum* and cultivar Helia of *T. aestivum* as the main plots. Each main plot was split for five periods, during which the weeds remained on plots: B. from sowing to 3-4 leaves of cereal; C. from sowing to tillering; D. from sowing to shooting; E. from sowing to earing; A. from sowing to maturity. The wheat was sown (550 grains m<sup>-2</sup>) on March 19, 2002 and April 18, 2003, according to agronomic recommendations. Each plots was only weeded, the timing of which was according to the appropriate stage of cereal growth as set out above (A, B, C, D or E). The weeds were pulled out manually from the whole area of the plots (1.5 m x 5.5 m) measured, air-dried and then weighed. There were differences in the growth rates of the above-ground weed matter (W<sub>m</sub>GR) g·m<sup>-2</sup>·day<sup>-1</sup>, the increase in the number of individual weeds present per day·m<sup>-2</sup> (W<sub>n</sub>GR) and frequencies of dominant weed species (%) on the basis of the studied parameters. Calculations of W<sub>m</sub>GR were performed on the basis of the study by Kocoń *et al.* (1977). The results were statistically evaluated by analysis of variance and a Tukey's test ( $\alpha=0.05$ ).

## Results and Discussion

During the vegetative growth of the wheat, which lasted about 120 days, the studies identified a greater average increase of the number of weeds (W<sub>n</sub>GR) in a canopy of *T. durum* (0.90-0.62 weeds·m<sup>-2</sup>·day<sup>-1</sup>) as compared to the canopy of *T. aestivum* (0.50 weeds·m<sup>-2</sup>·day<sup>-1</sup>). Besides, the rate of biomass accumulation in weeds (W<sub>m</sub>GR) in both lines of durum wheat was greater (1.54-1.17 g·m<sup>-2</sup>·day<sup>-1</sup>) as compared to the canopy of *T. aestivum* (0.70 g·m<sup>-2</sup>·day<sup>-1</sup>). The values of W<sub>m</sub>GR differed from each other in a significant way (LSD<sub>0.05</sub>=0.34). In addition, the studies found a significant dependence of both indexes on growth stages of wheat. The relationships shown in the figure presents a curvilinear function (Fig. 1). The maxima of W<sub>n</sub>GR in both lines of *T. durum* fall in the phase of tillering (21-22 BBCH), while in the case of *T. aestivum* it was 11 days later. They are generally convergent with the period of the largest emergence of weeds in spring cereals (Jędruszczak 1993). The accumulation of mass in weeds had a less dynamic course than in the case of emergence. The rate, which was slight in the beginning, increased together with the growth and development of wheat. Prior to harvest it slowed down, with an exception of line LGR 1359/8. A higher rate of increase in the number and mass of weeds in the short-strawed awnless line LGR 899/17A (67 cm in high) as compared to the medium-high awned line LGR 1359/8 (85 cm high) and 95 cm-high cv. Helia, indicate a significant relationship with the plants' height. Likewise, the

height of winter wheat plants affects the damage thresholds of *Apera spica-venti* and *Avena fatua* (Kapeluszny 1986). The average frequency of the dominant weed species in the canopy of the discussed species of wheat was: *Galeopsis tetrahit* – 73.6%, *Chenopodium album* – 63.9%, *Stellaria media* – 36.1%, *Viola arvensis* – 33.3% and *Cirsium arvense* – 26.3%.

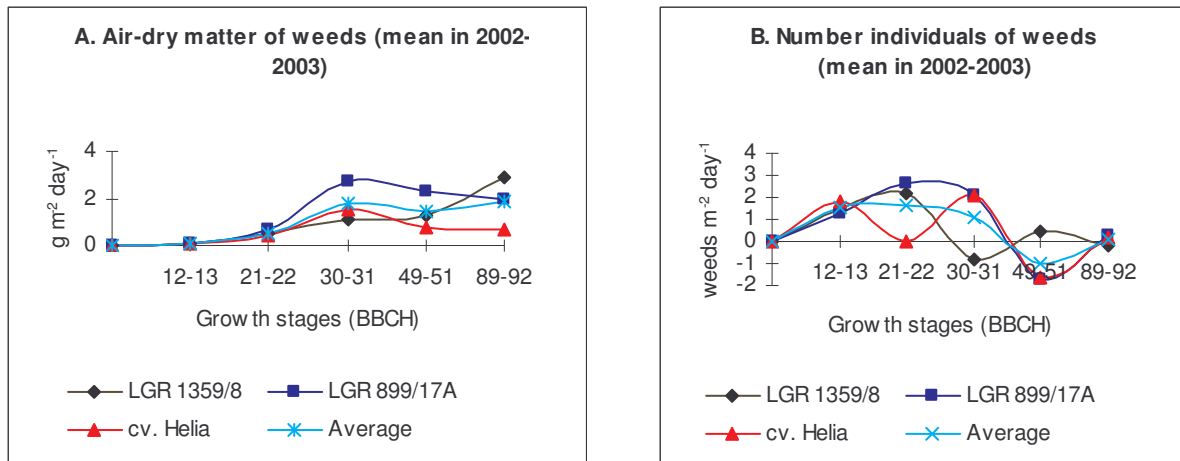


Figure 1. The dynamics of weed mass and number depending on the developmental stage of spring forms of *Triticum durum* and *T. aestivum*. A. LSD<sub>0.05</sub> for the air-dry matter of weeds: between species-0.34; between growth stages-1.46. B. LSD<sub>0.05</sub> for the number individuals of weeds: between species-not significant; between growth stages-2.55

## Conclusions

In the soil-climatic conditions of the Lublin region the studied lines of durum wheat showed smaller competitive ability in relation to weeds as compared to *Triticum aestivum*.

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# Studies on the seed production and germination characteristics of *Poa annua*, *Senecio vulgaris*, *Stellaria media* and *Tripleurospermum inodorum*

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## Introduction

A good understanding of the population dynamics of arable weeds is essential if lower input weed management is to be successfully promulgated for UK winter cereal crops. Such management approaches will result in the survival of plants in the crop and the production of seeds that may impact on infestation levels in subsequent crops. Sound data on seed production are needed to predict consequences for these crops. Such information can be used either as an aid to direct advice or as a component of computer-based decision support. The work reported here forms part of a larger area of work aimed at quantifying weed behaviour through a rotation and reports on seed production and germination of *Poa annua* L., *Senecio vulgaris* L., *Stellaria media* (L.) Vill. and *Tripleurospermum inodorum* (Schultz Bip).

## Materials and methods

In the seasons 2001/02 to 2003/04 seeds of *P. annua*, *S. vulgaris*, *S. media* and *T. inodorum* were broadcast in the autumn into plots of winter wheat, or at the same time into areas without wheat. The wheat and unsown areas were treated similarly, in that they both received fertiliser rates appropriate for the wheat. In most experiments herbicide treatments were used, where possible, to minimise other weed species but sample areas to be used for seed production studies were also hand-weeded when needed. In a sub set of experiments herbicide treatments that partially killed the *T. inodorum* and *S. vulgaris* were also studied.

Individual plants of *P. annua*, and *S. vulgaris* were harvested from the plots as they approached maturity in June/July. *T. inodorum* plants were harvested just before harvest in August. For *S. media* plant numbers in the sample areas were counted in the winter/spring and 0.25m<sup>2</sup> areas were harvested in June. Flower, panicle and capsule numbers were counted and the plants were dried and weighed. A sub-set of capsules (or panicles for *P. annua*) were dissected to calculate seed numbers. More details of methods are given in Lutman, (2002). Seeds were also collected from mature flower heads at intervals during the summer. These were dried and were tested for their dormancy status by germinating four sets of 50 seeds/plot in Petri-dishes in incubators at 10-20°C (12/12hr) either in light/dark (12/12hr) or in continuous darkness. Numbers of germinating seeds were recorded. All field experiments were of a randomised block design with 3 or 4 replications.

## Results and discussion

*Seed production* The numbers of seeds produced was regressed against plant weight for all the experiments. Good correlations between seed number and plant dry weight with Log/Log transformed data were found for all species in all experiments (e.g. Fig. 1). The presence of crop competition reduced seed production appreciably (Table 1) but did not markedly change the allometric relationship between seed numbers and plant weight. Variation in weather conditions caused variation in plant weight and hence seed production. Data from *S. vulgaris* and *T. inodorum* indicated no marked change in the allometric relationship (plant weight / seed number) when plant vigour (weight) was severely reduced by herbicide treatments. These data indicate that even modest weed infestations can produce appreciable numbers of seeds, especially in the absence of crop competition. The measurement techniques used estimate the maximum potential for seed production and require comparison with seed rain studies that generally predict lower numbers of seeds landing on the soil surface.



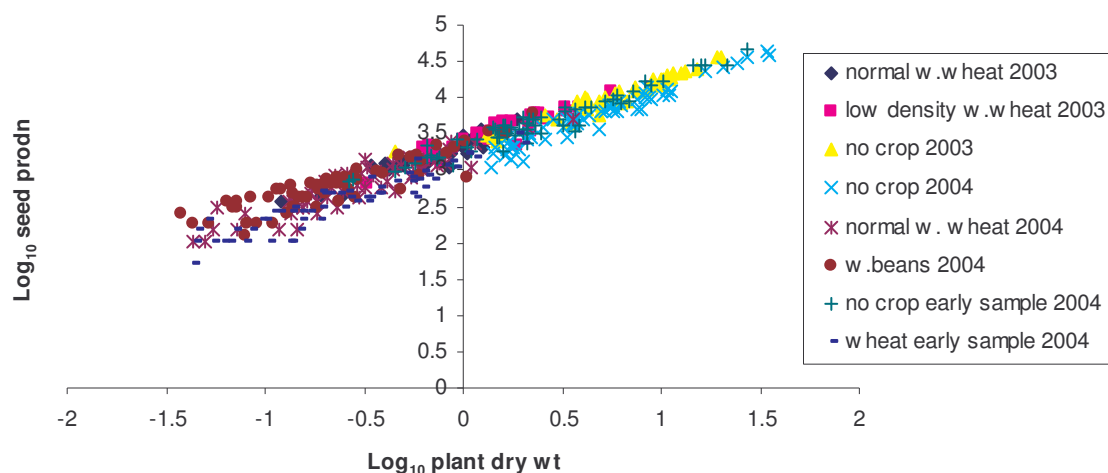


Figure 1. Relationship between  $\text{Log}_{10}$  plant weight and  $\text{Log}_{10}$  seed production for *S. vulgaris* (data collected in summer 2003 and 2004)

Table 1 Average seed number/plant from four annual weeds in winter wheat and when growing alone (estimated from regression curves) (data are grand means and year ranges)

Weed species	W. wheat	No crop
<i>P. annua</i>	1554 (845-2844)	8869 (3148 -18663)
<i>S. vulgaris</i>	984 (607-1503)	6166 (5470 – 8872)
<i>T. inodorum</i>	1082	30000
<i>S. media</i>	2953	20645

*Seed germination* Seeds of all four species germinated more readily in the presence of light. With *T. inodorum* there was very little germination in the dark. Seeds produced by early cohorts of *P. annua* flowers germinated more slowly than later produced ones. Interestingly, stored seeds (4 months storage) of this species, unlike fresh seeds, failed to show inhibition of germination in the dark. Although there was only low germination of seeds of *S. vulgaris* in the dark, very early and very late produced seed cohorts germinated more readily. *S. media* seed germination seemed unaffected by treatment of the parent plant with a number of herbicides. Generally there was high germination in light/dark, less germination in dark alone.

Overall, it seems clear that given adequate moisture many seeds will germinate as soon as they are shed, provided they are in the light. The creation of a persistent seedbank seems to depend on the incorporation of the seeds into the soil (darkness) either by natural processes or by cultivation. From our somewhat limited data it appears that partially effective herbicides applied to young seedlings do not affect the dormancy (and viability) of the seeds. Although there were detectable differences in behaviour between cohorts of seeds, these differences appeared relatively minor, in comparison to the effects of light and dark.

This information on seed production, dormancy and germination of annual arable weeds, forms a key element of the prediction of weed behaviour through a rotation, and aspects already feature in the biological models of the UK's weed management decision support system (Collings *et al.*, 2003).

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# Mapping *Orobanche* spp. infestation degree in arable crops by using Geographical Information Systems (G.I.S)

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## Introduction

Several arable crops (tobacco, tomato, sunflower, faba bean, etc.) are severely attacked by *Orobanche* spp. in different regions around Greece. In order to study the distribution of *Orobanche* spp., global positioning (GPS) and geographic information systems (GIS) are developed. It is well known that GPS can record site and boundary data directly in the field, and when linked with GIS can transfer those data to computerized equipment to compile and generate maps and other data sets. The recognized advantages of GPS for surveying *Orobanche* spp. are twofold. One is its ability to record accurately the boundaries of *Orobanche*'s infestation. A second is the ability to interface with GIS to position accurately the infestations in relationship to other data referring to topology, phenology, climatology, etc. (Lawrence W. Lass *et al.* 1993). The aim of this study is to provide baseline information, as far as the infestation level is concerned, registering, in a parallel way, all the obtained information from the sampling points (soil parameters, climatic data, etc.) in a database for future comparisons on local, regional and supra-regional level.

## Material and Methods

The sampling took place in Orestiada (41° 30' N latitude), Katerini (40° 16' 30'' N latitude), Kozani (40° 18' N latitude), Domoko (39° 07' 30'' N latitude), Agrinio (38° 37' 30'' N latitude), Tithorea (38° 36' N latitude), Psaxna (38° 34' 30'' N latitude), Marathonas (38° 09' N latitude) and Nafplio (37° 33' N latitude) provinces during both 2003 and 2004. Katerini, Kozani, Domokos, Agrinio and Tithorea are areas where tobacco is traditionally cultivated. *Orobanche* samples were collected from tomato fields in Nafplio. Sunflower is cultivated on a very large scale in Orestiada. Psaxna and Marathonas are regions where faba bean is cultivated on a smaller scale. The selection of the fields in each region was based on the GIS principles and a specific methodology was followed in order to select the sampling points in each field with some modifications (Kroschel J.). In general, the fields were crossed in a zigzag line and six sampling points were inspected in each field. The exact geographical position of the sampling point was provided through a portable G.P.S. (Global Position System). Each field was estimated for the degree of infestation using a scoring scale from 0 to 6; 0 = not infested, 1 = very low, 2 = low, 3 = moderate, 4 = strong, 5 = very strong, 6 = host plants completely destroyed (Schmitt U., 1981). All the data concerning each sampling point (infestation magnitude, soil parameters, climatic data, phenetic features of the *Orobanche* samples etc.) were collected and processed thoroughly in the laboratory.

## Results and discussion

*Orobanche cumana* infested sunflower in Orestiada at a moderate level, although in some cases the inoculation was severe. *Orobanche crenata* was localized in Psaxna and Marathonas where the infestation of faba bean was not so high as to cause dramatic economic loss to production. *Orobanche ramosa* severely infested tobacco fields in Domokos, whereas the situation was not so dramatic in tomato fields in Nafplio. *Orobanche aegyptiaca* parasitized tobacco fields in Katerini, Kozani, Agrinio, Tithorea, but varied in the magnitude of the infestation. The tobacco fields in Tithorea were attacked viciously by *O. aegyptiaca* which caused deterioration in the quality of the collected leaves. The symptoms to the host plants from *Orobanche* attack were more severe when the parasite had an earlier emergence and as a

result the tobacco was unable to compete with the heavy population occurrence. In contrast Agrinio had the lowest infestation compared to the other regions. The image of the infestation in the tobacco fields in Katerini and Kozani was in the middle of the two extreme cases mentioned above, although in some tobacco fields in Kozani the damage was severe. Figure 1 demonstrates the distribution of *Orobanche* species and the infestation magnitude in all the regions mentioned. This variability in the degree of infestation may be attributed to the macro- (or) micro-climatic conditions that define the local environment. GPS and GIS may completely explain this variation by using the information registered in a database.

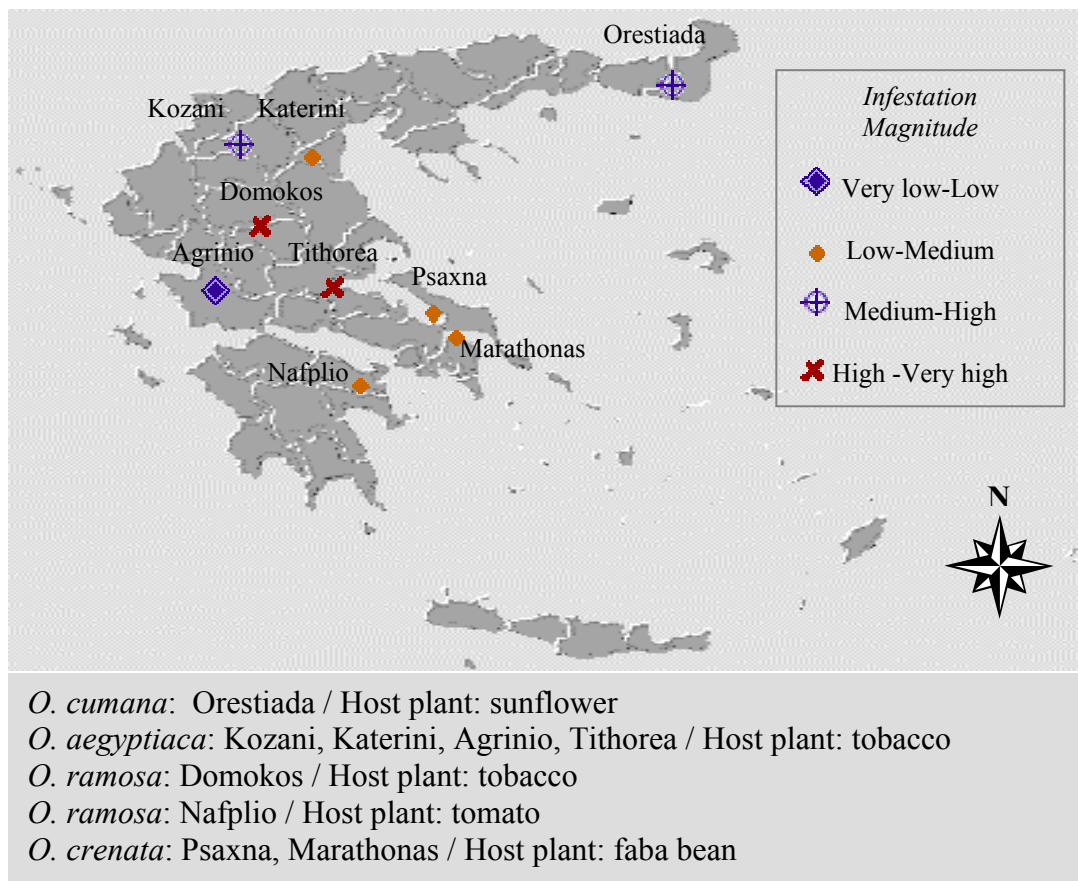


Figure 1: Infestation magnitude of *Orobanche* species in nine regions in Greece

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## Shrub flowering to aid weed control

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### Introduction

In organic farming multi-tactic, ecologically based, information-intensive weed management is necessary because the available control methods only guarantee a limited rate of kill (<50-70%). Whilst the range and efficiency of non-chemical weeding techniques is improving, the efficiency of mechanical weed control practices could be increased if the timing of control was strongly linked with the emergence dynamics of the weeds.

In agriculture, biological or phenological indicators are widely used to alert farmers about stress or parasite problems; knowing stress conditions or the timing of weed or pest phenology in advance enables farmers to plan their management practices more efficiently. Although there are many possible applications, the use of phenology in weed control is uncommon (Masin et al., 2005).

Indeed, the correct timing of weed control (whether mechanical or chemical) is the best guarantee for obtaining good treatment results and optimizing control strategies. Berti et al. (1996) also highlighted the need to understand and predict weed emergence patterns to improve decision-making capabilities and diversify Integrated Weed Management.

### Materials and methods

In a pre-alpine valley in northern Italy a study was conducted to evaluate whether the phenological phases of some spontaneous or planted shrubs could serve as reliable indicators of time of weed emergence, and consequently as a guide in management decisions.

In five sites of the Val di Gresta (northern Italy) at different altitudes (from 600 m to 1200 m a.s.l.), a phenological garden was created by planting two cloned individuals for each of seven species of shrubs (*Forsythia* sp., *Sambucus nigra*, *Syringa vulgaris*, *Rosa* sp., *Laburnum anagyroides*, *Prunus spinosa*, *Pyrachantha coccinea*). A series of spontaneous shrubs was also monitored (*Sorbus aucuparia*, *Evonimus europeus*, *Rosa* sp., *Cornus sanguinea*, *Cytisus* sp.). Plants were chosen, based on observations made in previous years, for having easily recognizable phases corresponding with the emergence period of annual weed species.

Plants were monitored weekly at all of the sites from February to September and the dates of the phenological phases were recorded. To make observation easy, every phase was identified with a code, as suggested by Zuin et al. (2003).

In those same sites the real weed flora was assessed using a permanently marked small sized plots ranging from 1,0 to 3,0 m<sup>2</sup>, where weed seedlings were counted once or twice a week, and removed. After each count, the top layer of the soil was disturbed (approx. 3 cm deep) to promote new seedlings emergence; on two occasion during summer the plots were watered to promote full expression of the weed seedbank in field conditions. Michelson & Stougaard (2003) showed that collecting a small number of large samples, rather than a large number of small samples, had little effect on precision. Rather, increasing the proportion of total area sampled is more important to improve precision.

Weather data and soil temperature at 5 cm depth were recorded in four out of the five sites for determining the degree day sums needed to reach the phenological phases of interest.

Cumulative relative seedling emergence (CRSE) was modelled using a Gompertz function, as follows:

$$\text{CRSE} = 100 * \exp(-a * \exp(-b * t))$$

where  $t$  is a proper time-dependent variable,  $a$  represents the lag before emergence starts, and  $b$  represents the rate of increase of emergence once it has been initiated.

Simple linear correlation analysis was used to measure the degree of linear association between the date corresponding to 5% of emergence for every species and the phenological events that could be used as potential predictors of this emergence.

## Results and discussion

Between sites there was a large effect of altitude, both for air and soil temperature. At the higher site the weed emergence started approx. 30 days later than at the lower altitude. No differences between rainfall were observed. In 2003 a total of 49 weed species were counted in the monitored fields. The number of species per field ranged from 20 to 30, with an average of  $26.6 \pm 1,8$  (mean  $\pm$  SE). Among these species, 27 were found in at least 3 fields and 10 (*Chenopodium album*, *Chenopodium polyspermum*, *Galinsoga parviflora*, *Amaranthus* spp. and others) were common to all fields, whereas 17 were restricted to only one field (“unique” species, e.g. *Digitaria sanguinalis*). The total number of weeds  $m^{-2}$  ranged from 475 to 1758, with an average of  $1210 \pm 153$ . The cumulative weed emergence pattern was a typical S-shape for all of the species that were recorded. In some case emergence can restart (eg. after rainfall in a dry period, as in 2003) (Figure 1). Correlations exist between phenological events and weed emergence timing, and their consequences in terms of integrated weed management are discussed.

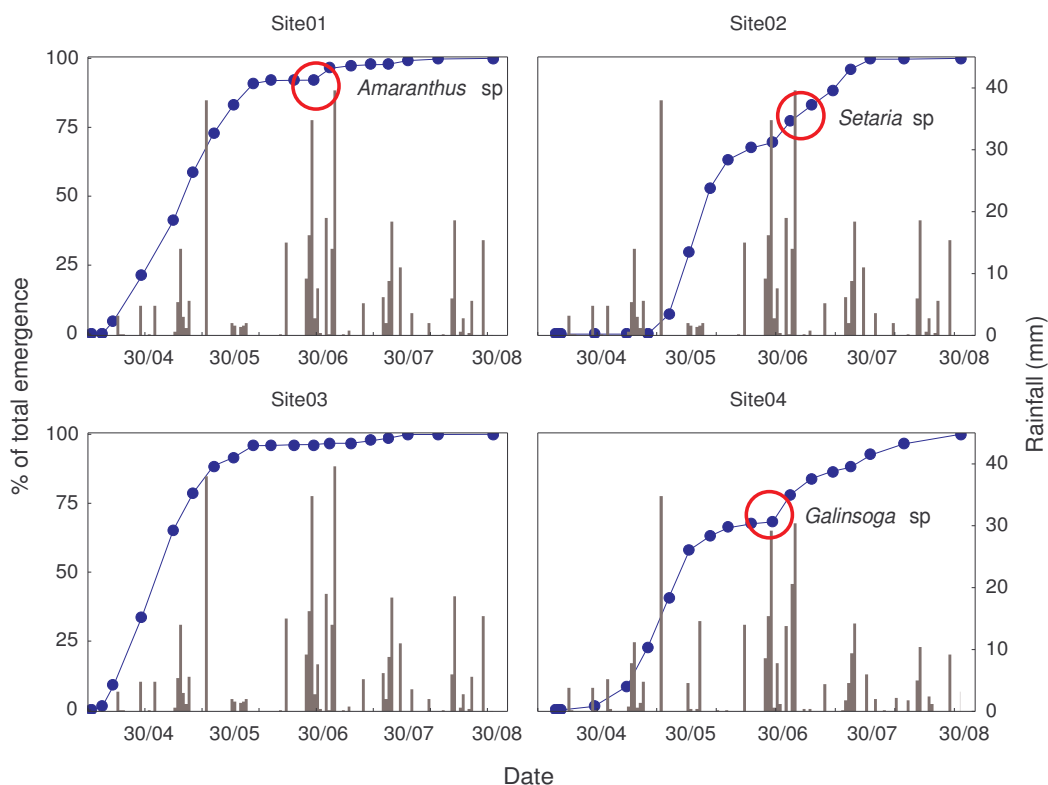


Figure 1. Weed emergence dynamics and rainfall for four monitored sites in 2003. Weed species restarting emergence are circled.

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## Assessing gene flow between and within populations of *Alopecurus myosuroides*

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### Introduction

*Alopecurus myosuroides* Huds. (black-grass) is a major annual winter weed in northern Europe where field conditions have led to the development of herbicide resistant populations. To control this weed, herbicides targeting the chloroplastic acetyl coenzyme A carboxylase (ACC'ase, EC 6.1.4.2) have been broadly used since the end of the 1980s. In France, black-grass resistance to ACC'ase-inhibiting herbicides very likely evolved following a "patchy" pattern. The large-scale evolution of herbicide resistance to herbicides in black-grass would result from the independent selection of various resistance mechanisms in local black-grass populations undergoing contrasted herbicide and agronomical selection pressures, and connected by gene flows the parameters of which remain unknown (Délye et al., 2004). In this study we used amplified fragment length polymorphism (AFLP) markers (Vos et al., 1995) to investigate the patterns of natural genetic variability in populations of *A. myosuroides* in Burgundy France. Our primary objectives were to (i) determine the extent of genetic variation between and within resistant and sensitive plants, and (ii) provide a preliminary estimate of gene flow between populations.

### Materials and methods:

#### *Plant Material*

Seeds of black-grass were collected in 2003 in 50 agricultural fields that have been sprayed with ACC'ase-inhibiting herbicides in the same region (Burgundy, France).

DNA was extracted from young leaf tissue from disease-free glasshouse grown plants (60 plants per population) as described by Doyle and Doyle (1990) with minor modifications.

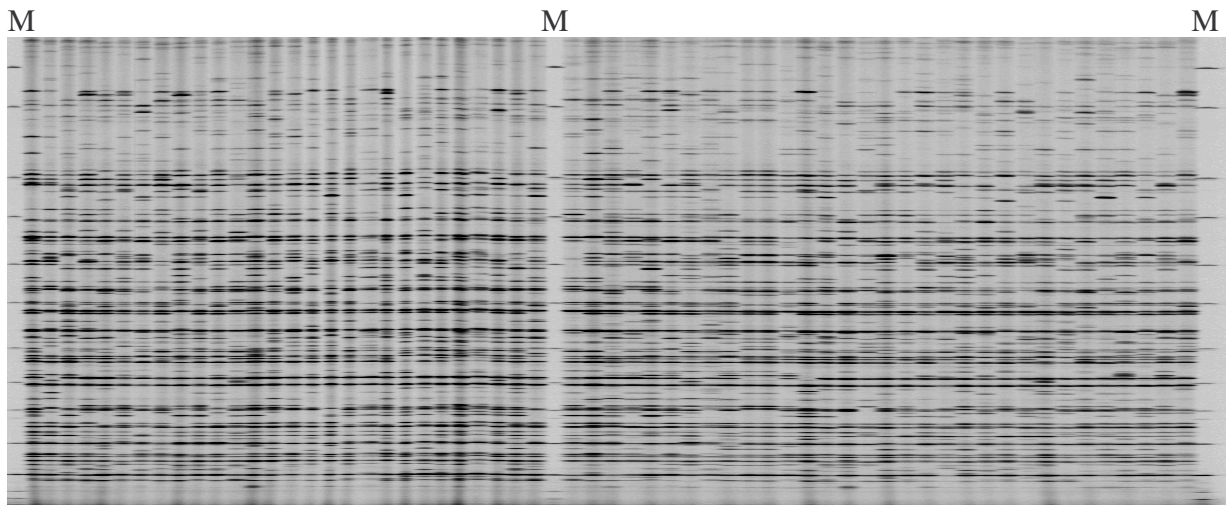
#### *AFLP analysis*

Following the restriction digestion of genomic DNA with restriction enzymes *EcoRI* and *MseI*, adapters specific to *EcoRI* and *MseI* were ligated. The first PCR step with non selective primers consists of 23 cycles of 5s at 95°C, 30s at 56°C and 60s at 72°C and a final cycle of 5min at 72°C. Selective amplification was conducted using pre-amplified DNA, diluted 5 times, as a template. Primers with three selective bases at their 3' ends were employed. The cycling program consists of one cycle of 5s at 95°C, 30s at 65°C and 60s at 72°C. The annealing and elongation temperatures were reduced by 0.7°C and 0.9°C per cycle respectively during the first 12 cycles, then 23 cycles were performed at 5s at 95°C, 30s at 56°C and 60s at 72°C and a final cycle of 5min at 72°C.

The DNA fingerprints were scored for presence (1) and absence (0) of bands using the dist AFLP software (Mougel et al., 2000).

### Results and Discussion:

To set up AFLP assay to analyze *A. myosuroides*, a total of 20 combinations of pairs of primers were tested, and one combination selected for AFLP analysis (Fig. 1).



**Fig. 1: Black-grass AFLP patterns. M, molecular mass marker**

This study will enable us to determine the genetic variation among 3000 black-grass plants issued from 50 field populations collected in the same region. Relationships among samples for the scored polymorphic bands will be estimated by coefficients of similarity (Nei, 1987; Dice, 1945). A cluster tree will be constructed and evaluated by a bootstrap analysis.

Association between the genotypic clustering of samples and their field of origin or the presence of any given mutation conferring resistance to ACC'ase-inhibiting herbicides will be determined. Analysis of Molecular Variance (AMOVA, Excoffier et al., 1992) will be performed to partition total genetic variation into its among individuals, within-population and between-population components. Genetic diversity, *He*, is used as a measure of genetic diversity within populations according to Nei (1987). The impact of herbicide selection pressures at the within population level will be examined by analysing the relationship between *He* for each field and the known herbicide application program during recent years, as well as between *He* and the frequency of resistant plants within the field.

Genetic differentiation among populations will be measured by *Gst* (Nei, 1987), a parameter which is correlated with the extent of gene flow among populations. The existence of a significant correlation between *Gst* values for pairs of field populations and their corresponding geographical distance will be investigated using Mantel tests (Mantel, 1967). This will provide a first estimation of the extent of gene flow in *A. myosuroides* at a regional scale.

Finally, comparison of the genotypes of resistant plants in the different populations will indicate whether resistance is propagated among fields via pollen or seed flow or arose from locally appeared mutations.

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# Development of weeds in organic crop rotation experiments

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## Introduction

Weeds are a major problem in organic farming. Preventive as well as curative measures must be utilised to manage the weeds and avoid proliferation. Besides direct weed control measures, many different aspects of planning and management in the cropping system affect the proliferation of weeds. However, it has rarely been investigated how the whole system affects weed populations.

## Materials and methods

A crop rotation experiment was designed to evaluate the possibilities for increasing production of cereals and pulses in arable organic farming without risking yield penalties, leaching of nutrients and increasing weed problems. The experiment was carried out at three locations in Denmark: Jyndevad with a coarse sandy soil, Foulum with a loamy sand and Flakkebjerg with a sandy loam. It had two replicates for eight years (1997-2004) and included three factors: occurrence of nitrogen-fixing crops in a four-year crop rotation (rotation 1 (R1): 1½ year grass-clover + 1 year pulses, rotation 2 (R2): 1 year grass-clover + 1 year pulses, rotation 4 (R4): 1 year pulses), manure application with (+M) or without (-M) animal manure) and use of catch crops with (+CC) or without (-CC) undersown catch crops or bi-cropped clover). All crops in the rotation were present every year. Preventive as well as curative measures for weed management were used where possible in each system, e.g. mechanical weed control was not used after sowing of catch crops, and post-harvest stubble cultivations were only carried out in systems without catch crops. Annual and perennial weeds were monitored over 7 years. The experiments are described in Olesen et al. (2000), yields from the first 4 years are described in Olesen et al. (2002) and the weed development during the first rotation is described in Rasmussen et al. 1999a, b and Rasmussen et al. 2000.

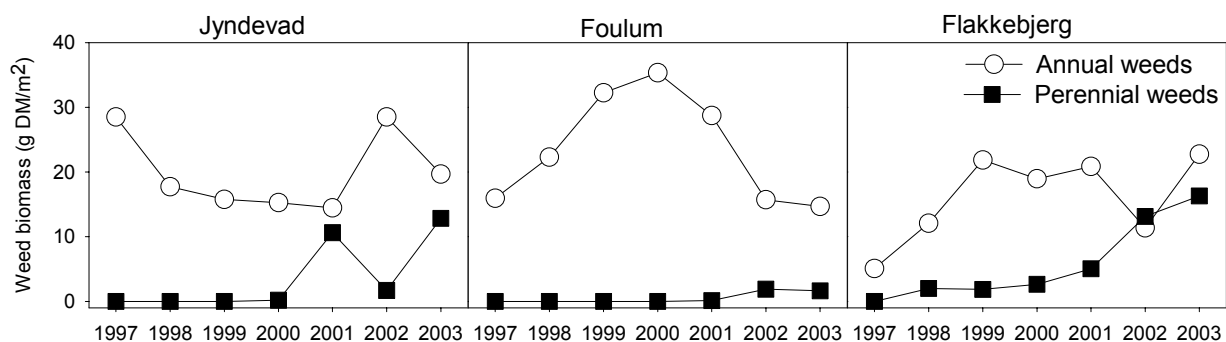


Figure 1. Development of annual and perennial weed biomass, mean of all treatments and crops at the three locations.

## Results and discussion

The general development of weeds, annual as well as perennial, during the experiment, differed between locations (Fig. 1). At Foulum the annual weed biomass was higher in the years 1999-2001 than the other years. There were two reasons for this: i) the winter cereals harvested in 2000 and 2001 were sown early the previous autumn, resulting in higher weed pressure, and ii) gradually increasing weed pressure in the spring crops was countered by

increased mechanical weed control from 2001 onwards. At Jyndevad and Flakkebjerg the amount of perennial weed biomass increased towards the end of the experiment in spite of efforts to counter this development.

Weed biomass as well as the proportion of weed biomass to total biomass was greatest in the +M treatment at Foulum and Flakkebjerg, while no effect on biomass and a reduced proportion of weeds in total biomass was seen at Jyndevad. This indicated that the crops benefited more than the weeds from the addition of manure on this sandy soil.

Weed biomass was greater in the winter cereals than in the spring-sown crops. Especially at Foulum there was more weed biomass in the –CC treatments, where weed control was less intensive, than in the +CC treatments. This difference between the catch crop treatments was also seen in some spring cereals at all three locations, but not in the pulse crops, and not in spring barley with undersown ley where no or the same weed control measures were carried out in all treatments.

In R2, which included grass-clover, there was significantly less thistle (*Cirsium arvense* L.) biomass than in R4 without grass-clover at Flakkebjerg, where *C. arvense* was prolific. This was most obvious in winter wheat that followed the grass-clover; no difference was found between the crop three years after grass-clover and the crops in R4 (Fig. 2a). Surprisingly, there were no significant differences between the amount of *C. arvense* in the –CC and +CC treatments, in spite of the fact that stubble cultivations were only carried out in the –CC treatment (Fig. 2b). Control of *Elytrigia repens* (L) Desv. ex Nevski at Jyndevad was carried out by repeated stubble cultivations, but this led to increased nutrient leaching and decreased fertility in the system.

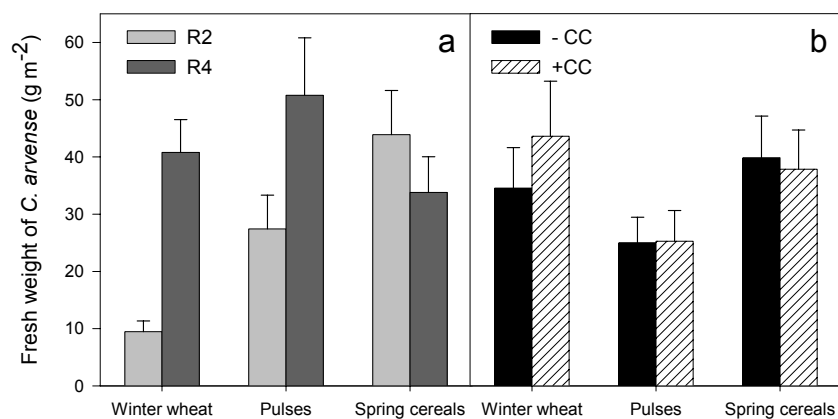


Figure 2. Fresh weight of *Cirsium arvense* in various crops in a. rotation 2 (R2) and 4 (R4). b with (+CC) or without (-CC) catch crops.

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# Reduction of the weed seedbank and manual weed control by preventing weed seed production during seven years

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## Introduction

In organic farming in the Netherlands, the required amount of manual labour is hardly available and the costs are high. Farmers want to reduce this labour input. Therefore, the relationships between the efficacy of weed management strategies, the required amount of hand weeding over the years, and the size of the weed seedbank need to be clarified. Specific goals of this research were (1) the comparison of the effects of three different weed management strategies on the required input on hand weeding over several years and (2) the comparison of the effects of these strategies on the weed seedbank in the soil.

## Materials and methods

The efficacy of three weed management strategies was studied and compared in a 7-year rotation. The three strategies were (1) control of weeds as carried out in standard organic farming practice, (2) prevention of all weed seed production in the field and (3) control of all residual weeds that grow above the crop. Thus, strategies 2 and 3 involved additional manual weed control, on top of the mechanical and manual weed control carried out in strategy 1. Field research has been carried out during 7 years at the organic experimental farm “Lovinkhoeve” in the Netherlands on seven fields. This farm was transferred from a conventional to an organic farm in 1996. The crop rotation scheme was: lucerne, sugar beet, winter wheat, spring wheat, potato, maize and tulip. Plots were 12 m x 40 m and were replicated twice on each field. Each year the required manual labour was determined for each strategy. At the start of the research (1996) and at the end of the experiments (spring 2003), the size of the weed seedbank was determined for the 0-25 cm top soil layer.

## Results and discussion

### *Manual weed control*

The additional manual weeding in strategy 2, compared to strategy 1, represents the extra effort to prevent all weed seed production, compared to the usual weed control effort on the organic farm. The linear regression equation of the required extra manual effort, expressed as additional manual weed control (h) over the years is:

$$\text{extra effort} = 53 - 7.45 * \text{years} \text{ (Figure 1).}$$

These results suggest that on average the extra effort to prevent all weed seed production, will be reduced to zero after 7 years. After this period, the required amount of manual weed control with strategy 2 will probably be lower than at present on organic farms (strategy 1). There was no difference in the amount of hand weeding between strategies 1 and 3.

### *Seedbank*

In spring 2003, the size of the weed seedbank was assessed and compared with the size in 1996 for each strategy. The number of seeds in the seedbank, after strategy 1 (reference organic farming) and strategy 3, was tripled. If weed seed production was prevented during 7



years, the size of the seedbank could be maintained at the same level as resulted from the earlier period of conventional farming with chemical control (table 1).

Table 1. Size of the weed seedbank before and after seven years of different weed management strategies. Strategy 1: standard organic farming practice, 2: prevention of all weed seed production, 3: removal of weeds growing above the crop. Seeds were collected from the 0-25 cm top soil layer.

Strategy	Size of seedbank (#/m <sup>2</sup> )	
	1996	2003
1	3806 <sup>a*</sup>	11758 <sup>b</sup>
2	5909 <sup>a</sup>	5724 <sup>a</sup>
3	3814 <sup>a</sup>	10830 <sup>b</sup>

\*) different characters indicate significant differences.  $F_{pr}=0.024$ . Lsd= 4473

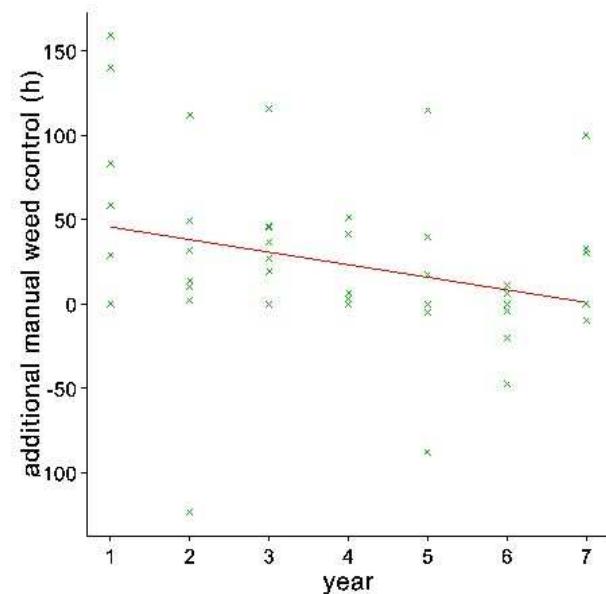


Figure 1. Extra manual effort required to prevent all weed seed production above the weed control in standard organic farming practice over multiple years.  $F_{pr}=0.038$ .

The results of this experiment suggest that it is possible to reduce the required labour input in weeding by preventing all weed seed production. Complete prevention of weed seed production resulted in a reduction of manual weed control effort over time, reaching the level of the standard manual control effort after 7 years. This effort may even become lower after a longer period. We could not demonstrate that this reduction in required hand weeding was linked to a decrease in the weed seedbank. A similar stability after prevention of seed production was found by Roberts and Neilson (1981). In our study, we can not guarantee that the aim of fully controlling all weeds that might produce seeds, was completely accomplished in the plots of strategy 2. Though machinery was carefully cleaned when moving from one plot to the other, in general import of seeds from the surroundings can not be fully excluded either. However, an analysis with a simple population model suggests that the number of germinating seeds, of which the seedlings were controlled, was small compared to the intrinsic spatial variability in total seedbank size. We hypothesize that the decline in required hand weeding is linked to a decrease in those weed seeds that form the transient part of the seed bank (Thompson and Grime, 1979).

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## Cruciferous weeds of canola in Iran.

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### Introduction

Annual weeds Cruciferous weeds are considered to be the most important weeds in canola, in that they affect yield and quality of canola. Taxonomically they are in the same family as canola and hence are difficult to control selectively in the crop. In Canada, the presence of *Sinapis arvensis* at 20 plants/m<sup>2</sup> resulted in 5% contamination of canola seeds (Anonymous, 1996). Likewise, early-season competition with *Sinapis arvensis* was most severe (Blackshaw, 1987). Several Cruciferous weed species have been observed in Canola crops in Iran. Suitable herbicides for controlling Cruciferous weeds, including *Rapistrum rugosum*, *Raphanus raphanistrum* and *Sinapis arvensis* have been investigated (Shimi 2001, 2004).

### Material and methods

To identify, determine density and study phenology of Cruciferous weeds in canola fields, an investigation was carried out in ten provinces in Iran (Tehran, West Azarbayjan, Khorasan, Bandar Abbas, Semnan, East Azarbayjan, North of Khouzestan, Fars, Golestan, Zanzan.) in two cropping years (2001 – 2002). Weed densities were determined by sampling quadrats in a W pattern at monthly intervals in each province and relative weed density expressed for each species recorded as a percentage of the total.

### Results and discussion

Results showed that the relative density of Cruciferous weeds was 1.8% in Bandar Abbas, 3.8% in the north of Khouzestan, 10% in Zanzan 16.1% in Semnan, 18.9% in west Azarbayjan, 20.4% in Khorasan, 22.2% in Golestan 35.1% in Tehran, 35.5% in Fars, 37.7% and in east Azarbayjan. The mean for the 10 Provinces was, 20.1%. Cruciferous weeds in each province were :

Tehran, *Descurainia sophia*, *Cardaria draba*, *Rapistrum rugosum*, *Goldbachia laevigata*, *Erysimum repandum*, *Capsella bursa – pastoris*, *Sisymbrium loeselii*, *Sisymbrium irio*, *Sinapis arvensis*.

West Azarbayjan, *Cardaria draba*, *Conringia orientalis*, *Raphanus raphanistrum*, *Sinapis arvensis*, *Capsella bursa – pastoris*, *Goldbachia laevigata*, *Descurainia sophia*, *Thlaspi arvensis*, *Lepidium latifolium*, *Brassica nigra*.

Khorasan, *Rapistrum rugosum*, *Goldbachia laevigata*, *Euclidium syriacum*, *Descurainia sophia*, *Eruca sativa*, *Cardaria draba*, *Sisymbrium irio*, *Malcolmia africana*, *Sinapis arvensis*.

Bandar Abbas, *Malcolmia africana*, *Eruca sativa*, *Descurainia sophia*.

Semnan, *Malcolmia africana*, *Descurainia sophia*, *Cardaria draba*, *Eruca sativa*.

East Azarbayjan, *Rapistrum rugosum*, *Goldbachia laevigata*, *Raphanus raphanistrum*, *Sisymbrium irio*.

Khouzestan, *Sinapis arvensis*, *Descurainia sophia*.

Fars, *Sinapis arvensis*, *Cardaria draba*, *Rapistrum rugosum*, *Hirshfeldia incana*, *Descurainia sophia*, *Neslia apiculata*, *Erysimum repandum*

Golestan, *Sinapis arvensis*, *Sinapis alba*, *Brassica sp.*, *Capsella bursa – pastoris*, *Raphanus raphanistrum*.

Zanzan, *Cardaria draba*, *Sinapis arvensis*.

The relative density of Cruciferous weeds in East Azarbayjan , Tehran and Fars was greater than in other Provinces ( Figure 1). In Fars, *Sinapis arvensis* was the dominant Cruciferous weed but in East Azarbayjan it was not observed. This species found only in one of the visited localities of Tehran in 2.4 %. However, Fars was the important Province of Iran for high density of Cruciferous weeds especially *S. arvensis*. *S. arvensis* was not observed in Semnan, East Azarbayjan and Bandar Abbas. The north of Khouzestan and Bandar Abbas had the lowest density of Cruciferous weeds .

Dominant Cruciferous weeds in Iran were: *Descurainia sophia* , *Cardaria draba* , *Rapistrum rugosum* , *Erysimum repandum* , *Capsella bursa – pastoris*, *Goldbachia laevigata*, *Sisymbrium loeselii*, *Sisymbrium irio*, *Conringia orientalis* , *Eruca sativa*, *Alyssum strigosum*, *Brassica nigra*, *Malcolmia africana*, , *Raphanus raphanistrum*, *Sinapis arvensis*.

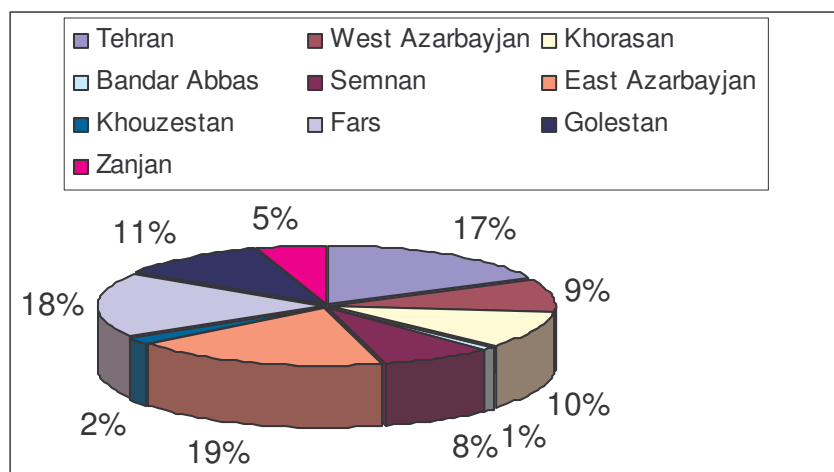


Figure1. Comparison of Cruciferous weeds densities in ten provinces.

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# Identification , density and phenology of dominant weeds of canola in Tehran Province

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## Introduction

Weeds are one of the most important factors that affect yield and quality of canola since canola is susceptible to weed interference (Daugovish *et al.* 2003). In order to develop the most appropriate methods of weed control it is necessary to identify species present, determine their density and understand their phenological development especially during the critical period of weed competition. Martin *et al.* (2001) showed that after the four – to six-leaf stage of crop development, few weeds emerged, and late-emerging weeds accumulated little shoot biomass. In Iran, canola is planted in autumn and hence susceptible to competition from both winter and spring emerging weeds. Shimi (2001, 2004) showed that different species of dicot and monocot weeds exist in canola fields in the Karaj area (30 km W of Tehran).

## Material and methods

In order to identify weed species composition, determine their density and investigate the phenology of weeds in canola, an investigation was carried out in four localities of Tehran province, namely south of Tehran, Nazar Abad, Hashtgerd and Varamin in each of two years (2001 – 2002). Weed densities and phenology were determined by quadrat sampling method in a W pattern on a monthly basis. Weeds were divided into three groups, Graminae, Cruciferae and other dicotyledonous species. Winter canola was planted in early autumn and harvested in late spring in Tehran province.

## Results and discussion

Results showed that the relative density of each of the groups of weeds (Graminae, Cruciferae and other dicot weeds) was 22.6 , 46.2, 31.2 % in south of Tehran, 4.1, 37.9, 42.0 % in Nazar Abad, 15.3, 35.7, 49.0 % in Hashtgerd , 6.2, 20.5, 73.3 % in Varamin, respectively. The mean for Tehran Province was 12.1, 35.1, 48.9 %. The relative density of Cruciferous weeds south of Tehran was greater than the other localities. *Sinapis arvensis* was only recorded in Varamin with a relative density of 2.4 %. Dicot weeds were dominant to other groups. The greatest density and diversity of weeds was in the spring at which time most were at the reproductive stage while canola was at the stem elongation stage. *Goldbachia laevigata* , *Erysimum repandum*, *Sisymbrium loeselii* , *Sisymbrium irio*, *Conringia orientalis* *Convolvulus arvensis*, *Rapistrum rugosum*, *Sonchus* spp , *Vicia vilosa* , *Descurainia Sophia*, *Capsella bursa-pastoris* and most of grass weeds were considered troublesome at harvest time because their seeds contaminated canola seed .

Table 1. Relative density (%) of dominant weeds in canola fields of Tehran province.

Weeds	Relative density ( % )	Family
<i>Goldbachia laevigata</i>	5.25	Brassicaceae
<i>Descurainia sophia</i>	13.9	Brassicaceae
<i>Capsella bursa-pastoris</i>	6.32	Brassicaceae

<i>Rapistrum rugosum</i>	2.15	Brassicaceae
<i>Erysimum repandum</i>	2.95	Brassicaceae
<i>Cardaria draba</i>	1.42	Brassicaceae
<i>Sinapis arvensis</i>	0.6	Brassicaceae
<i>Sisymbrium loeselii</i>	-	Brassicaceae
<i>Sisymbrium irio</i>	-	Brassicaceae
<i>Galium tricornutum</i>	2.15	Rubiaceae
<i>Lamium amplexicaule</i>	12.65	Labiatae
<i>Amaranthus retroflexus</i>	3.7	Amaranthaceae
<i>Sonchus spp</i>	7.35	Compositae
<i>Euphorbia helioscopia</i>	3.45	Euphorbiaceae
<i>Centaurea depressa</i>	2.4	Compositae
<i>Vicia vilosa</i>	2.2	Leguminosae
<i>Silene conoidea</i>	1.82	Caryophyllaceae
<i>Adonis aestivalis</i>	0.55	Ranunculaceae
<i>Veronica persica</i>	2.35	Scrophulariaceae
<i>Anchusa iranica</i>	2.95	Boraginaceae
<i>Malva neglecta</i>	2.4	Malvaceae
<i>Convolvulus arvensis</i>	3.4	Convolvulaceae
<i>Acroptylon repens</i>	-	Caryophyllaceae
<i>Anchusa ovata</i>	-	Boraginaceae
<i>Avena ludoviciana</i>	2.95	Poaceae
<i>Sorghum halepense</i>	0.4	Poaceae
<i>Echinochloa crus-galli</i>	0.75	Poaceae
<i>Bromus tectorum</i>	2.7	Poaceae
<i>Hordeum glaucum</i>	0.95	Poaceae
<i>Phalaris minor</i>	0.67	Poaceae
<i>Bromus danthoniae</i>	3.75	Poaceae
<i>Muscari neglectum</i>	4	Liliaceae

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# Seed predation by carabids in organic wheat fields in the Netherlands

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## Introduction

The life cycle of weeds can be divided into four stages: seedling, adult, seed dispersal and seed storage in the seedbank. The population size can be manipulated in each stage in the life cycle with variable success. For substantial reduction of weed populations only 25-50% seed kill is needed, while almost 98% of the adult plants need to be removed prior to seed dispersal for the same effect (Medd and Ridings, 1989). Isolated individual plants that survive weed control may produce vast quantities of seeds due to the absence of competitors (Leguizamón and Roberts, 1982). Therefore, weed control methods that increase seed kill are potentially very effective. This is particularly important in organic cereals, which are a problematic crop in terms of weed control.

The results of investigations on seed predation varied greatly between regions and crops because of different methodologies, experimental period or seeds used, such that observed predation oscillated between 18-78% (Harrison et al., 2003; Westerman et al., 2003b).

Potential seed predators include many vertebrate and invertebrate species such as mice, birds, ants, carabid beetles and slugs. Westerman et al. (2003a) concluded that rodents removed the greatest quantity of the seed from wheat fields in the Netherlands. Marino et al. (1997) found both invertebrates and vertebrates to be important seed predators in the United States. Honek et al. (2003) found carabid beetles to be most important invertebrate seed predators in the Czech Republic. Regardless of their relative importance for seed kill, different predators may also have different spatial and temporal patterns of predation (Marino et al., 1997) and different preferences for seed species and size (Mittelbach and Gross, 1984). Therefore all types of predators need to be studied in more detail to estimate their effect on the life cycle of weed species.

## Methods

This paper focuses on post-dispersal seed predation by carabid beetles (Coleoptera: Carabidae) in organic winter wheat fields in the Netherlands. The aim was to investigate the temporal and spatial variation in seed predation. Two preferred (*Capsella bursa-pastoris* and *Stellaria media*), two non-preferred (*Veronica persica* and *Lamium amplexicaule*) and one seed of unknown preference (*Poa annua*) were used. Fifty seeds of one species were glued on cards (4x9 cm) of sandpaper, using 3M SprayMount™ glue (Westerman et al. 2003a). Groups of five seed cards (one card per seed species) were exposed in seven transects within each of six locations: grass boundary, field edge, 4, 11, 24 and 49m from the field edge, in two organic fields of winter wheat near Wageningen, the Netherlands. Each group of cards was protected with a cage in order to prevent rodents from eating seeds. Two pitfall traps were placed close to cages in order to monitor activity of carabids. A further five sets of cards were wrapped in envelopes of fine mesh to exclude invertebrate access. These served as controls. At weekly intervals missing seeds were counted and all cards were replaced. IRREML analysis in Genstat 6.0 was used for data analysis. The numbers of remaining seeds were used as a response variable. The analysis was done in two steps: (i) the data from each field were compared with the controls to establish if there is significant seed predation; (ii) if there was significant predation, the effect of treatments (fields (as blocks), distance of the stand from the field edge, seed species and time duration (measured in weeks) on seed loss was tested.

## Results and Discussion

Seed loss varied between weeks. In some weeks, seed loss was not significant compared to control seed cards. Seed predation was significant for 12 out of 18 weeks. This was probably correlated with the activity of invertebrate seed predators, mainly carabid beetles. Activity of invertebrates varies with temperature (Honek, 1988), therefore in weeks with low temperature carabid activity and hence seed predation was low. Heavy rain negatively influenced significance of the results as it increased background seed loss. This was especially important in weeks with less predation.

Seed predation also varied between seed species. *C. bursa-pastoris* and *P. annua* were most predated. High seed predation of the former species was expected, as it was one of the most widely eaten seed species in laboratory trials (Honek et al., 2003). High consumption of the latter was also observed by Tooley et al. (1999). *S. media* was occasionally eaten but on average its consumption was low in contrast to results of Honek et al. (2003). *V. persica* and *L. amplexicaule* were not preferred, but the background seed losses were also highest.

Seed predation was greatest close to the field margin, and decreased with increasing distance towards the field interior. This was as expected and was probably caused by greater density and activity of ground beetles near to the field margin. Increased activity of carabids close to the field margin was reported by Thomas et al. (2000) and may correlate with microclimate resulting from higher weed density, sparse crop and greater food availability.

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# Weed emergence dynamics in Italian wheat crops: influence of sowing time and stale seedbed preparation

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## Introduction

Weed pressure in Italian wheat crops (and winter cereals in general) has increased in recent years, especially by the most troublesome weed species like grasses, *Papaver* spp. and *Galium aparine* (Montemurro *et al.*, 2000). This trend can be ascribed to three main reasons: wider use of monoculture (or very limited crop rotation), earlier crop sowing due to the adoption of high-producing varieties with a long life-cycle, and increasing herbicide resistance problems. In this changing agronomic situation, many farmers have continued to make their weed control treatments, mostly with herbicides, during the end of winter-early spring when weeds are larger and less susceptible to herbicides.

Successful weed management, based on integrated weed control techniques, relies on knowledge of the weed emergence dynamics of the key species. This is particularly important in planning integrated control strategies for dense and/or herbicide resistant weed populations (Sattin *et al.*, 2001).

Little is known about the time of weed emergence in Italian wheat crops. With the aim of filling this information gap, in 1999, a multi-site, pluri-annual field research project began looking at the influence of wheat sowing time and mock stale seed-bed preparation on weed emergence dynamics and weed infestation.

## Materials and methods

Sixteen field experiments have been made over five seasons in the major Italian wheat growing areas. They can be grouped as follows;

A) Seven experiments (four in southern and three in central Italy) to determine weed emergence dynamics, six of which considered the most important grass weeds (*Lolium* spp., *Avena sterilis*, *Phalaris paradoxa*) in autumn-sown durum wheat crops. One experiment in central Italy monitored all the major weeds of autumn sown bread wheat. Six to twelve (depending on the level and uniformity of infestation) steel rectangular quadrates (12 cm x 50 cm) were randomly fixed on the ground of each experimental field, positioned across the wheat rows. Seedlings within each quadrate were counted and removed every 15-20 days starting from mid-November to tillering stage (BBCH-scale 22-28).

B) Two experiments were made in central Italy to acquire information on the influence of durum wheat sowing time (four dates) on weed infestation. The soil was tilled (5-10 cm deep) just before each sowing time.

C) Seven experiments, in the absence of the crop (durum or winter wheat), simulating the influence of delay in sowing, after stale seedbed preparation, on weed infestation. Two experiments were located in southern Italy, three in central and two in the north. The soil was tilled once in early autumn and glyphosate was applied (1100-1450 g p.a. ha<sup>-1</sup>) at different times to kill any vegetation present. The layout for experiments of groups B and C was a randomised block design with three or four replicates, plot size was 15 m<sup>2</sup> (3 m x 5 m). The final level of infestation was evaluated through a visual estimate of the ground cover and/or weed counts in 3 steel rectangular quadrates (12 cm x 50 cm) randomly fixed on the ground of each plot.

## Results and discussion

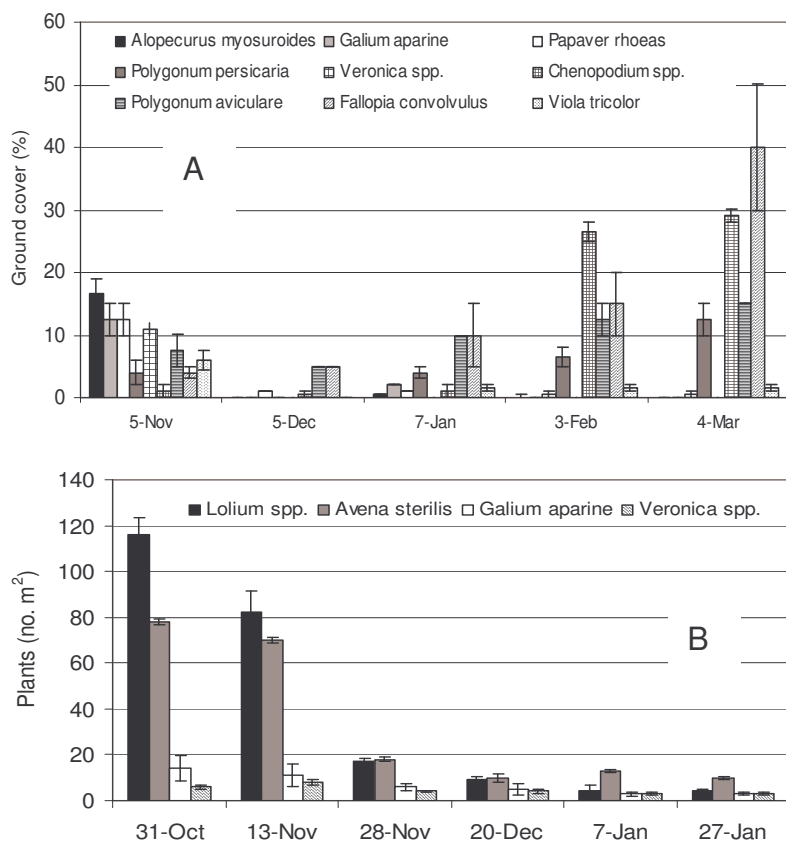


Figure 1. Simulated effect of delay of sowing, coupled with stale seedbed preparation, on the weed infestation evaluated in mid-May. Vertical bars represent standard errors (S.E.). A = average values of two experiments in north-western Italy (Po valley).

B = values from one experiment in central Italy.

The results from all experiments indicate that, providing the soil is moist enough and temperature is suitable for germination, the most troublesome weeds (all grasses plus *Galium aparine* and *Papaver* spp.) emerge before mid-January and mid-November in southern and northern Italy, respectively. This happens even in the absence of the crop (Fig. 1). Potentially, i.e. in the absence of the crop, several dicotyledenous weeds can emerge from the end of January onwards in the Po valley (Fig 1A) and in some central areas.

Stale seedbed preparation significantly reduced the pressure derived from the most troublesome autumn-emerging weed species (*Lolium* spp., *A. sterilis*, *P. paradoxa*, *P. rhoeas*, *G. aparine*), especially in the centre and south (Fig. 1B). This technique appears particularly effective where dense populations of herbicide resistant grasses infest the crop, when weed density can be reduced by up to 80% (Gasparetto *et al.*, 2003). However, stale seedbed preparation is sometimes rendered difficult because the soil is too wet in late autumn.

For weed management there is an optimum sowing time. When this optimum time is coupled with a stale seedbed preparation it minimises the infestation of the potentially most dangerous autumn-emerging weeds, whilst also producing a crop competitive enough to minimise the damage caused by spring-emerging weeds and yield loss due to delayed sowing. This data set suggests that the end of October and from mid-November to early December are the most suitable wheat sowing times in the north and south of Italy, respectively.

The experiments simulating, without the crop, the influence of the delay of crop sowing after stale seed-bed preparation, also indicate that early post-emergence herbicide treatments (e.g. February) are preferable because they are more effective (sometimes a lower herbicide dose can be applied) without risking a late emergence of competitive weeds (e.g. grasses).

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# Why is reproductive potential high in weed plants?

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## Introduction

There exists a trade-off between two groups of plants. The first group is flowering only once in their life, and/or plants with a short life cycle, that have a tendency to produce copious amounts of small seeds. On the contrary, the second group has a long life cycle usually producing a small quantity of large seeds (Baker 1972, Harper 1977).

Weed plants growing in cultivated fields usually have annual life cycles (*Avenella flexuosa*, *Chenopodium album*, *Lamium purpureum*, *Papaver rhoeas*, *Agrostemma githago*, *Stellaria media*, etc). Annual life cycle is usually connected with considerable production of diaspores, persistent seedbanks, and protracted seed dormancy. This is the reason, why some annual plants are very obstinate weeds.

Is it true, that all weeds produce a lot of seeds? There are many weed plants in which vegetation propagation is of greater importance than seed fecundity (*Elytrigia repens*, *Stachys palustris*, *Aegopodium podagraria*, etc). Whereas, many weeds appear as annuals, they are able to complete more than one life cycles per one annum (*Poa annua*, *Stellaria media*, *Veronica persica*, etc.), they probably produce more seeds per annum than “normal annual” plants. There are another two types of life cycle: biennial and monocarpic perennials. Plants with biennial or monocarpic perennial life cycles flower mostly once only in its life cycle. Which reproductive strategy do such plants have? It produces more or less seeds than annual plant? Which model of weed reproduction exists? Answers to such questions are important for understanding the reproductive strategy of weeds enabling better weed control.

## Materials and methods

All the data about weed characteristics (especially number of seeds) were obtained from Appendix of the work Šerá & Šerý 2004 ([www.ibot.cas.cz/folia/index.htm](http://www.ibot.cas.cz/folia/index.htm)) and processed with common mathematical and statistical procedures.

## Results and discussion

Monocarpic plants produce more seeds per growing season than polycarpic plants (Harper 1977, Šerá & Šerý 2004). In addition, annual plants produce more seeds than perennials, the number of seeds produced by biennials and monocarpic perennials is comparable, and annuals and monocarpic perennials have on average lighter seeds than polycarpic plants (Baker 1972, Silvertown 1981, Šerá & Šerý 2004). A similar trend was found in weed plants, but some differences were not significant (small number of compared species).

Monocarpic life cycle is typical for weed plants and R-strategists (Grime 1977, 2001). A high number of small seeds is their adaptation to dispersal in space and time (Southwood 1988). Species which invest into reproduction only once in their lives have a strategy to produce more seeds than species with repeated flowering throughout (Salisbury 1942). The repeated

flowering is connected with balance among investments energy to somatic growth, survival, and reproduction.

Persistence, vitality, and competitive ability of individuals are important for a successful establishment of weed populations in the field. The large output of small seeds of great dispersive quality or persistence within the soil seed bank is a sure guarantee for a weed-infested field.

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## Assessment of allelopathic effects of debris of *Ageratum conyzoides*

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### Introduction

Allelopathy is fast being recognized as one of the novel mechanism adopted by invasive species to successfully establish in an alien environment at the cost of native communities (Bais et al., 2003). In this context, *Ageratum conyzoides* L. (Asteraceae), an invasive aromatic weed of tropical America, is no exception. It has invaded several tropical and subtropical parts of the world including India. Being fast growing with high reproductive potential, *A. conyzoides* soon dominates the area upon invasion forming its own kingdom and replacing the native vegetation. As a principal weed of arable land and pastures, it reduces crop yields both qualitatively and quantitatively and causes fodder scarcity. Allelopathy seems to be the major reason of its dominance in invaded ecosystems as some reports point towards this (Kong et al. 2002, Singh et al., 2003). A number of allelochemicals, including water-soluble phenolics and volatile essential oils rich in mono- and sesquiterpenes, have been reported from the weed (Kong et al. 2002, Xuan et al., 2004). However, the reports on its interference with crops are still inadequate and studies are required to determine the extent of phytotoxicity towards a wide range of crops. Therefore, a study was planned to assess the effect of *A. conyzoides* debris (plentiful owing to large weed biomass) on the early growth of three winter season crops – *Triticum aestivum* (wheat), *Brassica campestris* (mustard) and *Raphanus sativus* (radish) – that are grown during the active growth period of this weed.

### Material and methods

Debris of *A. conyzoides* was collected from the plants growing wildly in the agricultural fields. Certified seeds of *Triticum aestivum* (var. HD 2329), *Brassica campestris* (var. Pusa Bold) and *Raphanus sativus* (var. Pusa Chetki) were used for the growth studies. Aqueous extracts were prepared by dipping 10 g of *A. conyzoides* debris in 500 ml of distilled water for 16 h followed by decantation and filtration through a Whatman paper no. 1 so as to get a 2% aqueous extract. It was further diluted to 0.5 and 1.0 % and their effect was studied on the early seedling growth of test crops in a laboratory bioassay. Further, the debris was amended into garden soil at the rate of 5, 10 and 20 g per 1000 g of soil. Amended soils were filled in 250 ml polystyrene cups and used for growth studies under greenhouse conditions. Debris aqueous extracts and amended soils were analyzed for the presence of phytotoxic phenolics using Folin-ciocalteu reagent as per the method of Swain and Hillis (1959).

### Results and discussion

The results indicate an appreciable reduction in the seedling length and dry weight of all the test crops in response to debris extracts of *A. conyzoides* (Table 1). The inhibitory effect increased with increasing concentrations of extracts. Among the three crops growth of radish was affected more than others (Table 1). The growth retardatory effect of extracts indicates the presence of some phytotoxins in debris that are probably released through leachation. To further establish this, debris was amended in the garden soil and its effect was studied on early growth of test crops. An inhibitory effect on early growth and dry weight of test crops was observed in amended soils (Table 2) thereby indicating that putative allelochemicals are released from the debris, accumulate in the soil and affect the growth of test crops. The study therefore demonstrates that debris of *A. conyzoides* contains some inhibitory chemicals which deleteriously affect the growth of test crops.

Table 1. Effect of *A. conyzoides* debris extracts on the seedling growth (after 1-week) of test crops.

Concentration (%, w/w)	<i>T. aestivum</i>		<i>B. campestris</i>		<i>R. sativus</i>	
	SL	DW	SL	DW	SL	DW
0 (Control)	18.3a	15.2a	17.5a	8.3a	16.6a	12.7a
0.5	15.7b	12.8b	15.8b	7.0b	14.6b	10.4b
1.0	12.3c	10.0c	10.8c	6.3c	10.0c	9.0c
2.0	8.8d	7.4d	7.1d	4.0d	6.8d	6.3d

SL: Seedling length (cm); DW: Dry weight (mg).

Different alphabets in a column indicate significant difference at P<0.05.

Table 2. Growth (after 2-weeks) of test crops in *A. conyzoides* debris amended soils.

Treatment (g / kg soil)	<i>T. aestivum</i>		<i>B. campestris</i>		<i>R. sativus</i>	
	SL	DW	SL	DW	SL	DW
0 (Control soil)	20.1a	17.6a	18.1a	11.7a	17.1a	18.3a
5	17.3b	15.7b	15.1b	11.0a	14.2b	13.3b
10	14.8c	13.3c	11.6c	9.0b	12.1c	11.0c
20	10.1d	9.8d	8.0d	6.4c	8.5d	8.3d

SL: Seedling length (cm); DW: Dry weight (mg).

Different alphabets in a column indicate significant difference at P<0.05.

To find out the nature and content of phytotoxins involved, the amount of water-soluble phenolics was determined since these are ubiquitously found and often implicated in allelopathy (Rice, 1984). Phenolics are easily released through leaching as well as decomposition from the decaying plant parts and residues (Rice, 1984). The debris of *A. conyzoides* was found to contain appreciable amount of phenolics, which were not only present in aqueous extracts but also in amended soils (Table 3). The amount of the phenolics increased with the increasing concentration of extracts as well as amendment.

Table 3. Amount of phenolics in the *A. conyzoides* debris extracts and debris amended soil.

Concentration (%, w/v or w/w)	Debris extracts ( $\mu\text{g/ml}$ )	Debris amended soil ( $\mu\text{g/g}$ )
0 (Control)	-	14.81 $\pm$ 1.34
0.5	225.12 $\pm$ 12.62	19.15 $\pm$ 1.19
1.0	382.19 $\pm$ 14.65	25.17 $\pm$ 2.67
2.0	449.72 $\pm$ 10.60	38.33 $\pm$ 2.40

Values in a column significant from each other at P<0.05.

The study thus demonstrates that water-soluble phenolics present in debris of *A. conyzoides* are responsible for the observed growth inhibitory effect on the test crops.

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# Weed seedbank after lambsquarter (*Chenopodium album*) burning

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## Introduction

Management methods to control herbicide-resistant populations combine cultural control with the use of alternative herbicide. Cultural practices include delayed sowing, stubble burning, increased crop density and capture of weed seeds at harvest (Gill & Holmes, 1997). In Portugal, *Chenopodium album* resistant to atrazine were identified in a reduced tillage maize field sprayed with atrazine+metolachlore for a period of 10 years (Calha & Rocha, 2004), where the maize alternated with a potato crop (but not during the last four years). To destroy the large infestation of *C. album* at harvest time, the farmer burned the crop and the weed. The aim of this study was to evaluate the effect of burning on the germination of seeds in the weed seedbank and on the behavior of atrazine resistant *C. album*.

## Materials and methods

Two sets of soil (ancient alluvium) samples were taken. The first was taken in October 2003, just after crop harvest, the second four months after crop burning (February 2004). Each set of samples comprised of 100 soil sampling units of 93.75 cm<sup>3</sup> which were systematically taken at 5 m intervals throughout the sampling area. Samples were placed in a glasshouse with sprinkle irrigation to encourage germination in plates with 10 cm diameter. Seedlings were identified in a total area of 0.785 m<sup>2</sup> (100 x 78.5 cm<sup>2</sup>). Each sample was examined separately and the seedlings of the weed species were identified and counted weekly (Sousa *et al.*, 2003). Ten percent of *C. album* seedlings that had emerged from the weed seedbank were analyzed for atrazine resistance with a portable fluorimeter (PEA – Hansatech). The relative fluorescence VJ [VJ=FJ-F0)/(FM-F0)] was used to compare chlorophyll fluorescence from resistant and susceptible plants after an incubation period of 17 h in 10<sup>-4</sup> M atrazine.

## Results and discussion

Thirty-one species were identified in the weed seedbank flora belonging to 19 families with a prevalence of broadleaved weeds. The number of species found before and after burning ranged from 22 to 23, respectively (Table 1). Twelve species had emerged in both situations; *Chenopodium album*, *Stellaria media* and *Urtica membranacea* decreased after burning, whilst the high temperatures favored the germination of monocotyledons such as *Poa annua*, *Setaria verticillata*, *Digitaria sanguinalis* (Gramineae) and *Juncus bufonius* (Juncaceae). Seed density of *Solanum nigrum*, *Conyza albida*, *Stachys arvensis* and *Oxalis corniculata* did not differ between the two treatments. *Chenopodium album* was the most abundant weed species before burning, whilst *P. annua* was the second most common after burning followed by *J. bufonius*. The sample of *C. album* plants analyzed from the seedbank, both before and after burning, were confirmed to be resistant to atrazine as compared to the wild type (VJ values ranged from 0.85 ± 0.03 and 0.85 ± 0.02). This is in agreement with the high level of infestation of *C. album* emerged in the above-ground flora. Such a large seed stock could be attributed to the considerable seed rain that occurred during the most recent maize crop cycle because of the lack of control caused by resistance. There are few studies of herbicide resistance in the weed seedbank; the frequency of the susceptible biotype in the weed seed stock and seed longevity are both important factors to reduce resistance. According to Barberi *et al.* (1998) *C. album* seed stocks at deeper soil levels (20-30 cm) are high, which is related

to the high seed longevity of this species (Barralis *et al.*, 1988). These studies will be continued to assess atrazine resistance the in the *C. album* seedbank at deeper soil levels.

Table 1. Weed species seedling density before and after crop burning

Species	Density (seedlings/m <sup>2</sup> )	
	before burning	after burning
<i>Amaranthus retroflexus</i> L. (AMARE)	14.01	14.01
<i>Anagallis arvensis</i> L. (ANAAR)	-	2.55
<i>Chamaemelum fuscatum</i> (Brot.) Vasc. (ANTPR)	-	1.27
<i>Chamaemelum mixtum</i> (L.) All. (ANTMI)	-	2.55
<i>Chenopodium album</i> L. (CHEAL)	3239.49	1633.12
<i>Chrysanthemum segetum</i> L. (CHYSE)	1.27	-
<i>Conyza albida</i> Spreng. (ERIFL)	5.10	8.92
<i>Conyza bonariensis</i> Cronq. (ERIBO)	1.27	-
<i>Datura stramonium</i> L. (DATST)	1.27	1.27
<i>Digitaria sanguinalis</i> (L.) Scop.(DIGSA)	7.64	11.46
<i>Echinochloa crus-galli</i> (L.) P. Beauv. ((ECHCG)	2.55	-
<i>Echium plantagineum</i> L. (EHIPL)	-	1.27
<i>Euphorbia peplus</i> L. (EPHPE)	-	8.92
<i>Heliotropium europaeum</i> L. (HEOEU)	-	1.27
<i>Hypericum perforatum</i> L. (HYPPE)	1.27	-
<i>Juncus bufonius</i> L. (JUNBU)	649.68	1126.12
<i>Lamium purpureum</i> L. (LAMPU)	2.55	2.55
<i>Lolium rigidum</i> Gaudin (LOLRI)	2.55	-
<i>Lythrum hyssopifolia</i> L. (LYTHY)	-	1.27
<i>Oxalis corniculata</i> L. (OXACO)	3.82	6.37
<i>Poa annua</i> L. (POAAN)	1443.31	1825.12
<i>Pseudognaphalium luteum-album</i> (L.) Hilliard& B. L. Burt	-	1.27
<i>Rumex crispus</i> L. (RUMCR)	1.27	-
<i>Setaria verticillata</i> (L.) P. Beauv.(SETVE)	3.82	20.38
<i>Sinapis arvensis</i> L. (SINAR)	-	3.82
<i>Solanum nigrum</i> L. (SOLNI)	370.70	340.13
<i>Spergula arvensis</i> L. (SPEAR)	3.82	-
<i>Stachys arvensis</i> (L.) L. (STAAR)	3.82	7.64
<i>Stellaria media</i> L. (STEME)	216.56	99.36
<i>Trifolium campestre</i> Schreb. (TRFCA)	1.27	-
<i>Urtica membranaceae</i> Poir. (URTDU)	19.11	6.37

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# Weed seed and weed plant dynamics in arable fields in central Latvia

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## Introduction

The weed flora in five individual arable fields in central Latvia (Rīga, Cēsis, Valka, Valmiera, and Limbaži) was surveyed each year from 1994 to 2002 to investigate the dynamics of the flora in relation to cropping and crop husbandry practices. The methodology and the results for the plant surveys have been reported previously (Vanaga, in press). From 1997, soil samples for weed seed assessment were taken at the same time as the plant counts were recorded. This paper summarises the results of the weed seedbank determinations (potential flora) and compares them with those from the vegetation assessments (actual flora).

## Materials and methods

Soil samples to a depth of 5 cm were taken with a small scoop from 25 – 40 sampling points in each field at the beginning of July each year from 1997 to 2002. The field samples were thoroughly mixed and a 1 kg sub-sample taken to the laboratory where it was allowed to dry. The moisture content of a sub-sample of the air-dried soil was determined by further drying in an oven at 105 °C for 6 hours. Four sub-samples of the air-dried soil, each of 100 g, were washed separately on a nylon net of 0.25 mm square mesh. The retained material from each sub-sample was transferred to a filter paper and dried in a warm oven. Undamaged seeds were identified to species with the aid of a binocular microscope in comparison to a seed reference collection held by the Institute and pictorial reference texts. Seed numbers per square metre to a soil depth of 5 cm were calculated by the method of Доспехов *et al.* (1977) using a soil bulk density of 1.1 g cm<sup>-3</sup>. The seed and species numbers were transformed to log<sub>10</sub> (number + 1) and analysed by the same statistical techniques as the previously reported plant data.

## Results and discussion

The seeds of 40 weed species were identified in this survey, compared with 64 species of weed plants. Five species represented by seed occurred in all five fields (20 species of plants); 12 occurred in only one field (14 species of plants); with seed of only one species, *Chenopodium album*, recorded in all five fields in all six years. No species of plant was observed in all fields in all nine years. The highest number of seed species recorded in any field in any year was 15 (Valmiera 2002) (which contained 32 species of plant) and the lowest was three (three fields in 1998 and one in 1997) (containing two plant species). The highest density of seeds was 13 425 m<sup>-2</sup> (Limbaži 1997), of which 9 080 were *Chenopodium album* and 1 650 were *Tripleurospermum inodorum*. The lowest density of seeds was 2 201 m<sup>-2</sup> (Valmiera 1998). Nine species represented as seeds were not recorded in the actual flora. Seeds of most of these species occurred only once in one or two fields and in low numbers, but seeds of *Euphorbia peplus* were found in four years in the Limbaži field and once in the Valmiera field. Thirty three species were not recorded in the seedbank. Most of these species occurred in small numbers at a low frequency, but *Fumaria officinalis* was recorded in all five fields and on 25 of the 45 possible field-year observations. Other species that occurred fairly frequently as plants (18-19/45) but were not found as seeds were *Veronica arvensis*, *Artemisia vulgaris* and *Mentha arvensis*.

For statistical analysis the components of the seedbank were considered in the same three groups as the actual flora: annual dicots, perennial dicots and monocots.

Seeds of 29 species of annual dicots were identified (36 species of annual dicot plants): 5 occurred in all five fields; 11 species were found in only one field and 8 of these occurred only once. The most frequent species of annual dicot (as determined by frequency of occurrence in the 30 seedbank determinations) were: *Chenopodium album* (30), *Stellaria media* (21), *Tripleurospermum inodorum* (18), *Viola arvensis* (15) and *Polygonum lapathifolium* (15). These 5 species were the most abundant overall. *Chenopodium album* was the most abundant seed, with an average density of 4 111 seeds m<sup>-2</sup>. Although much less frequent, the next most abundant species were *Spergula arvensis* (frequency 5/30) with an average density of 902 seeds m<sup>-2</sup> where it was found and *Capsella bursa-pastoris* (frequency 1/30) with a density of 825 seeds m<sup>-2</sup>.

Seeds of perennial dicot and monocot species were recovered much less frequently than those of annual dicots. Seeds of only 8 perennial dicot species were recovered (compared with 23 in the actual flora, the most frequent of which, *Rumex crispus* and *Sonchus arvensis*, occurred in only six of the thirty samples. Three species of monocot seeds were recovered (5 species of plants): *Elymus repens* (4/30), *Poa annua* (4/30) and *Echinochloa crus-galli* (2/30).

In contrast to the actual flora, there were no statistically significant differences among fields in the mean numbers of seed species for any of the three weed groups or for all species combined. There were, however, significant differences among years in the mean number of species recorded as seeds, with the lowest numbers for all weeds groups occurring in 1998 and the highest numbers occurring in 2002. There were no comparable differences among years in the numbers of species recorded in the plant assessments between 1997 and 2002.

There were significant differences among fields in the mean numbers of seeds for all three weed groups and overall, but only between the lowest and highest values. There were fewer species of annual dicots for Cesis, higher numbers of perennial dicot species at Riga and no monocot species at the Valka site. There were also significant differences among years in the mean numbers of seeds. No perennial dicot seeds were recovered from the 1998 samples, but the numbers increased successively from 1999 to 2002. No monocot seeds were recovered in 1997 or 1998, but significantly greater numbers were recovered in 2000 and 2002. The year to year effects on the mean numbers of annual dicot seeds were much smaller.

Choice of crop had little effect on either species diversity or seed density. However, the previous crop had a greater effect; species number of annual dicot seeds were particularly low where the previous crop was grass. Herbicide had no significant effects on the seedbank.

As with the plant data, regression analysis was employed to assess the relative importance of field, year, crop, previous crop and herbicide as factors to explain the observed variation in numbers of species and numbers of seeds. Compared with the plant data, there were few significant regressions and the percentages of variance accounted for (adjusted R<sup>2</sup>) were much lower. For species numbers, year was the single factor that accounted for the greatest percentages of the variance for all three weed groups and for all species combined: 34, 32, 28 and 58% respectively. Crop accounted for 13, 19, 10 and 12% respectively. Previous crop accounted for 29% of the variance for numbers of species of annual dicots, but less than zero for the other two groups. For seed numbers, the only significant regression was for year and monocots (28% of variance), but year also accounted for 21% of the variance for perennial dicots. Multiple regressions including crop, previous crop and herbicide gave some improvements, but the effects were inconsistent and the percentages of variance accounted for were generally much lower than for the corresponding plant data. There were no significant correlations between the numbers of species of seeds and plants or between the numbers of seeds and numbers of plants for any weed group.

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# Effects of crop species, tillage regime and herbicide treatment on dynamics of Mediterranean arable weed seedbank

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## Introduction

Analysis of the size and composition of the weed seedbank in the soil can provide information of past and present weed populations, reveal the effect of agricultural management systems and forecast any future weed problems (Wilson *et al.*, 1985). Seed density and species composition of the weed seedbank are largely governed by soil and environmental characteristics and their interaction with management practices. The variables over which growers have some control include tillage system, crop rotation, and weed control practices (Ball, 1992). The objective of this study was to characterize the weed seedbank and population dynamics at the beginning of the research and then to determine changes due to different tillage systems, cropping sequences, and herbicide treatments.

## Materials and methods

Field research was conducted in 2003-2004 growing season, at Alexandria, Imathia, northern Greece. A split-split plot approach was used in a randomized complete block design (RCBD) with three replications. Main plot treatments were the three tillage systems (moldboard plough, chisel plough and rotary-hoe), and sub plot treatments the three cropping sequences (cotton-cotton, cotton-sugar beet, tobacco-tobacco) by herbicide application or untreated.

After seedbed preparation in late April 2003-2004, soil-core samples were taken from each subplot at two depths, 0-15cm and 15-30cm by using a 5cm diameter core sampler device. Four soil samples were taken randomly from each subplot and then bulked together. Seed extraction from the soil-core samples took place by passing each sample through a 0.5 mm mesh screen under running water. Seeds retained were identified by the means of a magnifying lens and recorded. Only seeds that were resistant to a slight pressure with the forceps were considered to be viable.

Three weed control assessments were performed in the experimental area during June, July, and August 2003-2004, where the individuals of each weed species were recorded in an area of 25 cm x 2 m oriented in the middle of each subplot. After each assessment, hoeing was performed between the rows only for the herbicide treated subplots. All data recorded were subjected to analysis of variance (ANOVA) by using a split-split plot design. Treatment means were compared by using LSD procedures at  $P=0.05$  level of significance.

## Results and discussion

*Amaranthus* spp. and *Portulaca oleracea* were the most abundant species in the soil seedbank ranging from 76% of the total seed density (at 0-15 cm) to 90% (at 15-30 cm). These results are similar to those found by Uremis *et al.* (2003). All weed species identified in the above-ground flora were also found in the weed seedbank, but the emerged *Amaranthus* spp. plants were less than the expected according to their abundance in the seedbank. However, the opposite occurred with the *Cyperus rotundus* and *Sorghum halepense* plants, which were found to be more common in the field than in the seedbank. Similar findings were also reported by Uremis *et al.* (2003). Total seed density (0-30 cm) was significantly reduced on the herbicide treated plots. Seed density was also reduced near the surface of the plots treated with moldboard plough in comparison with those treated with the other tillage systems. This could be resulted from the burial of seeds at 15-30 cm by the moldboard plough, which is in accordance with the results of Ball (1992).

Weed population dynamics in field were significantly affected by crop species and herbicide treatments during the two years of the study. The results obtained are related to different establishment time and competitiveness, but also different herbicide applications (Andersson & Milberg, 1998). Seedbed preparation for cotton sowing in mid May, in 2004, and late establishment of tobacco crop resulted in the reduction of *Echinochloa crus-galli* and *Solanum nigrum* populations as their period of emergence ranges from spring till early summer. This was in contrast to 2003, where cotton sowing was made in late April and reduced *Amaranthus blitoides* and *S. nigrum* populations. Seedbed preparation for tobacco transplanting in late May probably broke up some of the rhizomes of *S. halepense* and bulbs of *C. rotundus* found in the upper 15 cm, or brought them to the surface where the sprouting conditions were not ideal, resulting in the reduction of the two perennials. *Solanum nigrum* control in herbicide treated plots was expected as prometryn and pendimethalin applied in soil tend to provide good and partial control of *S. nigrum*, similar to the observations reported by Keeley & Thullen (1991). However, *Sorghum halepense* population reduction was probably caused by trifluralin soil incorporated pre-sowing treatment, which is in accordance to the results found by McWhorter (1974). *Cyperus rotundus* control, in 2004, was probably a result of hoeing that was performed after each assessment, since the herbicides applied are not effective on perennial weeds like *C. rotundus*.

Table 1. Effect of crop species, tillage regime, and herbicide treatment on weed population dynamics.

	Mean values (plants/m <sup>2</sup> )													
	AMABL		ECHCG		CYPRO		SOLNI		SORHA		Others		Total	
	2003	2004	2003	2004	2003	2004	2003	2004	2003	2004	2003	2004	2003	2004
MP	2.8	1	3.2	5.7	79.3	39.9	4.6	2.8	11.7	2.6	2.8	3.4	104.4	55.4
CP	1.8	0.8	0.9*	6.1	92.3	42.3	4.9	2.7	10.3	3	2.3	2.6	112.5	57.5
RH	3	1.6	2.2	5.9	88.7	45.7	4.3	2.3	8.4	2	2.3	1.9*	108.9	59.4
<b>LSD</b>	<b>3.6</b>	<b>3</b>	<b>1.1</b>	<b>5.4</b>	<b>53.3</b>	<b>44.7</b>	<b>4.2</b>	<b>2.8</b>	<b>5.9</b>	<b>3.1</b>	<b>1.3</b>	<b>0.9</b>	<b>47.8</b>	<b>44.6</b>
C	0.8*	1.4	1.3	5.8*	97.9	51.2	3.2*	1.1*	12	2.7	2.4	3.2	117.6	65.4
C/SB	2.7	1.6	2.2	8.7	100.1	41.4	5.1	5.8	10.9	3.4	1.6*	2.5	122.6	63.4
T	4.1	0.5	2.8	3.3*	62.3**	35.3	5.4	0.8*	7.5*	1.5	3.4	2.3	85.5**	43.7*
<b>LSD</b>	<b>2.3</b>	<b>1.4</b>	<b>3.5</b>	<b>2.7</b>	<b>13.6</b>	<b>16.5</b>	<b>1.7</b>	<b>2.7</b>	<b>3.2</b>	<b>1.8</b>	<b>1.6</b>	<b>1.6</b>	<b>16.7</b>	<b>14.4</b>
Herbicide	0.9**	0.6	0.9*	5.7	87.3	38.7*	1.8**	0.5*	7.2**	1.8*	1.2**	1**	99.3**	48.3**
Utreated	4.2	1.7	3.3	6.1	86.2	46.6	7.4	4.7	13.1	3.3	3.8	4.3	118	66.7
<b>LSD</b>	<b>1.6</b>	<b>1.2</b>	<b>1.7</b>	<b>1.4</b>	<b>7.7</b>	<b>5.1</b>	<b>2.3</b>	<b>2.4</b>	<b>1.9</b>	<b>1.4</b>	<b>1</b>	<b>1.1</b>	<b>8.5</b>	<b>4.6</b>

Abbreviations: MP, moldboard plough; CP, chisel plough; RH, rotary- hoe; C, cotton; C/SB, cotton→sugar beet; T, tobacco; AMABL, *Amaranthus blitoides*; CYPRO, *Cyperus rotundus*; ECHCG, *Echinochloa crus-galli*; SOLNI, *Solanum nigrum*; SORHA, *Sorghum halepense*. \* and \*\* = significant difference at P<0.05 and P<.001.

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# Effects of organic farming on weed abundance - long-term results from a site in Northern Germany

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## Introduction

One of the aims of organic farming systems is to maintain or enhance the agricultural and natural biodiversity in agro-ecosystems. Weeds can contribute to biodiversity but also need to be controlled. Compared to conventional farming the weed density and diversity in organic farming is expected to be enhanced because of reduced fertilisation and low efficacy of control measures. It is also assumed that the diversity of crops, especially alternation of summer and winter crops, results in a greater range of weed species. However, relatively few data are available confirming this benefit of organic farming. Thus, continuous monitoring studies have been started in 1996 on a trial area for organic farming in northern Germany.

## Materials and methods

Trials on organic farming have been conducted since 1996 on an experimental area of 10 ha certified according to EC Regulation No 2092/91. This site ('Ahlum, BBA') is located south of Braunschweig and consists of homogenous loam with a high nutrient level, an annual precipitation of 579 mm and an annual mean temperature of 9.3°C. The area is managed under an arable farming system with a crop rotation of 50% cereals and 62.5% summer crops. Legumes were grown every 4 years.

Weeds were controlled by harrowing at least once in each crop and year. The soil was regularly ploughed to a depth of 20–25 cm in autumn or spring. Stubble cultivation was mainly done with a wing share cultivator (cultivation depth 10 cm). Weeds were assessed annually in the field at 205 fixed reference points in a grid of 24 x 24 m by DGPS. The same points were also used for soil samples (to 30 cm) to determine the weed seedbank by germination tests.

## Results and discussion

Since 1996 both weed density and species diversity increased markedly. Weed density assessed before direct control measures ranged between 62 and 223 plants m<sup>-2</sup> throughout the observation period. The following annual species were abundant each year:

- monocotyledonous weeds: *Alopecurus myosuroides*, *Apera spica-venti* and *Poa annua*
- dicotyledonous weeds: *Galium aparine*, *Lamium* spp., *Matricaria* spp., *Sonchus oleraceus*, *Stellaria media*, *Thlaspi arvense*, *Urtica urens*, *Veronica* spp. and *Viola arvensis*

Depending on the year, these species made 63% to 80% of the entire infestation. During the nine years of organic management the number of species estimated on the field increased from 19 in 1996 to 36 in 2003 (Fig. 1). In contrast to these field estimations more weed species were found in the seedbank, but the increase was similar. Over all years 43 weed species were estimated in the field compared to 53 in the seedbank. Species like *Gnaphalium uliginosum*, *Juncus bufonius* and *Solanum nigrum* were found in the seedbank, but have never been observed by field estimations. Some other weeds appeared sporadically in the field including *Aphanes arvensis*, *Taraxacum officinale* and *Rumex* spp.. This observation has been reported by others (van Elsen, 2000) and might be due to different dormancy patterns depending on weed species and actual growth conditions (Albrecht, 2002).

Out of the 12 most frequent species mentioned above only 1 decreased, 7 increased and the others remained virtually constant. In particular numbers of monocotyledonous weeds in the soil seedbank rose to an extremely level by 2004 (Tab. 1).

Figure 1. Number of weed species on the field and in the seedbank at Ahlum, 1996-2004.

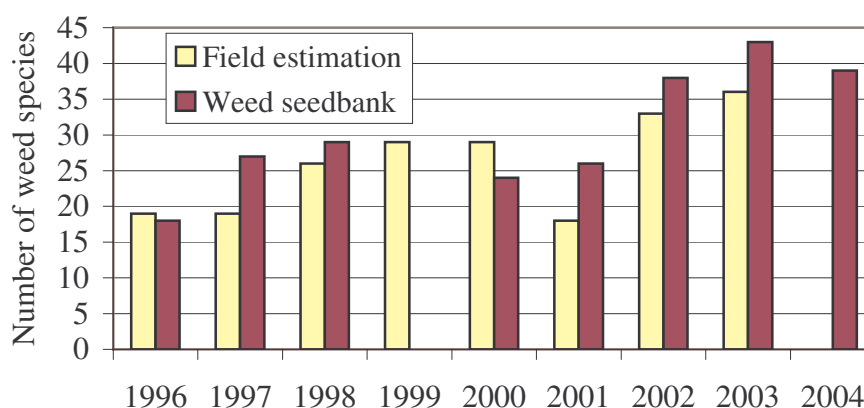


Table 1. Number of weed seeds m<sup>-2</sup> to 30 cm soil depth at Ahlum, 1996-2004.

	1996	1997	1998	2000	2001	2002	2003	2004
Monocotyledonous	479	924	686	442	286	205	922	2264
Dicotyledonous	3709	6376	6171	4444	9068	6022	11537	9264
All	4188	7300	6857	4886	9354	6227	12459	11528

Despite the low fertilisation level non-nitrophilous weed species could not profit from the organic farming system. This is clearly related to the high soil fertility such that even *Galium aparine* and other nitrophilous species have not been repressed during the past nine years. The area infested with *Polygonum convolvulus* and *Cirsium arvense* has increased considerably with up to 50 and 63% respectively. Especially, *C. arvense* has become a serious problem due to less effective weed control and crop competition (Verschwele & Häusler, 2003).

Weed communities are not only affected by the production system but also by soil conditions and the diversity of the landscape (Jüttersonke & Arlt, 2002). Among other reasons this may account for the fact that weed populations have changed only little at this site: Even after nine years of organic farming the most abundant weed species were those which were highly abundant before and those recently abundant on the adjacent conventional field. Although some further weed species appeared, rare or even endangered species could not be found. Significant changes in weed communities appear difficult to achieve exclusively by organic farming. Under optimal growing conditions rare species can be suppressed by both the crop and other competitive weeds. These findings are similar to those of other investigations on organic fields (Albrecht, 2002; Hyvönen et al., 2003).

Apart from the contribution of weeds to arable biodiversity the risk of weed competition and the necessity for control measures has to be considered. If the numbers of weed species is just a function of the total number of plants (Hyvönen et al., 2003), intensive and improved control measures will reduce both weed density and, unfortunately, species diversity too. Therefore a balance is needed between the control intensity and the intended weed diversity, challenging researchers as well as farmers.

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## Weed flora in Italian rice fields.

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### Introduction

The intense and widespread use of herbicides, together with trades of agricultural goods even between very distant countries, are the major causes of floristic modifications in weed populations around the world. This change has also been evident in rice. At the beginning of the '60 weed flora in Italian rice fields was made up of a vast number of species (Pirola, 1964), but its diversity sharply declined in recent decades (Sparacino *et al.*, 1985, Tabacchi and Romani, 2001). Exotic plants appeared, which were completely absent in the indigenous flora. In this situation a periodic monitoring of weeds in the various rice areas can also be fundamental to deciding on the best control strategy. The present investigation was carried out with this aim.

### Materials and methods

During the summer 2001 weed floras of 50 rice fields in the main Italian rice producing areas were assessed by Braun-Blanquet phytosociological method. The fields were sited in the following regions: Piedmont, Lombardy, Emilia Romagna, Veneto, Tuscany, Sardinia and were untreated with herbicides. In 2004 a series of interviews with growers and surveys were also conducted in the same areas to assess the dynamics of major weeds in rice. Concerning chemical control in Italy, propanile is the most used herbicide in rice fields; profloroxim, quinclorac and azimsulfuron are frequently applied, together with bispyribac-sodium, but only in 2004. Other herbicides used are ciclossidim and glyphosate, within the false-sowing technique against *Oryza sativa* var. *silvatica*, and MCPA against sulfonylurea-resistant monocotyledonous not gramineous weeds.

### Results and discussion

In the 2001 survey, 51 spontaneous species were recorded in total. This number was certainly less than the potential weed flora diversity that can be found in Italian rice environments (Viggiani *et al.*, 2004). The most widespread weeds, in order of frequency over all regions, were: *Echinochloa crus-galli* (L.) Beauv. (98%), *Heteranthera reniformis* Ruiz et Pavon (74%), *Bolboschoenus maritimus* (L.) Palla (52%), *Schoenoplectus mucronatus* (L.) Palla (48%), *O. sativa* var. *silvatica* Chiappelli (43%), *Lindernia* spp. All. (43%), *Alisma* spp. L. (30%), *Heteranthera rotundifolia* (Kunth) Griseb. (26%), *Ammania* spp. L. (22%), *Leersia oryzoides* (L.) Swartz. (22%), *Paspalum paspaloides* (Michx.) Scribner (22%), *Cyperus* spp. L. (22%), and *Butomus umbellatus* L. (17%).

*Echinochloa crus-galli* was present everywhere along with its subsp. *E. crus-pavonis* (H.B.K.) Schultes. However, its supremacy is now threatened by other barnyardgrass species, which, in Italy, are commonly called "white". In particular, the importance of *E. phylloporon* auct. non Stapf and *E. erecta* (Pollacci) Pignatti is increasing where *E. crus-galli* is successfully controlled by herbicides. In the near future *E. erecta* will become a serious problem because, like *E. crus-galli*, it can infest many crops, as was frequently found in 2004 in maize after rice in Ferrara province. Among other white barnyardgrasses, *E. oryzoides* (Arduino) Fritsch is spreading particularly in Veneto and Emilia Romagna. In contrast, *E. colonum* (L.) Link was reported only in some restricted areas of Lombardy.

In the field investigation we only found the following *Heteranthera* species: *H. reniformis* and *H. rotundifolia*; the first one more frequently, but the second was rapidly increasing,



particularly in Lombardy and Piedmont. Everywhere both species have overcome *H. limosa* Willd which was frequently recorded in Italian rice fields a few years ago. Now it is reported only in Tuscany and Sardinia, but its real presence is uncertain.

The most important *Cyperaceae*, species were *S. mucronatus* and *B. maritimus*. For the latter, the subspecies *maritimus* prevailed in Veneto and Emilia Romagna; the *compactus* (Hoffm.) Hejn subsp. in Lombardy and Piedmont. Other important *Cyperaceae* were *Cyperus difformis* L., *C. fuscus* L., *C. glomeratus* L., *C. longus* L., *C. strigosus* L. and the rapidly spreading *C. serotinus* Rottb. and *C. esculentus* L.. *Oryza sativa* var. *silvatica* was ubiquitous in Italy, with a lower frequency in Tuscany.

Three genera were observed, each with two species: *Lindernia* (*L. dubia* [L.] Pennel and *L. procumbens* [Krocher] Philcox), *Alisma* (*A. lanceolatum* With. and *A. plantago-aquatica* L.) and *Ammania* (*A. auricolata* Willd. and *A. coccinea* Rothb.). *Lindernia* spp. were mainly found in Sardinia, whilst the frequency of *Alisma* spp. and *Ammania* spp. was equally distributed all over Italy.

In the Italian peninsula *L. oryzoides* and *B. umbellatus* infestations were much more frequent than in Sardinia. The frequency of *P. paspaloides* was particularly high in Emilia Romagna, but its occurrence also showed an increase in other parts of Italy. Two species of *Leptochloa* were also observed to be spreading rapidly: *L. fascicularis* (Lam.) Gray in Lombardy and Piedmont; *L. uninervia* (J. Presl) H&C in Emilia Romagna and Veneto. *Alopecurus geniculatus* L. frequently appeared in Lombardy and Piedmont, while *Sparganium erectum* L. and *Sagittaria* L. spp. (*S. latifolia* Willd. and *S. sagittifolia* L.) showed a sporadic presence.

In addition to these weeds, which showed a nationwide distribution, the following species were observed only at specific sites, located in the bracketed provinces: *Ottelia alismoides* (L.) Pers. (Biella); *Murdannia keisak* (Hassk.) Brenan and *Commelina communis* L. (Vercelli); *Limnophyla indica* (L.) Druce x *L. sessiliflora* Bl., *Salvinia natans* (L.) All. and *Elatine triandra* Schkuhr (Ferrara), *Veronica anagallis-acquatica* L. (Siena); *Myriophyllum verticillatum* L. in all Emilia Romagna, and *Eclipta prostrata* L. in Sardinia Isle.

Finally, it should be noted that the following species were present especially along ditches but can easily invade rice fields: *Bidens* spp. L. and *Typha* spp. L. were found almost everywhere, *Polypogon monspeliensis* (L.) Desf, was detected to be widely spread in Emilia Romagna, *Glyceria maxima* (Hort.) Holberg, *Lithrum salicaria* L., *Nasturtium officinale* R. Br., *Panicum dichotomiflorum* Michx., *Polygonum amphibium* L., *Polygonum hydropiper* L., *Potamogeton natans* L., *Rorippa* spp. (L.) Besser, *Rotala* spp. L., *Scirpus* spp. L., *Sphaeroplea annulina* Agardh and *Stachys palustris* L.

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# The influence of *Brassica* crops on the soil seed bank and amount of the weeds in following wheat crops

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## Introduction

It is well known that field brassica crops are good preceding crops for many agricultural plants, including cereals. The phytosanitary role of these crops is well known as well as their ability to stimulate and to decrease the growth and development of the other biotic components of phytocoenosis. Their positive role in reducing the amount of weeds in the following crops has been established. Some scientists explain it by the allelopathic activity of the cabbage crops (Jimenez-Osornio, Gliessman, 1987), others by their high competition with weeds (Kahnt, 1988), which leads to the lowering of the seed productivity of the weed plants and as result to the decreasing of the soil seedbank. The differences between the ability for some brassica crops to suppress the weed plants and hence decrease the amount of weed seeds in soil, has been previously established (Wlassenko, 1998; Vlasenko, 2004). It has been interesting to research how such differences are displayed during the agrocoenosis formation of the following cereal crops.

## Materials and methods

The experiments were carried out in 2000-2004 in the forest steppe of Western Siberia. The soil of the experimental plots was a loamy leached chernozem with a humus content of about 5 % in the 0-30 cm soil layer. The weather conditions varied over a wide range during the different years. There were 379 mm of precipitation from May to August in 2000, 297 mm in 2001, 248 mm in 2002, 113 mm in 2003 and 217 mm in 2004. The average precipitation for several years was 232 mm. The sum of effective temperature (> 5 °C) for the same period was 1392, 1498, 1402, 1531 and 1505 °C for 2000, 2001, 2002, 2003 and 2004 respectively. The average for several years was 1299 °C.

The influence of the species of crop, nitrogen fertilizer and herbicides on the soil seedbank and the amount of the weeds in the following wheat crop, were studied. Spring rape (*Brassica napus*), mustard (*B. juncea*) and bigseed falseflax (*Camelina sativa*) were sown after wheat (preceding crop) and fallow (pre-preceding crop) on the 10, 16 and 19 of May in 2000, 2001 and 2003. N<sub>90</sub> was applied to the soil before sowing. The crops were treated with the insecticide Fastak ( $\alpha$ -Cypermethrin) for pest control. The application rate was 0.15 l/ha. For weed control a tank mixture of Furore Super (1.0 l/ha; active ingredient Fenoxaprop-P-ethyl) and DPX-A-7881-22 (15 g/ha; active ingredient Methyl-2-[4-ethoxy-6-methylamino-1,3,5-triazin-2-yl-carbamoyl-sulfanoyl-benzoat) was used. Weed control was made at the leaf rosette growth stage. The second herbicide was not used in the bigseed falseflax because of phytotoxic risks. Spring wheat was sowed after the brassica crops on the same areas on the 10, 18 and 19 of May in 2001, 2002 and 2004. Each crop was grown in an area about 50 m<sup>2</sup> and replicated four times. The weed seedbank in the 0-10 cm soil layer was measured after harvesting of the Brassica crops. Weed density was estimated on an area of 0.25 m<sup>2</sup> at the wheat tillering growth stage.

## Results and discussion

It was not possible to determine the influence of the species of brassica crop on the amount of weed seeds in the soil (Coefficient of variation (V) = 1.9% in 2000, 0.3% in 2001 and 11.7% in 2003). It was established that in 2000 the soil seedbank increased by 15% on average in the treatments that omitted fertilizer and herbicides (controls), whereas in contrasts it declined by

16% in 2001 and in 2003 the seedbank remained constant. In 2000 and 2001 nitrogen fertilizer caused a considerable difference in the amount of weed seeds found in the soil ( $V = 54.6\%$  and  $38.4\%$ ), in 2003 there was an average change ( $V = 15.6\%$ ). Cropped areas treated with herbicide but with no added fertilizer, caused considerable variation of this index from an average of  $19.6\%$  in 2000 to  $37.6\%$  in 2001 and  $31.5\%$  in 2003. Using fertilizer in wet periods (2000-2001) decreased the amount of weed seeds in the soil by  $45\%$  on average, herbicides by  $40\%$ , and both of herbicides and fertilizer by  $52\%$ . Where there was a deficit of precipitation (2003) these indices were  $23\%$ ,  $28\%$  and  $28\%$  accordingly.

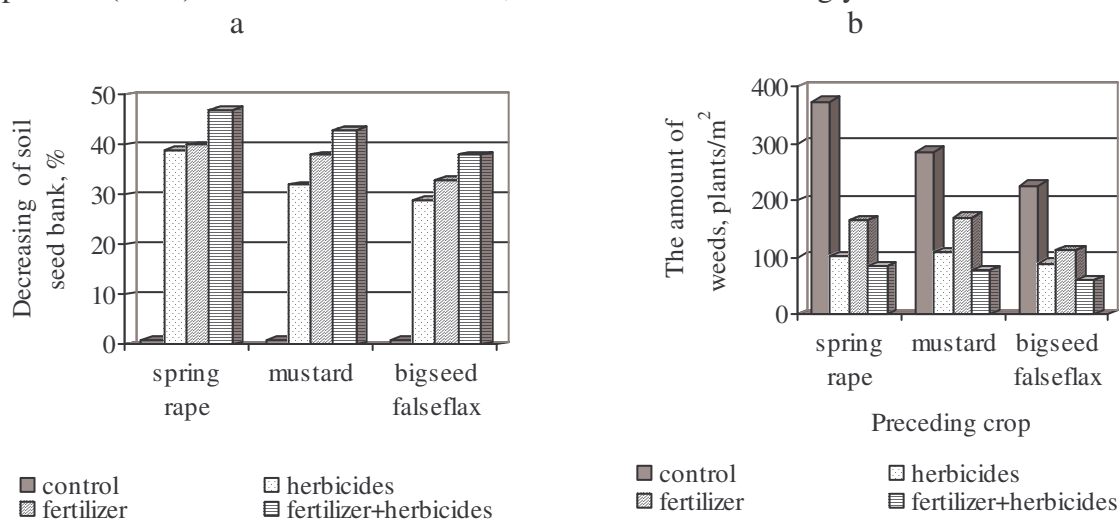


Figure 1: Influence of the cultivation of brassica crops on the soil seedbank (a) and amount of weeds in the following wheat crop (b) (average of 3 years).

After cultivation of brassica crops, the amount of weed seeds in the soil decreased by  $1\%$  on average for three years, independent of the crop species. The application of herbicides on the spring rape decreased the soil seedbank by  $39\%$ , nitrogen fertilizers by  $40\%$ , and using both herbicides and fertilizer by  $47\%$  (Figure 1 a). These indices were slightly less after mustard harvesting ( $32\%$ ,  $38\%$  and  $43\%$ ). Bigseed falseflax decreased the amount of weed seeds in the soil by the least amount ( $29\%$ ,  $33\%$  and  $38\%$ ).

However, the amount of weeds in the wheat crop when grown after bigseed falseflax, were less than after other two crops. This was observed for both the control and also the different experimental treatments. In comparison with bigseed falseflax, the amount of weeds in the wheat after mustard increased in control by  $27\%$ , with herbicides by  $22\%$ , with fertilizers by  $52\%$ , and with fertilizer + herbicides by  $57\%$ . These indices were  $65\%$ ,  $15\%$ ,  $46\%$  and  $41\%$  when spring rape was used as preceding crop. Application of herbicides and fertilizer in spring rape and mustard led to a stabilizing of the amount of weeds in the following wheat crop. Comparing the data from soil seedbank with the amount of weeds found in the following wheat crop, it is possible to assume that bigseed falseflax is not only competitive with weeds, but also has some potential allelopathic effect.

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## Pasture weeds as possible accumulators of heavy metals

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### Introduction

Natural grasslands in Serbia occupy huge areas accounting to near 1.65 million ha and exhibit significant diversity in flora due to various environmental influences, such as altitude, climate, relief, soil etc.. Regarding the composition of the herbaceous vegetation, all plant species participate equally. However, to classify grassland management and utilization, they could be divided into two groups: a) favorable (useful) species – plants of high fodder value readily taken by cattle in grazing, and b) unfavorable (useless, harmful) species – plants of low fodder value, prickly and poisonous, which are almost always avoided by cattle in grazing (Kojic et al., 1994). The latter category of grassland inhabitants may be designated as a weed component in meadows and pastures. Such plant species contribute more than 50 % of the floristic composition of natural Serbian grasslands (Kojic et al, 2000). Many plant species have the capacity to accumulate some biogenic elements, heavy metals and radionucleides above toxic values. The quantity of heavy metals that can be taken up from the soil depends both on the plant species and the soil properties, first of all the pH, and on the properties of the element itself (Kashem and Singh, 2002). Considering the fact that many plants of natural grasslands are found to be hyper-accumulators of heavy metals (Keller et al., 2003), several widespread pasture species of Mt. Stara Planina (east Serbia) were surveyed for content of heavy metals in their shoots.

### Material and methods

The plant samples for analysis of heavy metals were taken from pastures on the mountain Stara Planina (N= 43° 14' 24.7", E= 22° 51' 36.8") during the first week of July, 2003. The height of the vegetation cover was 95-100 cm, and the general degree of coverage was 100%. The slope of the study locality is 20-25%, exposure - N, altitude 1288 ± 6 m. The content of heavy metals (Cu, Zn, Pb, Ni, Cr and Mn) was determined in five species which are supposed to be bioaccumulators: *Cytisus albus* Hacq., *Hypericum perforatum* L., *Thymus vandasii* Vil., *Verbascum longifolium* Ten. and *Centaurea phrygia* L.. The total concentration of heavy metal was determined by atomic absorption spectrophotometry. The soil of the studied locality was previously determined as dystric humus-siliceous in two varieties: 1) lithic and brownised and 2) dystric brown soil and then characterized by its good quality based on acidification index and heavy metal content (Belanovic et al., 2003).

### Results and discussion

The differences in concentration of heavy metals among studied plants (Table 1.) depends primarily on their genetic characteristics, on the effect of root area and its capacity of ion adsorption, form of root exudates and the rate of evapotranspiration (Alloway, 1995).

Table 1. Concentration of heavy metals (mg.kg<sup>-1</sup>) in shoots of pasture weeds of Mt. Stara Planina in Serbia

Species	Cu	Zn	Pb	Ni	Cr	Mn
<i>Hypericum perforatum</i>	14.01	30.02	3.00	1.00	2.00	219.15
<i>Centaurea phrygia</i>	13.99	18.98	2.00	3.00	2.00	105.40
<i>Thymus vandasii</i>	12.0	44.99	3.50	5.00	4.50	363.43
<i>Verbascum longifolium</i>	12.96	23.43	3.50	1.00	1.49	321.50
<i>Cytisus albus</i>	12.45	32.39	0.99	2.49	1.00	472.32

The concentration of Cu in all studied plants and concentration of Pb in *T. vandasii* and *V. longifolium* are somewhat above the range reported for unpolluted areas (de Vries and Bakker, 1996). In the species *C. phrygia*, *T. vandasii* and *C. albus* the concentrations of Ni are higher than the natural concentrations in grasses according to Kabata-Pendias and Pendias, 1989. Higher concentrations of Cr and Ni were found in *T. vandasii* (after ECCE, 1994). Measured concentrations of Mn were above the limits in *T. vandasii* and *C. albus*.

Preliminary results of the heavy metal content in several pasture weeds showed that among them *T. vandasii* exhibited the highest bioaccumulation of heavy metals, primarily Pb, Ni, Cr and Mn, whilst bioaccumulation of Ni and Mn in *C. albus* was considerable. However, it is necessary to carry out further research on the soil – plant system, primarily to study the adsorption complex, and also to analyse the forms of heavy metal fixation and their origin, as well as the affinity of individual plant species to the specific element.

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# Influence of emergence time and density of redroot pigweed (*Amaranthus retroflexus* L.) on soybean (*Glycine max* L.)

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## Introduction

The major objective of a crop production system is the interception, fixation and storage of sunlight energy and for every crop producer, minimizing of inputs to the system and maximizing of yield is ideal. Integrated weed management (IWM) represents an interdisciplinary approach to linking social, economic and environmental interactions in the field (Swanton and Murphy, 1996). Redroot pigweed is a major weed occurring in fields of soybean. Yield losses caused by interference from various species of pigweed have been reported for numerous crops. The objective of this study is to determine the influence of selected redroot pigweed densities and times of emergence on soybean growth and yield.

## Materials and methods

Field study was conducted at agricultural Research Station of Tehran University in Karaj during 2001. The soil type was a clay loam soil (35% sand, 28.8% silt, 37.2% clay, 7.8 pH). Soybean cultivar Williams was planted in 50 cm row spacing, at 5 cm deep and 40 plants  $m^{-2}$  on 14 May 2001. Redroot pigweed was planted with soybean simultaneously and it emerged at the same time with the soybean (Ve), redroot pigweed was planted a week after the first soybean planting date and it emerged at the cotyledon leaf stage of soybean (Vc) and redroot pigweed was planted two weeks after the first soybean planting date and it emerged at the second trifoliolate leaf stage of soybean (V2). Redroot pigweed seed was manually sown in a 15 cm band over the soybean row. Redroot pigweed seedlings were thinned to densities of 10, 20 and 40 plants  $m^{-1}$  of row for each emergence date. To define the maximum yield loss of soybean due to high weeds density and weed-free condition, two plots were included additional, one of high weed density (all of weeds emerged from seed bank) and one weed-free by hand hoeing with three replications. Experimental design was a factorial in a Randomized Complete Block Design with three replications. The inter-row space for all densities of redroot pigweed was kept weed-free by hand hoeing. Plots were 2.5 m wide (four crop rows) by 8 m in length with weeds established in all crop rows. At physiological maturity stage, 20 soybean plants  $m^{-2}$  were sampled in each plot and number of pod per soybean plant, number of seed per soybean pod and 1000 seed-weight were determined. Soybean was harvested manually on October 7, 2001. The crop was hand-clipped from a 3 m section of the two center rows of each plot, hand-fed into a mechanical thresher, and yield at 14% of moisture was measured. Means were compared using the Duncan's multiple range test at the 5% significant level.

## Results

Soybean grain yield and percentage loss varied depending on density and time of redroot pigweed emergence. Soybean yield reduction was 25, 41 and 68% respectively for redroot pigweed densities of 10, 20 and 40 plants  $m^{-1}$  of row when redroot pigweed emerged at Ve. Soybean yield reduction when redroot pigweed emerged at Vc and V2 and increasing of redroot pigweed density is showed at Figure (1). Klingman and Oliver (1994) found that when weed emerging was at the same time with soybean, weed interference was increased and soybean competitive ability increased with delaying of weed emergence time. The number of soybean pods per plant reduction by 10, 20 and 40 plants  $m^{-1}$  of row was 14, 38, and 50% when redroot pigweed emerged at Ve. The number of soybean pods per plant loss due to second redroot pigweed emergence time was 10, 22 and 38% and it was 6, 7 and 8%

for third emergence time respectively (Figure 2). The number of soybean pods per plant has been the yield component most affected by weed competition. One can expect greater yield losses from weeds that shade soybean than from weeds that do not. Shade during the flowering period has been reported to increase the number of aborted soybean pods. This may have resulted from reduced sugar levels in leaves, caused by a lower rate of photosynthesis in the shade (Bloomberg *et al.* 1982). The effect of emergence time and redroot pigweed density on number of seeds per pod and 1000 seed weight was not significant.

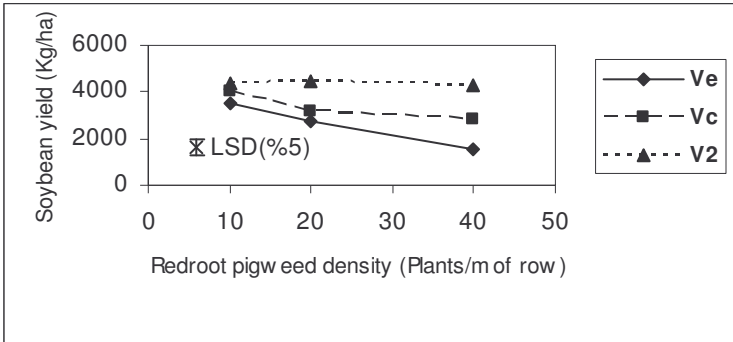


Figure 1. Soybean yield affected by emergence time and density of redroot pigweed

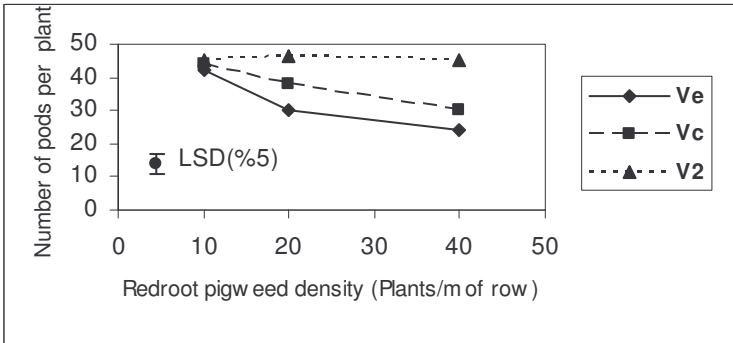


Figure 2. Number of pods per plant affected by emergence time and density of redroot pigweed

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# The Effect of Nitrogen Fertilizer and Relative Plant Density on Wheat and Wild Oat Competitive indices

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## Introduction

Plants competing for a factor necessary for their growth and development show different reactions depending on the subject of competition. Competitive interactions of wheat and wild oat can be affected by plant density of both species and nutritional conditions. A replacement series experiment, analysing some competitive indices, can give quantitative information on crop-weed intractions.

This research was conducted to study competitive indices of wheat and wild oat at different relative plant densities and nitrogen fertilizer levels.

## Material and methods

A replacement series experiment was conducted to investigate the effect of nitrogen fertilizer and relative plant density on competitive indices of wheat (*Triticum aestivum*) and wild oat (*Avena ludoviciana*) under constant total plant density with different proportion plant density of species. The experiment was carried out as a factorial experiment, based on a randomized complete block design, with 4 replications, in a greenhouse of Agricultural Faculty of Mashhad, Iran in 2001. Treatments were 5 rates of nitrogen topdressing (0, 0.5, 1, 1.5 and 2 grams per pot, corresponding to 0, 70, 140, 210 and 280 kg N ha<sup>-1</sup> respectively) applied as urea and 5 relative plant densities of wheat and wild oat (100:0, 75:25, 50:50, 25:75 and 0:100). Total plant density was 20 plants per pot, corresponding to 350 plants m<sup>-2</sup>. The pot volume was 16000 cm<sup>-3</sup> (18 cm diameter and 26 cm depth). By measuring the individual plant dry weight and total above ground biomass of species at the harvest, different competitive functions such as Relative Yield of species (RYi) (Dewit, 1960), Competition Index (CI) and Relative Competitive Ability (RCA) (Mark and Renner, 1990) were calculated. In addition intra- and interspecific competition indices of both species and the Nich Differentiation Index (NDI) were analysed by a multiple linear regression analysis according to the following equations, suggested by Spitters (1983):

$$1/w_{cw} = b_{c0} + b_{cc}N_c + b_{cw}N_w \quad (1)$$

$$1/w_{wc} = b_{w0} + b_{ww}N_w + b_{wc}N_c \quad (2)$$

$$NDI = (b_{cc}/b_{ww})(b_{cw}/b_{wc}) \quad (3)$$

Where  $N_c$  and  $N_w$  are crop and weed plant density in plants m<sup>-2</sup> respectively.  $b_{c0}$  and  $b_{w0}$  are individual plant dry weight of crop and weed without competition respectively.  $b_{cc}$  and  $b_{ww}$  are intra-specific competition coefficients of crop and weed respectively.  $b_{cw}$  and  $b_{wc}$  are interspecific competition coefficients of weed on crop and crop on weed respectively.

## Results and discussion

The results indicated that the above ground dry matter and individual plant dry weight of both species were significantly affected by relative plant density and nitrogen fertilizer, while there was not a significant interaction between relative plant density and N rates (data not shown). By increasing the proportion of wheat plants, individual plant dry weight of wheat

increased, in contrast individual plant dry weight of wild oat decreased as its relative plant density increased (data not shown). This reaction indicates that interspecific competition ability of wild oat against wheat is more than its intra-specific competition.

With all nitrogen levels, the wheat intra-specific competition index ( $b_{cc}$ ) was less than its interspecific competition index ( $b_{cw}$ ), while the wild oat showed the opposite reaction. Nich Differentiation Index (NDI) of competing species (Tab. 1) in all treatments was less than unity, indicating that both species compete for joint resources. Decreasing NDI in response to nitrogen fertilizer indicate that nitrogen increases interference competition of species.

With all nitrogen levels, the Relative Competitive Ability (RCA) of wild oat was more than wheat and the Competition Index (CI) of wild oat was more than unity, and those of wheat was less than unity (Tab. 2). Decreasing proportion density of wild oat increased its RCA and CI while decreased those of wheat (data not shown).

Relative Yield (RY) of wild oat was more than its relative plant density in all proportions and those of wheat was less than its proportion density (data not shown).

Analysing competitive indices showed that wild oat is more competitive than wheat and increasing nitrogen fertilizer increases its competitive ability.

Table 1. Intra- and interspecific competition indices and Nich Differentiation Index (NDI) of wheat and wild oat at different nitrogen topdressing levels.

Intra- and interspecific competitive indices	Nitrogen topdressing (gr/pot)					$r^2$	Fitted Equation
	0	0.5	1	1.5	2		
$b_{cc}$	0.344	0.344	0.343	0.344	0.343	0.33	$y = -0.0002x + 0.3442$
$b_{cw}$	0.355	0.353	0.354	0.352	0.352	0.72	$y = -0.0007x + 0.3553$
$b_{ww}$	0.349	0.348	0.346	0.346	0.345	0.93	$y = -0.001x + 0.3498$
$b_{wc}$	0.341	0.342	0.342	0.343	0.343	0.89	$y = 0.0005x + 0.3407$
NDI	0.992	0.992	0.983	0.984	0.980	0.85	$y = -0.0032x + 0.9958$

Table 2. Relative Competitive Ability, Competition Index and Relative Yield of wheat and wild oat at different rates of nitrogen topdressing.

Relative Competitive Ability	Nitrogen topdressing (gr/pot)					$r^2$	Fitted Equation
	0	0.5	1	1.5	2		
Wheat	3.18	3.22	3.13	3.13	3.13	0.54	$y = -0.038x + 3.19$
Wild oat	5.03	4.80	4.90	4.50	4.50	0.81	$y = -0.272x + 5.018$
Competition Index							
Wheat	0.80	0.81	0.78	0.78	0.78	0.61	$y = -0.014x + 0.804$
Wild oat	1.26	1.20	1.22	1.13	1.12	0.56	$y = -0.07x + 1.256$
Relative Yield							
Wheat	0.39	0.39	0.38	0.37	0.38	0.57	$y = -0.008x + 0.39$
Wild oat	0.60	0.60	0.62	0.57	0.58	0.32	$y = -0.014x + 0.608$
Total	0.99	0.99	1.0	0.95	0.96	0.48	$y = -0.022x + 0.99$

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# Light Demand, Capture and Use Efficiency in Wheat and Wild Oat Mixed Canopy

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## Introduction

Competition for light in mixed plant canopies involves both interception and photosynthetic utilisation of the intercepted light by the species. A simple model (Nassiri, 1998) based on the linear relationships between cumulative dry matter production and time integral of absorbed light provides a useful framework for analysis of the efficiency of canopies in capture and conversion of solar energy to dry matter. The slope of this linear relation, radiation use efficiency (RUE), displays the net assimilation gain of the crop per quantity of intercepted light (Sinclair and Horry, 1989). The light absorption is mainly a geometrical issue and depends on the size and structure of the canopy (Baldocchi and Collineau, 1994). Distribution of leaves over canopy height is the most important structural factor that determines the pattern of light availability and radiation interception within the canopy. The profile of light absorption by each species in a mixed canopy is in accordance with the distribution of leaf area over height.

## Material and methods

In order to investigate the light demand, supply and use efficiency in wheat (*Triticum aestivum*) and wild oat (*Avena ludoviciana*) mixed canopy, an experiment was conducted at College of Agriculture, Ferdowsi University of Mashhad, Iran in 2001. The experiment was carried out as a split plot design with 4 replications. Rates of nitrogen (25, 50, 75 and 100 kg ha<sup>-1</sup>) were assigned to the main plots and wild oat plant densities (0, 30, 50 and 80 plant m<sup>-2</sup>) to the subplots. The plant density of wheat was 400 plants m<sup>-2</sup>. Dry matter accumulation, leaf area index, canopy height and light absorption by mixed canopy were measured every two weeks during the growth season. Vertical distribution of leaf area, dry matter and light absorption in 20 cm interval layers of canopy were measured at two stages (2 weeks before and 2 weeks after the canopy closure). Light competition in mixed canopy was quantified using a demand-supply analysis, following the approach used in the CropSys model (Sinquit and Caldwell, 1995) for mixed canopy where the distinguished fractions are the light fraction actually captured by wheat (C), actually captured by wild oat (W), actually captured by both species (CW). The values for the fraction that theoretically could have been captured by wheat if all wild oat leaves had been removed (fraction C<sup>+</sup>), or the fraction that theoretically could have been captured by wild oat if all wheat leaves had been removed (fraction W<sup>+</sup>), were estimated by simulation. C<sup>+</sup> and W<sup>+</sup> are termed the light demanded by wheat and by wild oat, respectively. The fraction (C<sup>+</sup>-C) + (W<sup>+</sup>-W) is thus the light fraction subjected to competition. If (C<sup>+</sup>-C) < (W<sup>+</sup>-W), then wheat is the stronger light competitor and if (C<sup>+</sup>-C) > (W<sup>+</sup>-W) wild oat is the better competitor. When the two fractions are identical, light is equally shared between the two species.

The RUE (g DM MJ<sup>-1</sup> PAR) of each species was calculated as the slope of linear relationships, between the cumulative intercepted PAR and the accumulative aboveground DM of that species. The cumulative intercepted PAR for each species was calculated from the model.

## Results and discussion

The profile of simulated absorbed PAR density over height of the canopy was different between species. After canopy closure, the light absorption by top layers of wild oat stand was higher compared to that of wheat (data not shown). This advantage of wild oat is due to its

canopy structure with a higher proportion of its leaf area at a higher position in the canopy and its taller plant height.

Before canopy closure, the amount of light subjected to competition between species was more compared to that measured after canopy closure, and this term was increased by increasing wild oat plant density, and nitrogen fertilization (Tab. 1).

The light captured by wild oat after canopy closure was mainly closed to its demand. The fraction ( $W^+ - W$ ) was also lower than ( $C^+ - C$ ) (Tab. 1). This indicated that the light subjected to competition was mainly intercepted by wild oat.

RUE of both species (Fig. 1 right) showed a significant negative response to wild oat plant density. Nitrogen fertilization increased RUE of both species (Fig. 1 left). Nitrogen had a higher effect on RUE of wild oat than on that of wheat. It can be resulted that wild oat is a higher competitor than wheat, because of its better distribution of leaves within the canopy, and higher leaf area duration. In addition nitrogen fertilization will increase the competitiveness of wild oat against wheat.

Table 1. Simulated fraction of captured and demanded PAR by wheat and wild oat and the fraction of light subjected to competition in 25 and 100 kg ha<sup>-1</sup> nitrogen and 30 and 80 plants m<sup>-2</sup> wild oat plant density at 150 and 180 days after emergence (2 weeks before and 2 weeks after canopy closure respectively). See text for details.

Nitrogen (kg ha <sup>-1</sup> )	Wild oat plant density (plants m <sup>-2</sup> )	Day after emergence	C	C <sup>+</sup>	W	W <sup>+</sup>	C <sup>+</sup> - C	W <sup>+</sup> - W	(C <sup>+</sup> - C) + (W <sup>+</sup> - W)
25	30	150	0.88	0.90	0.03	0.11	0.03	0.08	0.11
25	30	180	0.52	0.54	0.05	0.07	0.02	0.02	0.04
25	80	150	0.79	0.87	0.10	0.28	0.08	0.18	0.25
25	80	180	0.36	0.41	0.15	0.17	0.05	0.02	0.07
100	30	150	0.89	0.93	0.05	0.20	0.04	0.15	0.19
100	30	180	0.72	0.77	0.07	0.12	0.05	0.05	0.10
100	80	150	0.76	0.89	0.16	0.42	0.13	0.26	0.39
100	80	180	0.78	0.63	0.26	0.34	0.15	0.08	0.23

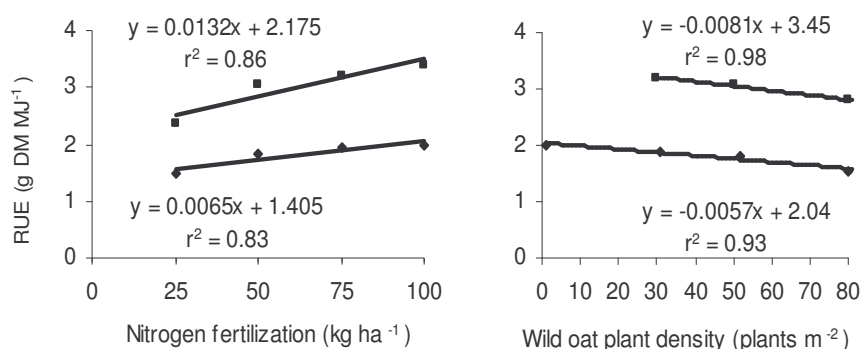


Fig 1. Simulated (lines) and observed (symbols) RUE (g DM MJ<sup>-1</sup>) of wheat (♦) and wild oat (▪) in function of N rates (left) and wild oat plant densities (right).

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# Investigation on competition between winter wheat (*Triticum aestivum*) and rye (*Secale cereale* L.) using a reciprocal yield model

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## Introduction

Winter wheat is the most important crop in Iran with approximately 6.3 million hectares sown in 2002 and production of 12 million tons (FAO 2002). Winter rye in Iran has been a problem because no selective herbicide is available for its control in winter wheat. Controlling rye is difficult because of its similar growth characteristics to winter wheat. It is important to evaluate the competitive relationship between winter wheat and rye at different density ratios. The method used to determine competition was a reciprocal yield model. This is based on the hyperbolic relationship between yield and density. Relationship between plant yield and density is hyperbolic, whereas relationship between one-plant reciprocal yield and crop density is linear, and explanation and application of linear models is easier. Because the degree of freedom of this model is high less replication is required (Pantone & Baker, 1991). Spitters (1983) used the hyperbolic yield-density model for studying competition of mixtures.

## Materials and methods

This study was from 2001 to 2002 at the research farm of Plant Pests and Diseases Research Institute in Karaj (Iran) (Lat 35°34'N, long 50°56'E). The experimental design was a randomized complete block with 24 treatments and 4 replications. The competition design was a bivariate factorial. The treatments included the pure stands of wheat cv. Mahdavi at four-plant density 350, 450, 550 and 650 plant/m<sup>2</sup>, the pure stands of volunteer rye (*Secale cereale*) at 10, 30, 50 and 70 plant/m<sup>2</sup> and mixed densities of both species at complete factorial densities. The competitive ability of wheat was investigated using a reciprocal yield model. In order to measure one-plant yield, this parameter was measured for 20 plants of winter wheat and rye individually in each plot. The data were incorporated into a weighted multiple linear regression (the reciprocal yield model) (Spitters 1983):

$$\frac{1}{W_w} = b_{w_o} + b_{ww} N_w + b_{wr} N_r \quad (1)$$

$$\frac{1}{W_r} = b_{r_o} + b_{rr} N_r + b_{rw} N_w \quad (2)$$

Where  $W_w$  and  $W_r$  were the average yields per plant for winter wheat and rye, respectively, and  $N_w$  and  $N_r$  were their densities. The intercepts ( $b_{w0}$  and  $b_{r0}$ ) estimated the reciprocal of the maximum yield of isolated plants. Intra-specific competition was estimated by the partial regression coefficients  $b_{ww}$  and  $b_{rr}$ , while inter-specific competition was estimated by  $b_{wr}$  and  $b_{rw}$ . RCA (Relative Competitive Ability) is the ratios of intra-specific competition coefficients to inter-specific competition coefficients and indicates the competitive ability of winter wheat and rye to each other. RCA (Relative Competitive Ability) indicates which of them is stronger competitor and how many plants of winter wheat are equivalent to one plant of rye in competitive ability.

$$RCA_w = \frac{b_{ww}}{b_{wr}} \quad (3) \qquad RCA_r = \frac{b_{rr}}{b_{rw}} \quad (4)$$

NDI (Niche Differentiation Index) is calculated by multiplying RCA for winter wheat and rye (Pantone & Baker 1991). To determine if the two species were competing for the same resources, the niche differentiation index (NDI) was calculated as follows:

$$NDI = RCA_w \cdot RCA_r = \left(\frac{b_{ww}}{b_{wr}}\right) \times \left(\frac{b_{rr}}{b_{rw}}\right) \quad (5)$$



## Results and Discussion

Reciprocal yield model for winter wheat grain yield and biomass, were fitted and relationship between reciprocal one-plant yield and biomass with winter wheat and rye density were obtained by multiple linear regression.

$$\frac{1}{W_w} = -0.2566 + 0.0034 N_w + 0.0067 N_r \quad r^2 = 0.91 \quad \text{Grain yield} \quad (6)$$

$$\frac{1}{W_w} = 0.0561 + 0.0011 N_w + 0.0019 N_r \quad r^2 = 0.81 \quad \text{Biomass} \quad (7)$$

Estimated regression coefficients (equations 6, 7) indicated that winter wheat biomass and grain yield, were influenced more by inter-specific competition than with intra-specific competition because the coefficient for inter-specific competition is greater than the intra-specific competition coefficient. Comparing the two equations (6, 7) it is apparent that winter wheat grain yield is influenced more than biomass by inter-specific and intra-specific competition and indicates that winter wheat grain yield is more susceptible to competition caused by rye. Tanji & Zimdahl (1997) indicated that winter wheat was influenced more by ryegrass (*Lolium*) inter-specific competition. RCA based on grain yield was 0.5 indicating that each two plants of winter wheat are approximately equivalent to one plant of rye. Using the reciprocal grain yield model coefficients NDI is 0.608 and indicates that there is no niche differentiation. Winter wheat and rye compete for identical resources and significant niche differentiation did not occur.

Results of analysis of variance indicated that increasing rye density had a significant effect on winter wheat grain yield decreasing it 3 to 40% (Figure 1A). The results showed that the optimum wheat density for this study was 550 plants/m<sup>2</sup>. The wheat density of 550 plants/m<sup>2</sup> also produced the maximum crop biomass (Figure 1B).

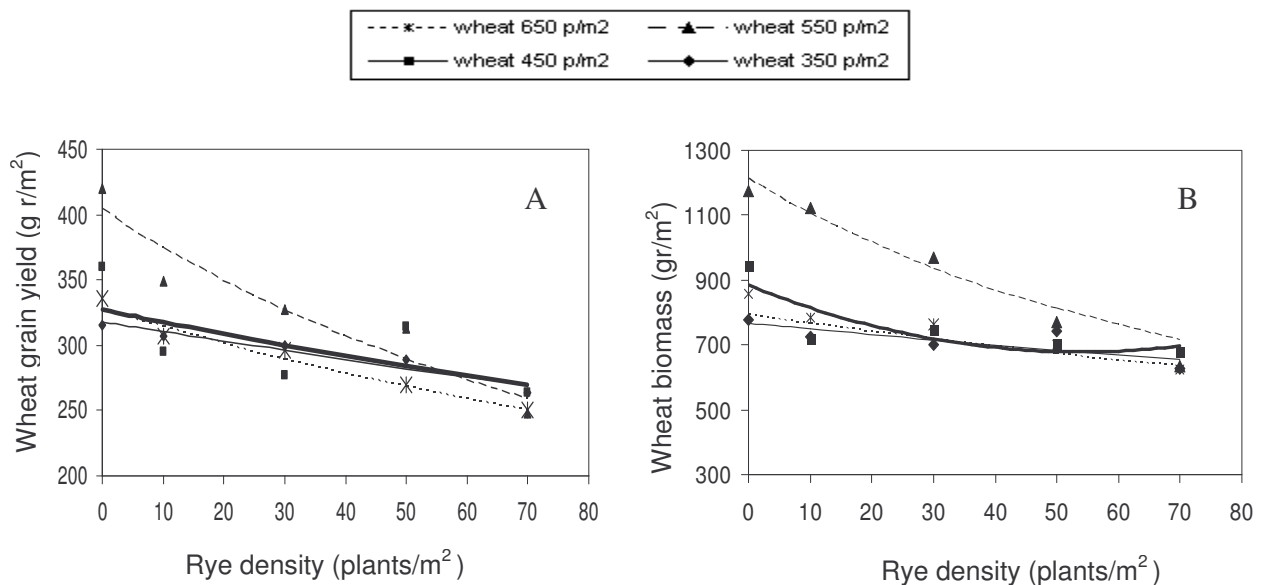


Figure 1- Effect of rye density on winter wheat grain yield (A) and biomass (B)

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# **A comparative morpho–anatomical analysis of aquatic and terrestrial forms of *Ranunculus repens* in the lake district Vlasina, Southeast Serbia**

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## **Introduction**

*Ranunculus repens* is a perennial plant 20-40 cm high, with a short rhizome sending out runners (stolons) above and thin slightly branched roots below. The species is widely spread in Europe and Asia, as well as in North Africa (Coles, 1977). Besides, it expanded its area as adventive species in North and South America, Australia and New Zealand (Benson, 1948; Louertig, 1951). Generally, its habitats are wet and flooded meadows although it appears on cultivated fields where it can come into competition with cultivated plants (Harper, 1957).

The aim of this work was to elucidate the differences between the aquatic and terrestrial forms of *R. repens* found on flooded fields around the Vlasina Lake, in Southeastern Serbia. Structural differentiation of aquatic and terrestrial forms indicates adaptive ability i.e., ecological plasticity of the investigated species in changing environmental conditions on this unstable wet habitat.

## **Materials and methods**

A morpho-anatomical analysis of leaves, stems, stolons and roots of *Ranunculus repens* was performed on material collected in the period from May to October 2002 on flooded fields around the Vlasina Lake at the altitude of 1200 m. Plant material was fixed in the field with 50% alcohol and then the anatomical analysis of plant organs was carried out in laboratory by standard method for light microscopy (Jensen, 1962; Blaženčić, 1994). Leaves, stems and roots were measured with a special image analyzer “Ozarya”. The data were analyzed using “STATISTICA 4.5” for Windows. Comparisons between aquatic and terrestrial plants were analyzed by ANOVA.

## **Results and discussion**

Lower and upper leaves of both aquatic and land forms are triangular-ovate-shaped divided in three lobes and with markedly long petiole. Terrestrial form leaves are longer compared to the aquatic, while they are approximately of the same width. The leaves of both ecomorphs are dorsiventral or bifacial since the mesophyll is clearly differentiated into adaxial palisade and abaxial spongy tissue layers. The whole leaf thickness as well as the thickness of palisade and spongy tissue is higher in terrestrial than in aquatic form.

The stem in both forms is erected, being three times higher in plants from aquatic environment compared to stems of plants from flooded meadows around the lake. Stems of aquatic form usually consist of 4-5 internodes, while in terrestrial form there are 2-3 internodes. In the herbaceous stems of both aquatic and terrestrial form are present only primary tissues, i.e. epidermis, cortex and vascular cylinder. In the center of the mature stem is formed the rhixigenous pith cavity. The terrestrial form stem cortex is more and better developed than aquatic stem, which, otherwise, is characterised by larger parenchyma cells. There are 5-6 vascular bundles in the vascular cylinder of the aquatic form, while in terrestrial form stem they number vary from 13-20. The pith cavity of the stem of terrestrial form is approximately two times larger than in aquatic form stem.

Exodermis, primary cortex and vascular cylinder are clearly differentiated in the roots of both aquatic and terrestrial forms. The cortex diameter in aquatic form is 10% wider than in terrestrial form. There are three xylem strands in the central cylinder of aquatic (triarch root), while in the root of terrestrial form five or more xylem plates can be present (pentarch or polyarch root).

Stolons length of aquatic form of *R. repens* can reach 0.2 m. The structure of stolons is similar to that of stem.

### Conclusions

The differences between terrestrial and aquatic form of *R. repens* are evident in morphological (external) appearance of the whole plant, i.e. its ecomorphs as well as in anatomical structure of above- and underground organs. On the basis of general anatomy the plant belongs to the hygrophytes. The structural differences exhibited in all plant organs of both terrestrial and aquatic forms imply its adaptive ability of the rapid adjustments to the spatial changes in the environment. The aquatic form is characterised by several degrees of general structural reduction, which is in connection with the uniformity of its environmental conditions. More complex anatomical structure attributes the specimens thriving in wet terrestrial environment, which correlate with greater ecological heterogeneity in such type of habitats.

Table 1. Morpho-anatomical characters of aquatic and land forms of *Ranunculus repens* (mean  $\pm$  SE).

Morpho-anatomical character	Aquatic	Land
Leaf length (mm)	18.2 $\pm$ 0.06	32 $\pm$ 0.9
Leaf width (mm)	58.5 $\pm$ 0.2	57.3 $\pm$ 0.2
Leaf thickness ( $\mu$ )	124 $\pm$ 0.1	167 $\pm$ 0.01
Palisade thickness ( $\mu$ )	38 $\pm$ 0.03	55 $\pm$ 0.02
Spongy thickness ( $\mu$ )	60 $\pm$ 0.01	79 $\pm$ 0.03
Stem length (m)	0.124 $\pm$ 0.01	0.043 $\pm$ 0.003
Stem cortex thickness ( $\mu$ )	568 $\pm$ 0.02	1311 $\pm$ 0.09
Pith cavity of stem ( $\mu$ )	478 $\pm$ 0.07	896 $\pm$ 0.09
Cortex diameter of root ( $\mu$ )	2204 $\pm$ 0.02	1904 $\pm$ 0.02

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# Allelopathic effects of *Parthenium* residues: effect of residue age on phytotoxicity

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## Introduction

Allelopathy is a type of chemical interference where a plant releases chemicals that are generally detrimental to the growth and establishment of neighbouring plants (Wardle *et al.*, 1998). It provides a number of advantages to the donor plants such as gaining dominance over the other vegetation, greater competitiveness, and ability to quickly occupy available niches. A number of weed species are known to exploit allelopathic interference in order to gain selective advantage compared to others. *Parthenium hysterophorus* L., an annual exotic weed from tropical America is one such weed that has caused a major problem in the Indian subcontinent due to its allelopathic interference (Kohli & Rani, 1994). A number of allelochemicals such as phenolics and sesquiterpene lactones, particularly parthenin, have been identified from the weed (Kohli & Rani, 1994). Even the residues of the weed that are found in plenty especially at the end of the growing season of the weed have been reported to be allelopathic and thus affect the early growth of next season crop plants (Singh *et al.*, 2003). There is thus a need to manage these residues. However, before any management practice is undertaken, there is a need to understand at what stage of decomposition these residues have minimal allelopathic activity so that these can be utilized as effective bioresources. With this objective, a study was planned to find out the changes in the phytotoxicity of residues at different stages of decomposition vis-à-vis quantity of phytotoxins involved.

## Material and methods

Residues of *P. hysterophorus* were collected from the wild strands of the weed in the agricultural fields after the harvest of summer crops. These were allowed to decompose under field conditions and their phytotoxicity was studied at 0 (freshly harvested), 2<sup>nd</sup>, 4<sup>th</sup> and 6<sup>th</sup> week. Aqueous extracts (1 %, w/v) of residues of different ages were prepared and their effect was determined on seedling growth of mustard (*Brassica campestris* L. var. Pusa Bold) in a 15 cm Petri dish lined with Whatman no. 1 filter paper moistened with 7 ml of extract or distilled water (control). Further, residues of different ages were mixed into soil (10 g/kg soil) and growth of mustard in terms of seedling length (from tip of root to tip of shoot) and dry weight was studied in the amended or unamended control soil two-weeks after sowing. The aqueous extracts and amended soils were analyzed for the presence of water-soluble phenolics as suggested by Swain and Hillis (1959). Data on seedling length and weight is presented as mean  $\pm$  SE and that on phenolic content was analyzed using polynomial regression analysis

## Results and discussion

It is clear from the results that extracts of *P. hysterophorus* residues inhibit the growth of mustard measured in terms of radicle and plumule length and seedling dry weight (Figure 1a). Freshly harvested residues were most phytotoxic and with increasing residue age (decomposition period) the toxicity of the extracts gradually decreased and was almost negligible in 6-week-old residues (Figure 1a). Not only the aqueous extracts, even the residues of different ages amended in the soil also showed a similar growth retardatory effect (Figure 1b). Here also maximum inhibitory effect was observed in the soil amended with fresh residues whereas with older residues of 2-, 4-, and 6-week old the phytotoxic effect decreased (Figure 1b). This indicates that the amount of inhibitors present in *P. hysterophorus*

residues possibly decreases with increasing residue age. To establish this, residue extracts and amended soils were analyzed for phenolics, often implicated in allelopathy (Rice, 1984).

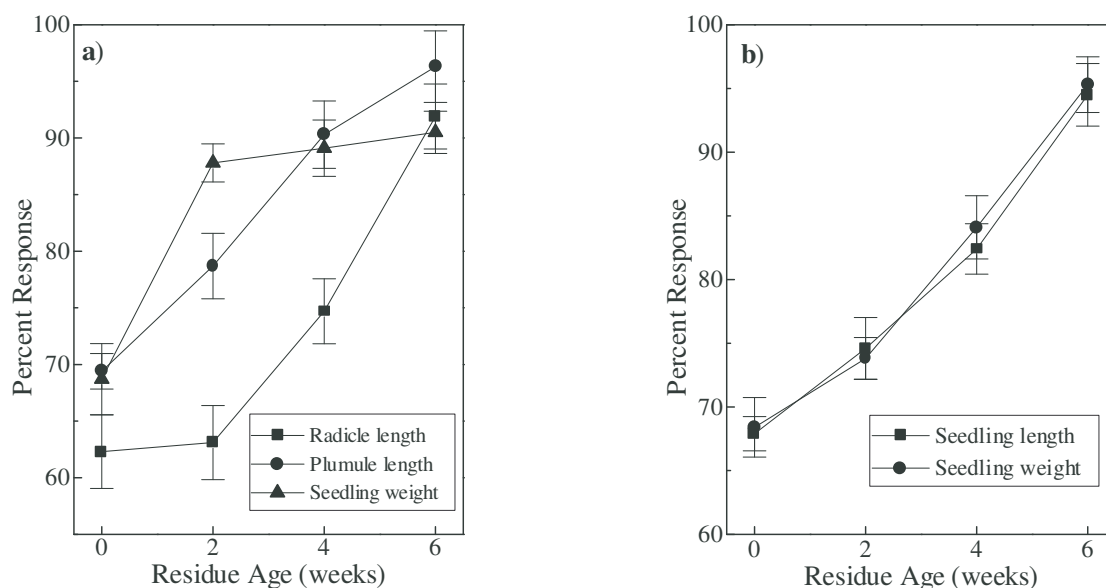


Figure 1. Percent response in comparison to water treated control of seedling growth of mustard in (a) residue aqueous extracts or (b) soil amended with residues of different ages of *P. hysterophorus*.

As expected, the amount of phenolics was the maximum in freshly harvested residue extracts or soils amended with these and it decreased with increasing age of residues. After 6-weeks, phenolic content was reduced by around 75 % of that in fresh residues (Figure 2). In other words, the phytotoxic allelochemicals are gradually lost as decomposition proceeds.

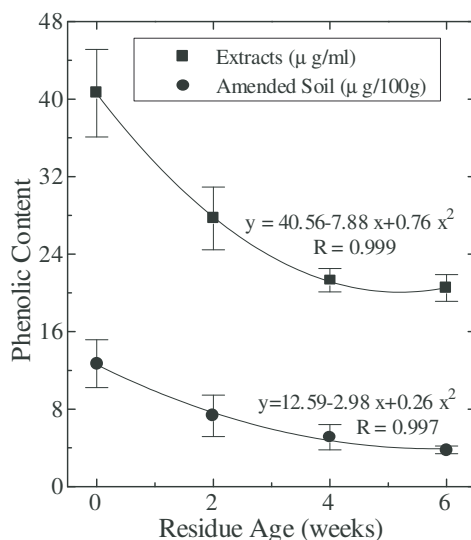


Figure 2. Amount of phenolics in *P. hysterophorus* residue extracts and residue amended soil.

From results, it is clear that phenolics in residues have a direct relevance with observed phytotoxicity. Such observations may be useful for management of *P. hysterophorus* residues.

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# Role of Emergence Time for Weed Seed Production in Corn (*Zea mays*) Crop

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## Introduction

It is widely believed that rational management of weeds in agroecosystems demands knowledge of the biology of the various species (Mortensen et al. 2000), in order to predict and optimize the relationships between agronomic practices and weed growth dynamics. The fast and elevated seed production capacity has led to the definition of ruderal species, which devote most of their energy resources to rapid production of large amounts of seeds. Agronomic measures designed to minimize annual seed production in such ruderal species constitutes a fundamental management practice for prevention of infestation.

Of increasing interest is also the assessment of the seed production capacity of those weed species for which it is not economical to use herbicides, in that such weeds remain below the intervention threshold (Cardina and Norquay 1997). In these cases weeds tend to develop freely and hence succeed in completing their biological cycle and dispersing their seeds.

The aim of this study was to investigate seed production in some important weeds infesting a typical summer crop, assessing seed production as a function of emergence time. In addition, we investigated whether different species respond differently depending on their ability to adapt to the unfavorable shade environment caused by the overlying crop canopy.

## Materials and methods

The test was carried out in 2000 and 2001 on a farm located in Asciano (Pisa) (43° 43' North, 10° 26' East; level ground; 2 m above sea level) on silty clay soil (clay 35%, sand 45%, sand 20%; 1.7 organic matter, pH 7.9) heavily infested by *Datura stramonium*, *Solanum nigrum* and *Abutilon theophrasti*. Corn (*Zea mays* L.) was sown (hybrid FAO 700) on 27 April 2000 and 23 April 2001 at a density of 6.6 plants m<sup>-2</sup>.

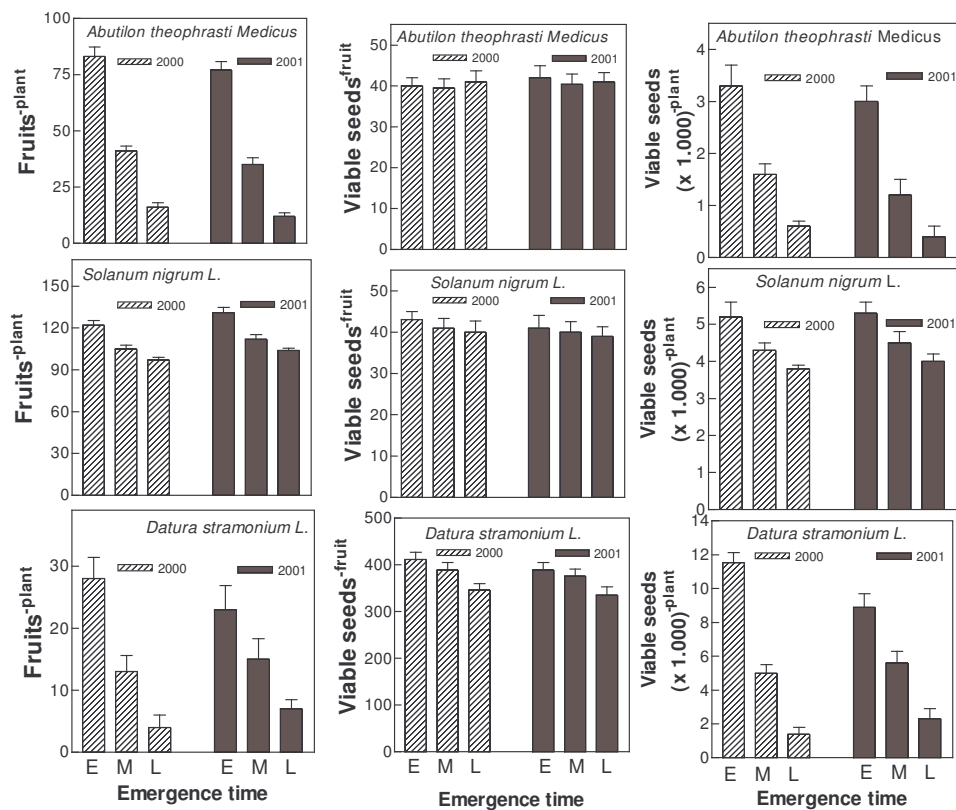
Emerged weeds were identified and labeled with their respective date of emergence in order to group them into three periods: early (emergence peak around early May), medium (emergence peak around early of June), late (emergence peak around early July). Only the three most abundant species were taken into account: *D. stramonium*, *S. nigrum* and *A. theophrasti*, to ensure a sufficient number of emerged seedlings for each of the periods mentioned above. After labeling, the chosen weeds were isolated from other weeds (manual thinning): this was designed to promote competition between crop and weeds only, avoiding undesired and uncontrollable weed-weed interference. Reproductive measurements (number of fruits per plant, number of seeds per fruit) were recorded at the normal time of crop harvest (October). In such cases (not senescent fruits) seed tetrazolium tests were also performed.

The experimental design was a randomized complete block for plant development and the corresponding reproduction study. Seed viability evaluation was based on a completely randomized design. All data were subjected ANOVA using Fisher's Protected LSD test (0.05 level of probability) for means separations.

## Results and discussion

In spite of the various competition strategies adopted by the weeds under investigation, fruit production per plant was found to be strictly dependent on time of emergence (Fig.1). Thus delayed emergence consistently resulted in a significant decrease ( $P = 0.05$ ) in the number of fruits in all species studied. The decrease was, however, species-dependent: late emergence





**Fig.1** Weed reproduction activity (fruit<sup>-plant</sup>; seed<sup>-fruit</sup>; seed<sup>-plant</sup>) as a function of the emergence time (E= early, M= medium and L= late).

led to a 15%, 25% and 75% fruit reduction as compared to early emergence, in *D. stramonium*, *A. theophrasti* and *S. nigrum* respectively. Moreover, the number of seeds per fruit was found to be constant in *A. theophrasti* (around 40 seeds per capsule) light variable in *S. nigrum* and variable in *D. stramonium*.

This considerable reduction in reproductive activity has been reported previously (Benvenuti et al. 1994) but was attributed almost exclusively to the decrease in fruits per plant, since no statistically significant variation in number of seeds per fruit was recorded. As a consequence of the drop in number of fruits per plant and (partially) seeds per fruit, seed production per plant was dramatically reduced with late emergence. When weeds and crops emerged almost simultaneously, *D. stramonium* produced roughly 10,000 seeds and black nightshade about 5,000, while *A. theophrasti* produced over 3,500. However, only *S. nigrum* maintained 75% of the quantity of seeds produced in more favorable growth conditions. This behavior is probably linked to the growth strategy characterizing the latter species, which is capable of competing and reproducing even in the presence of poor light thanks to its photomorphogenic and photosynthetic adaptation. Thus overall, *D. stramonium* and *A. theophrasti* seem to have a competitive-ruderal growth strategy, while *S. nigrum*, although also a ruderal species, appears to have greater stress tolerance and adapts better to growing in shaded conditions. In conclusion, time of emergence is of major importance in determining seed production, but its influence varies as a function of the strategies evolved by weeds to adapt to adverse environmental conditions (*i.e.* shade tolerance developed by *S. nigrum*). Such strategies crucially enable weeds to continue good seed production and thus to preserve the size of the “seed bank” at sufficient levels to ensure survival in a disturbed agroecosystem.

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# Within-field stability of weed density–crop yield interactions in *Lolium rigidum* and *Avena sterilis* mixed populations

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## Introduction

Several crop–weed associations show variation in interference relationships at distinct spatial (i.e. regions, fields) and temporal scales (Moechnig et al., 2003). This variation in predictions of yield loss from theoretical models is relevant for the development of decision support systems for integrated weed management. However, within-field variation has not been described in depth, despite the general recognition of the fractal nature of biological variability. *Lolium rigidum* Gaudin (annual ryegrass) and *Avena sterilis* L. (wild oat) are two of the most troublesome weeds in Mediterranean dryland crops. Weed communities of winter wheat crops in north-eastern Spain are usually dominated by *L. rigidum* and *A. sterilis* mixed populations, and few broad-leaved weeds are present in such agro-ecosystems. In spite of this, attempts to characterize wheat performance and geographical and temporal variability as a result of competition have been made separately for these plants (Lemerle et al., 1995; Murphy et al., 2002); only a study by Pannell and Gill (1994) has considered the effects of mixed populations of *Avena sp.* and *L. rigidum*. Here we present data from a two-year study on natural *L. rigidum* and *A. sterilis* mixture interference on wheat yield, and determine whether yield loss caused by this weed assembly is stable both across locations within a field and interannually.

## Materials and methods

A study was carried out in an 8 ha-field located in Calonge de Segarra (Central Catalonia, north-eastern Spain, 41°45'32" North 1°31'29" East) in 2002 and 2003. Three equally sized field areas measuring 31 m × 51 m (which will be referred to as areas 1, 2 and 3) were selected to represent contrasting topographical positions. Differences in soil texture and chemistry between areas were reasonably small. Twenty four plots of 1 m × 1 m, 9 m apart were delimited within each area. They were georeferenced using Ashtech G-12 DGPS, which provides submetre accuracy. *L. rigidum* and *A. sterilis* density was evaluated in winter, using nine 10 cm × 10 cm samples randomly located within each plot. Wheat yield was measured by hand harvesting four 25 cm × 25 cm quadrats, randomly selected within each plot. Field data were described by means of the model proposed by Swinton et al. (1994), who proposed a reformulation of the rectangular hyperbola to take into account additional weed species; estimated competition curves were tested for significance by means of an approximate F-statistic; lack of fit test and residual analysis were used to test the appropriateness of the model. When regression was significant and error variance between data sets homogeneous, the extra sum of squares principle for regression analysis comparisons was used to evaluate the equality of the estimated parameters among data sets (Ratkowsky, 1983)

## Results and discussion

The multi-species hyperbolic model did not provide a satisfactory fit for area 3 in either year. Some authors have also reported a lack of fit in situations in which crop yield is low. Many factors interact with weeds, thus masking their effect. Area 3 was the topmost of the field; it was also slightly sloping. These factors increase soil erosion and consequently decrease water holding capacity, which can affect wheat growth to a larger extent than weed density.

The regressions between crop yield loss and weed density of each species and year were significant in areas 1 and 2. Nevertheless, there was a high variability in data, which was not completely explained by the inclusion of the two species in the model. All three null hypotheses, testing whether the effect of *L. rigidum*, the effect of *A. sterilis* or the asymptotic yield loss were equal between areas for a given year, were accepted (Table 1). This result indicates that none of these parameters varies at this small spatial scale. To test the stability of parameters across years, data from distinct locations was pooled for each year, as there was no evidence of spatial differences. The effect of *L. rigidum* was significantly higher in the first year (Table 2), indicating that the competitiveness of this weed varies over time. However, while the effect of *A. sterilis* was higher in the second year, the difference is not significant (Table 2). This finding shows that the effect of *A. sterilis* on wheat yield was stable between years. *A. sterilis* is affected by drought to a greater extent than wheat. Thus the lack of effect of *A. sterilis* on yield loss in the first year could be a result of differential effects of water stress on wheat and wild oat. In the second year, when no water stress was detected throughout the growing season, this weed took on an important role in crop competition. Weed-free yield might be the main parameter for evaluating economic threshold because differences determine whether a treatment is worthwhile or not, especially in areas with low crop yields, independently of the effect of weeds. Our results indicate that care should be taken before uncritically applying the same economic threshold criteria in different years and different zones.

**Table 1:** Parameter estimates for each area each year (with their standard errors) and *P*-values from difference tests between areas

	parameter*	area 1	area 2	<i>P</i> -value of test
<b>First year</b>	<i>I (Lolium)</i>	0.110 (0.0285)	0.142 (0.0623)	0.6387
	<i>I (Avena)</i>	1.575 (1.2037)	0.285 (0.3511)	0.2931
	A	146.785 (41.8321)	81.51 (21.623)	0.1647
<b>Second year</b>	<i>I (Lolium)</i>	0.013 (0.0073)	0.001 (0.0105)	0.2865
	<i>I (Avena)</i>	0.003 (0.0043)	1.012 (0.5720)	0.3929
	A	67.784 (62.774)	76.72 (16.3850)	0.8909

*I* represents percentage loss in crop yield per unit of weed density as density approaches zero; *A* represents the maximum percentage crop yield loss asymptote as weed density approaches infinity.

**Table 2:** Parameter estimates for each year (with their standard errors) and *P*-values from difference tests between years

parameter	first year	second year	<i>P</i> -value of test
<i>I (Lolium)</i>	0.129 (0.0298)	0.010 (0.0023)	< 0.0001
<i>I (Avena)</i>	0.163 (0.1780)	0.578 (0.2514)	0.1557
A	111.081 (18.482)	89.667 (30.320)	0.5636

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# Growth Inhibitory Effect of Sumac (*Rhus typhina* L.) Root and Leaf Water Soluble Exudates on Selected Crops

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## Introduction

Plant exudates often exhibit clear allelopathic phenomena upon adjacent vegetation growing within encroaching rhizospheres (Altieri, 1995; Gliessman, 1998; Yamamoto et al., 1999). Therefore, as Zucconi (1996) pointed out, an accumulation of these excreta in the soil may become inimical to crop production thus requiring a better alternation of species cultivation on the farm, including a more appropriate management of soil organic matter. Growth inhibition is exerted also against seed germination, depending upon concentration and characteristics of these allelochemicals (Kefeli & Kalevitch, 2003). The purpose of our study consisted in evaluating the inhibitory effect of phenolic substances extracted from leaves and roots of sumac (*Rhus typhina* L.) on root development of cuttings of willow (*Salix discolor*) and bean (*Phaseolus vulgaris*). Sumac trees abound as pioneer species in early succession in western Pennsylvania and appear to possess potent allelopathic capabilities. We support the idea that isolating sumac exudates and studying their effect upon root formation and seed germination of unrelated plant species could enhance an interest in developing more environmentally friendly herbicides. Our effort aims at promoting a sustainable approach to agriculture in modern weed control and management.

## Materials and methods

Sumac abscised leaves (2 kg) and roots (2 kg) were macerated for a total of 30 days in two separate water tanks (20 L), at room temperature (24°C) in our laboratory. Concurrently, we germinated four samples (n = 15) of bean seed by soaking them in water for 24 hours, prior to planting them in potting soil, at one-week interval, in order to produce cuttings of consistent length (16 cm) with those of willow. The latter were obtained from shoots collected from fully-grown trees. We immersed bean and willow cuttings in containers filled with sumac water extracts at one-week interval since the beginning of the maceration process. Through paper chromatography we were able to isolate and identify four different kinds of allelochemicals (flavonoids, anthocyanins, coumarins and phenolic acids), as proposed by Furuya (1965) and Turetskaya et al. (1968). The pH of the sumac macerates was also measured weekly, throughout the duration of the experiment. A two-factor ANOVA consisted in the methodology applied to verify statistical significance between the two different, rooting treatment conditions, being the concentration of sumac leaf and root exudates the two interdependent variables of this study. We analyzed our data with SPSS software.

## Results and discussion

Initially, (at the end of the first week) the sumac leaves macerate had a pH of 3.5 whereas the root-water solution had a lower level of acidity (pH=5.6). Kefeli and collaborators (2003) argued that lower pH levels indicate high concentrations of phenolic compounds, thus explaining the more powerful growth inhibiting action exerted by the sumac water-extract upon our two test crops. The data analysis revealed that root growth inhibition is significantly diminished with the age of the sumac leaf and root water extracts.

Table 1. Number of roots grown over course of treatment in bean cuttings (SEs in brackets).

Sumac water extract from	Week			
	1	2	3	4
Leaves	0	1 (0.2)	2 (0.3)	5 (0.5)
Roots	6 (0.5)	7 (0.5)	10 (0.9)	12 (0.9)

Table 2. Number of roots grown over course of treatment in willow cuttings (SEs in brackets).

Sumac water extract from	Week			
	1	2	3	4
Leaves	2 (0.2)	3 (0.2)	3 (0.3)	4 (0.2)
Roots	4 (0.2)	4 (0.2)	5 (0.3)	5 (0.3)

High levels of acidity in the water extract had an effect in inhibiting root formation and elongation. Additionally, we observed that the root length of both types of cuttings was affected by a low pH. The aging of the water extracts attenuated root growth inhibition. As Wallace and Fry (1994) indicated, there is a possibility for microorganisms to inactivate phenolic compounds, or converting these molecules to less effective allelochemicals. In accordance with this hypothesis, we recommend an emphasis about the role of microorganisms on the effect of plant exudates in future studies.

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# Double-row planting influences the critical period for weed control in early-planted maize

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## Introduction

Determining the critical period (CP) of weed competition can help us to minimize weed interference and to design the best weed management system (Knezevic et al., 2002). Several factors such as climate, genotype, cultural practices (weed composition and density, planting date or seeding rate, tillage, fertilization) may influence the CP (Norsworthy and Oliveira, 2004). Several studies have shown that narrower rows or higher maize population may lead to a shorter CP because of quicker canopy closure and higher crop competitive ability (Teasdale 1995). Planting maize at double-rows could be very easy with the recent introduction of several makes of planters that have easily adjustable planting row length. Maize yield data suggest that increased plant population has frequently manifested yield losses and lodging particularly at double density (2X) populations (Teasdale 1998). However, under Greek conditions, previous research (Chachalis and Zanakis, unpublished data) has shown that early- or ultra-early planted maize may be more suitable for a higher population density. One reason might be the lower maize biomass and higher root/shoot ratio that reduces the intracrop competition for light and nutrients. The objective of this research was to determine the CP in a double-row system with different plant populations in early-planted maize.

## Materials and methods

Field experiments were conducted at Armenio, Larisa, Greece under normal agronomic practices in 2003 and 2004. Maize was planted at double rows (20 cm apart) at three densities (1X, 73,000 plants/ha; 1.35X, 98,550 plants/ha; 2X, 146,000 plants/ha). Trials were established as a randomized complete block with a split-plot design, with plant densities as the main plot (4 m length) and duration of weed competition as sub-plots. Weeds were allowed to grow for 2, 4, 6, 8, or 10 weeks after emergence (WAE) and then the crop was maintained weed-free until the final harvest (early competition treatments); crop were maintained weed-free for 2, 4, 6, 8, or 10 WAE and then weeds were allowed to emerge and growth until the final harvest (late competition treatments). Sub-plots with weedy and weed-free crop for the whole cycle were also included as checks. A Gompertz curve [ $y = a \exp(-b \exp(-kT))$ ] for early competition treatments and a logistic curve [ $y = [(1 / \{\exp[c*(T-d)] + f\}) + ((f-1)/f)] * 100$ ] for late competition treatments were fit to observed yield data (Norsworthy and Oliveira, 2004). Parameters for the Gompertz curve were as follows:  $a$  is the yield asymptote,  $b$  and  $k$  are constants, and  $T$  is time after emergence. Parameters for the logistic model are as follows:  $T$  is the time after emergence,  $d$  is the inflection point, and  $c$  and  $f$  are constants. Critical period was determined from the curves as a 5% reduction of the yield.

## Results and Discussion

Naturally occurring weed populations included mainly *Solanum nigrum*, *Chenopodium album*, *Amaranthus retroflexus*, *Convolvulus arvensis*, *Sinapis arvensis*. The results of 2004 showed that yield was the highest at 1.35X planting rate (i.e. 98,550 plants/ha) in both weedy and weed-free treatments (4,390 and 15,030 Kg/ha, respectively). These findings agree with those of others that maximum yield was obtained at 96,000 pl/ha (Teasdale, 1998). The CP was longest (4.5 to 6.7 WAE) at 2X double-row planting than that of 1X (4.2 to 5.6 WAE)

(Table 1). At 1.35X planting density the critical period was non-existent. Practical implications are that high density through double-row planting although increases the critical time for weed removal and requires a longer critical period for weed control than that of normal density of double-row planting.

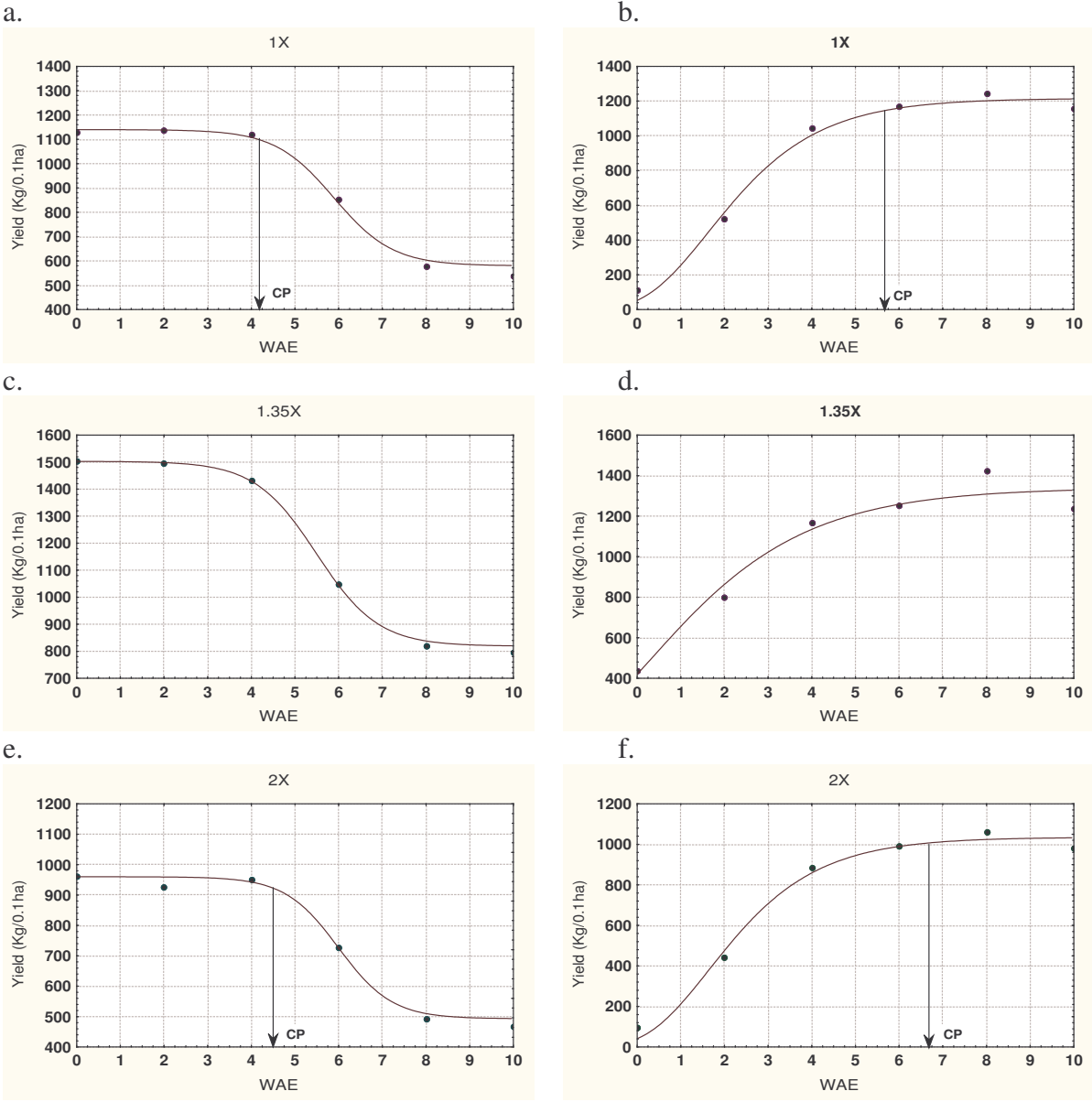


Figure 1. The influence of weed-free (a, c, e) and weed interference (b, d, f) at various durations of weeks after emergence (WAE) and different plant densities 1X 73,000 p/ha (a, b), 1.35X (c, d), and 2X (e,f). Arrows indicate critical periods (CP).

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# Integration of the influence of morphological plasticity on plants competition for light

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## Introduction

A plurispecific weed population dynamics model is currently developed to simulate the long-term evolution of the weed flora in field crops in relation to cultural practices (Collard et al., 2004). Accurate weed seed production modelling is needed to improve estimation of seed return to the soil seed bank and weed population in later years. Weed seed production is firstly determined by crop/weed competition. Most existing mechanistic competition models (Kiniry et al., 1992; Kropff et van Laar, 1993) simulate weed biomass per unit area. However as plants interact with their neighbours, heterogeneity in crop spatial arrangement at the very local scale creates different growing situations and can result in considerable variability in the number of seeds produced by individual weed plants. As a consequence modelling per unit area can lead to under- or overestimation of weed seed production, depending on the competitive environment.

Weeds can exhibit strong morphological plasticity in order to best adapt to their competitive environments. As a result, morphological parameters, like the stem biomass/leaf biomass partitioning ratio (SLR) or the specific leaf area (SLA,  $\text{cm}^2.\text{g}^{-1}$ ) can show important variation. In order to incorporate morphological plasticity into crop/weed modelling, those parameters are related to shade that plants are exposed to (Cavero et al., 2000; Brainard & Bellinder, 2004). So far models developed in this way remain mono-specific and do not account for possible response of morphological plasticity by weed plants before direct competition for light (Ballaré et al., 1990). Generic models that can deal with most weed species are still to be developed. Objective of this paper is to present first modelling and experimental work to build such a model.

## Materials and methods

To simulate light absorption and plant growth at the individual scale, a 3D modelling approach was used. Space discretization in 'voxel' is based on the TROLL model (Chave, 1999). In our model, voxels are elementary volumes of area 5 cm x 5 cm and height 10 cm where light absorption is computed. Individual plants are defined by their location, biomass, height, breadth and leaf area profile. In order to have a common representation for most species, plants are represented in the 'voxelised' space by a cylinder with diameter and height corresponding to the plant breadth and height (Fig. 1).

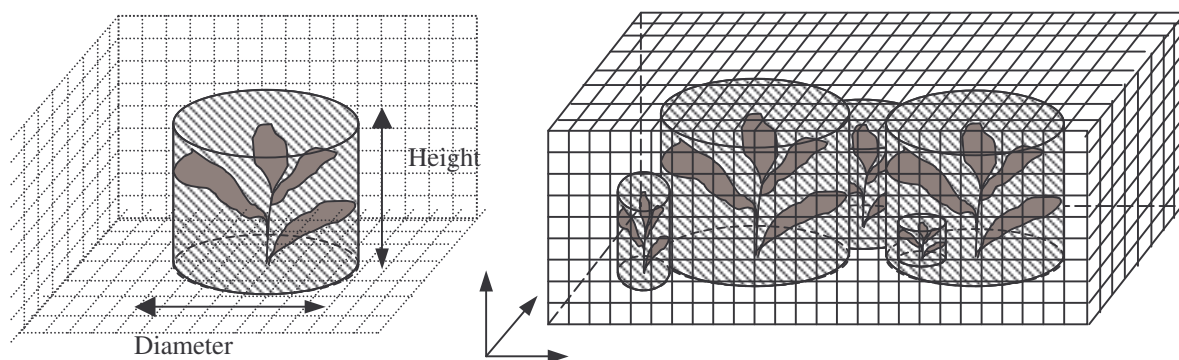


Figure 1. Geometrical description of a plant and its representation into the 'voxelised' space.

Daily light absorption was computed through the Beer's law and photosynthesis was simulated to calculate daily biomass assimilation of each plant. So far only the light interception sub-model has been developed. The specific plant height (SPH, cm.g<sup>-1</sup>) and diameter (SPD, cm.g<sup>-1</sup>), SLR, SLA and leaf area profile were used to determine daily plant growth and morphology.

Because those parameters are subject to morphological plasticity, a field experiment was conducted to parameterise relationships between them and the competitive environment that plants experience. Different competitive environments were produced by creating gaps of variable diameter (240 cm, 100 cm, 40 cm) in an oilseed rape crop, sown on September 10<sup>th</sup>, 2004. Seedlings of 5 weed species (*Amaranthus retroflexus*, *Alopecurus myosuroides*, *Galium aparine*, *Sinapis arvensis*, *Stellaria media*) were planted out on September, 18<sup>th</sup> at the centre of the gaps. Two other situations were created by planting out weed seedlings either in isolated conditions or in the oilseed rape crop with no gap. Such experimental design was used to delay the onset of competitive interactions for light between crop and weeds and to assess early plant response to it. Light sensors were placed in the different situations, at 4 cm height, to measure the share of the incoming radiation that is absorbed by neighbouring plants. Individual weed plants were harvested regularly. A picture of the profile of each harvested plant was taken at harvesting so that plant height, diameter and leaf area profile can be evaluated through image processing. Leaf area and leaf and stem weights were measured. To define competitive environment of weeds, we also harvested neighbouring oilseed rape plants, take a picture of them and measure their leaf area.

### Results and discussion

On November, 3<sup>rd</sup>, weed plants in the situations 'no gap' and '40 cm' were exposed to reduced light levels (9 and 65% of the incoming radiation, respectively). This results for the species *A. myosuroides*, *G. aparine*, *S. media* and *S. arvensis* in an increase of SPH, in the situation 'no gap' as compared to the situation 'isolated', of 266%, 490%, 500% and 410%, respectively. SLA is also increased by 17%, 39%, 55% and 34%, respectively. Increase in SLR was only observed for the species *S. arvensis* (700%) which exhibited a very strong modification of its stem size. Morphology of *A. retroflexus*, a summer species, was not affected by shading treatments as it could not grow long enough. At that time, proximity of neighbouring plants without direct competition for light (situation '240 cm' and '100 cm') did not cause important modification of plants morphology, regardless of weed species.

Integrating morphological plasticity in competition models requires to link together variables characterising plant morphology and competitive environments that plants experience. Data collected on neighbouring plants will be used in the light absorption sub-model to calculate a shading index for each weed plant. The shading index is defined as the proportion of the incoming radiation that reaches the plant. Relationships will be drawn up between the index and the morphological parameters and integrated in the competition model to allow weed morphology to be affected by their local competitive environment and eventually to improve simulation of weeds biomass and seed production.

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# Testing a simple approach to estimating the seed production of arable weeds within contrasting crops

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## Introduction

Weed seed production data are required within population dynamics models to improve the understanding of weed species lifecycles (Lutman, 2002; Norris, 2003), particularly those of high biodiversity value. Such information is lacking for many species (Wilson & Lawson, 1992; Lutman, 2002) and is vital for validating future weed decision support systems, aimed at targeting weed control to prevent crop yield loss whilst retaining beneficial weed species.

A number of researchers have defined reproductive effort (or reproductive allometry), as measured by the ratio of vegetative to reproductive biomass at maturity, as a stable characteristic of individual species. If this trait is constant across contrasting environments it provides a useful simple approach to estimating potential seed production in any cropping situation by measurement of total weed biomass.

To demonstrate the robustness of this approach in both winter and spring crops, the seed production of 6 common UK arable weeds has been investigated through a series of field experiments, over a three year period (2002-2004). In addition to validating the estimation of seed production these results will provide information to quantify potential weed fecundity in different crops within arable rotations. Target species were *Polygonum aviculare*, *Chenopodium album*, *Sinapis arvensis*, *Papaver rhoeas*, *Veronica persica* and *Fumaria officinalis*, although not every species has been sampled in every crop.

## Materials and methods

Plant samples were taken from fields at ADAS Boxworth (Cambridgeshire), Terrington (Norfolk) and High Mowthorpe (Yorkshire), from a range of winter and spring sown crops (winter and spring wheat, field beans, oilseed rape and sugar beet), between May and July of 2002, 2003 and 2004. Each individual species were measured differently according to the nature of the plant and sampling times varied according to the flowering times of each species. All weeds sampled were natural populations identified at each site.

*Plant sampling.* For each individual species sampled, 20 plants were randomly collected from within untreated field trial plots. Samples were labelled individually and assessed in the lab. The total dry weight (g) per plant, the ratio of reproductive to vegetative dry weight (g) per plant and the number of seeds per plant were recorded. For each of the 20 plants the number of immature and ripened capsules or pods were counted, including any capsules that had already shed their seed. The plants were then dissected into a) reproductive parts (capsules or pods excluding any stalks) and b) non-reproductive parts (leaves, stem and any stalks). The reproductive and non-reproductive parts were then dried in an oven at 80°C for 48 hours and the dry weights (g) were recorded.

*Number of seeds per capsule.* The average number of seeds per capsule was assessed by sampling a further 30 capsules or pods, from within the same field area as the main plant samples were taken. The plant heads or capsules selected were mature heads that had not already senesced and shed their seed, but were not immature. In the case of *S. arvensis*, the heads were not flowering on collection, but for *C. album*, *P. aviculare*, *V. persica*, *F. officinalis* and *P. rhoeas* some flowers were still present despite being mature. Each capsule was assessed individually and the total number of seeds per capsule was counted. Species with high numbers of seeds per capsule, such as *P. rhoeas*, required a seed sub-sample for assessment.

*Data analysis.* Data sets were compared using simple linear regression in Genstat (Payne et al., 1995) and averaged together for both winter and spring crops.

## Results and discussion

The average number of weed seeds per g, meaned across all crops is shown in Table 1. The  $R^2$  value (Table 1) is of the linear regression between seed production and total plant biomass.

Table 1. Weed seed production for six common arable weed species averaged across a range of winter and spring crops.

Species	No Seeds/g DW ( <i>s.e</i> )	Crops*	Number of trials	$R^2$
<i>Chenopodium album</i>	1584.2 (92.7)	SW, Sbeet	2	0.84
<i>Polygonum aviculare</i>	205.7 (15.2)	WW, SW, Sbeet	3	0.86
<i>Sinapis arvensis</i>	138.5 (10.1)	WW, SW, SB	3	0.74
<i>Veronica persica</i>	709.7 (46.9)	WW, SW	3	0.79
<i>Papaver rhoeas</i>	1856.0 (134.0)	WOSR, SOSR, SB	6	0.73
<i>Fumaria officinalis</i>	233.5 (24.4)	WW, SW, SB	3	0.66

\*WW = winter wheat, SW = spring wheat, WOSR = winter oilseed rape, SOSR = spring oilseed rape, SB = spring beans, Sbeet = Sugar beet

There were no significant differences between seed production in the winter or spring crops for *V. persica* or *P. aviculare*. Both *S. arvensis* and *F. officinalis* showed significant differences ( $P < 0.001$ ) between the number of seeds produced in both winter and spring wheat and spring field beans. Similarly *C. album* showed significant differences ( $P < 0.001$ ) between the seed production in two spring-sown crops, spring wheat and sugar beet. However, for *P. rhoeas* there were no significant differences between spring and winter oilseed rape or spring beans, but a significant difference ( $P < 0.001$ ) between the seed production of two different crops of winter oilseed rape.

A difficult aspect of assessing weed seed production in natural weed populations in the field is the timing of sampling. Seed production occurs over a protracted period of time, varying not only between different species, but also within different populations of the same species (Norris, 1996). The methodology used by Norris (1996) involved collecting seed rain at 10-day intervals until completion. In these experiments seed production was only assessed on one occasion, resulting in an underestimation of total seed production, due to losses from seed rain or predation. However, plants were growing in competition with the crop and other weed species, representing a natural population. These data are used in weed population dynamics models to determine the impact of rotational weed management strategies in arable crops.

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## Patterns in *Setaria* Seedling Emergence

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### Introduction

Knowledge of a weedy pests behavior, especially at the critical time of seedling emergence and agro-community assembly in annual cropping systems is crucial to timing of control tactics, management of risk and optimization of economic returns from investment. Predicting seedling emergence is a worthy goal and a complex biological problem. Three morpho-physiological mechanisms regulate *Setaria* behavior via transduction of oxygen-water signals received from the soil environment. During the seeds' life in the soil the inherent dormancy of an individual seed responds to these oxygen-water-temperature signals determining its behavior in the soil. Three "maps" are defined by several studies from our research (Dekker et al., 2003; Aspects of Applied Biology 69:247-259) : 1] maps of dormancy heterogeneity shed by individual plants; 2] maps of calculated environmental oxygen-water-temperature signals received by those seeds in the soil; and, 3] timing of emergence of these seeds over several years after entry into the soil. Forecasts of *Setaria* seedling emergence can be made based on several different strategies including predictions of emergence responses (map 3) based on known dormancy (map 1) responding to actual field conditions (map 2). Herein we provide a prediction schema based on observed, historical, consistencies in patterns of seedling emergence (map 3). This consistent pattern of seedling emergence timing is the "realized hedge-bet" for individual *Setaria* fitness directly arising from the heterogeneous seed dormancy "blueprint" instilled during embryogenesis.

### Methods and materials

Seed of 44 locally adapted weedy *Setaria faberii* populations were buried in the soil (0-10 cm) at two different common nurseries (Crawfordsville and Ames, Iowa) in autumn 1997, 1998 and 1999. Weekly counts of seedlings emerged were recorded in the burial year and well as year 1, 2 and 3 (the majority ended after year 1). The field plots were planted to no-till soybeans (*Glycine max*) for the duration of the experiments to ensure agricultural seed bank conditions.

### Results and discussion

Seedling emergence in the first half of the season (Julian weeks 14-31) showed a complex oscillating pattern (fig. 1). This was unexpected, and occurred in every individual population studied. These changes were gradual and continuous over the first half of the season; there were not large week to week oscillations. There are two possible explanations for this oscillating pattern. One may arise from inherent somatic polymorphism in seed dormancy states. The seed rain may consist of several discrete dormancy phenotype cohorts, each with different requirements for germination. This suggests that the spring emergence pattern would not be a single distribution, but a mixture of several—one for each dormancy phenotype cohort. The alternative explanation is that there is an unknown mechanism within the seed that inhibits germination despite these favorable conditions, thus resulting in the observed oscillating pattern. Of the two possible explanations, the first is the most logical and does not require an unknown mechanism.

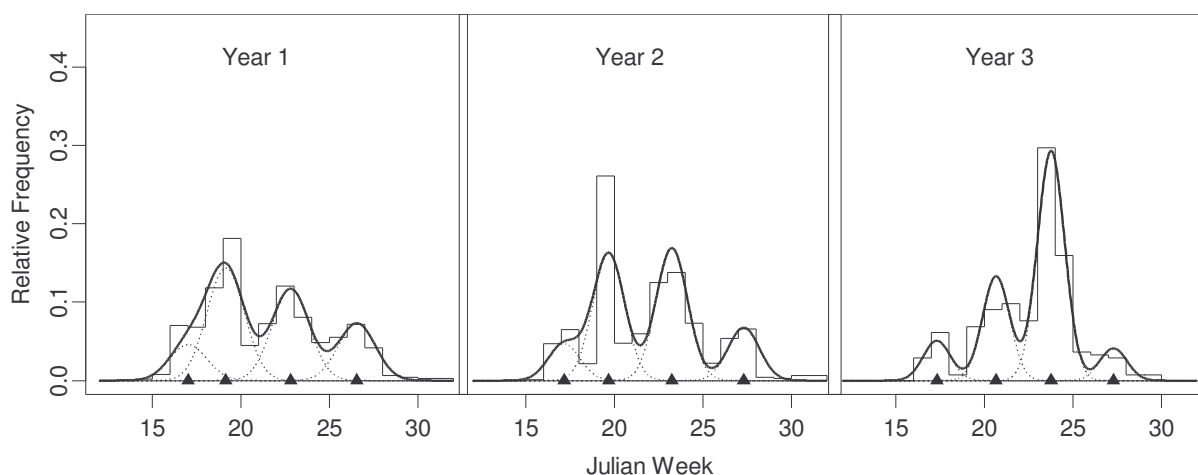


Figure 1. Relative frequency of seedling emergence of buried *Setaria faberii* (giant foxtail) populations in year 1 (left), year 2 (middle) and year 3 (right); bars are the histogram of data; solid line is the mixture model (see below); dashed lines are the four normal distributions components of the mixture model; triangles indicate the means of each normal distribution.

If we assume distinct dormancy phenotypes, the emergence pattern of a population can be represented with a mixture model, a model consisting of a mixture of two or more simple distributions. A mixture model which consisted of four normal distributions—and thus four distinct phenotype cohorts—was most appropriate. To relate seedling emergence to the timing of farming practices in typical summer annual cropping systems used in Corn Belt USA, these four normal distributions were identified and associated with julian week, calendar dates, seasonal name and management name (Table 1).

Table 1. Timing and names for each of the four normal distributions. Julian week and date are the model estimates for all data combined.

Distribution	Julian Week (All data)	Dates	Season Name	Management Name
1	17	April 23-29	Early Spring	Corn Planting
2	19.1	May 8-14	Mid Spring	Soybean planting
3	22.9	June 3-9	Late Spring	Late Planting
4	26.6	June 28-July 4	Early Summer	Corn Layby

The parameter that most affected the relative amount of seedling emergence in each distribution period was year. Differences in the timing of the distribution means had a relatively small effect on the overall patterns, although they tended to be later in the Crawfordsville nursery compared to that at the Ames nursery: the timing of these distributions was stable over years and locations. A general trend can be seen when comparing the emergence in the spring of the first, second and third years after burial. The first year, the largest peak is from the mid spring distribution. The second year, the mid and late spring distributions are more equal in size. The third year, the late spring distribution forms the largest peak. This implies a shift in emergence timing with seed age in the soil to later emergence.



## Allelopathy: from field evidences to agronomic utilizations

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### Introduction

Synthetic herbicides have been used successfully for decades for weed control in agriculture. Unfortunately, their widespread utilization generates increasingly acute problems such as the loss of botanical diversity in agricultural areas, the development of herbicide-resistant weed biotypes and the contamination of surface and ground water. In this context, allelopathy - *the effect of one plant on other plants through the release of chemical compounds into the environment*- is presently receiving an increasing interest in agriculture. Obviously, a better understanding of this phenomenon could offer promising alternatives in weed management and help reduce synthetic herbicide applications (Bhowmik & Inderjit, 2003). However, the real relevance of allelopathy in ecosystems and agro-systems is presently still strongly under debate. The discussion feeds mainly on the experimental difficulty in discriminating allelopathy from competitive effects. Traditionally, interferences among plants have been attributed mainly to competition for common resources, such as space, light, water and nutrients. The design of experiments, which clearly demonstrate the occurrence and importance of chemically mediated interactions among plants, is not trivial, especially at the field level (Inderjit & Callaway, 2003). Still, new techniques and improved methodological approaches have now been applied to research in allelopathy and recent results plead for its importance in both natural (Bais et al., 2003) and agricultural ecosystems (Kholi et al., 2001). In this presentation, we provide some examples of field evidences about the phenomenon of allelopathy and discuss some recent developments for its utilization in weed management.

### Demonstration of allelopathy at the field level

A recent and well-documented example of allelopathy in the field concerns the invasive behaviour of the Eurasian spotted knapweed, *Centaurea maculosa*, in North-Western United States. Bais et al. (2003) showed that *C. maculosa* produces and exudes a very potent phytotoxine ((-)-catechin) in the soil, around its roots, at concentrations largely sufficient to inhibit the germination and growth of native North American species. These findings strongly suggest that chemical interactions might play an important role in the displacement of native plant species. Another example is given by *Artemisia annua*, a plant producing artemisinin, a sesquiterpene lactone endoperoxide with known phytotoxic properties (Duke et al., 1987). Using two *A. annua* strains characterized by low and high artemisinin contents, we were able to demonstrate, in laboratory, glasshouse and field experiments, the strong inhibitory effects of artemisinin on weed development, and the reality of the phenomenon of allelopathy. In all the trials the same trends were observed: a strong and significant inhibition of the emergence and growth of crops and weeds was measured with artemisinin-rich plants, while no or only slight effects were observed with the artemisinin-poor strain (Delabays et al., 2004).

### Utilization of allelopathy in weed management

Schematically, we distinguish between three approaches to exploit allelopathy as a means of weed control in agriculture:

- install allelopathic cover crops in the rotation on arable fields and manipulate their residues (Foley, 1999).
- install allelopathic ground covers in perennial crops (Delabays et al., 2000),

- grow crop cultivars with allelopathic properties that enhance the competitive ability of these crops against weeds (Wu et al., 1999).

Using allelopathic cover crops for weed management has already been largely documented (Kholi et al., 2001). A good example is provided by Sorghum (*Shorghum bicolor*), a plant producing, amongst other allelochemicals, sorgoleone. This long chain hydroquinone induces inhibition of photosynthesis in higher plants (Weston et al., 1999).

Concerning the installation of allelopathic ground covers, we have initiated in Switzerland a research program aimed at the selection of plants offering a good soil protection and an efficient weed control, together with a low impact on crop yield, when sown in the inter-rows in vineyards and berry fields (Delabays, 2000). Several species, such as *Agrostis tenuis*, *Bromus tectorum* and *Hordeum murinum*, offer excellent weed suppression in the field, while revealing strong allelopathic properties in laboratory and glasshouse bioassays.

Lastly, one of the most promising projects in the breeding of crop cultivars with allelopathic properties is carried out on rice (Olofsdotter et al., 2002).

### Prospects

The identification of the natural compounds responsible for the allelopathic effects is crucial in order to design and carry out convincing experiments aimed at demonstrating the reality of allelopathy. Such identification is also required to optimize the utilization of this phenomenon in weed management as this largely relies on the breeding of highly allelopathic plants: crop cultivars, cover crops or ground covers. Breeding procedures are greatly facilitated once the molecules involved are known. As a matter of fact, artemisinin-rich strains of *Artemisia annua* are precisely the result of a breeding programme aimed at enhancing the production of the molecule in the plant (Delabays et al., 2001), which resulted also in a clear increase in the allelopathic properties of the plant.

More studies are still needed to better understand and master allelopathy, but today, accepting that resource and non-resource mechanisms may work simultaneously in plant-plant interference is certainly the most constructive approach.

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# The influence of plant residues on the germination and sprouting of *Agropyron repens* and *Galium aparine*

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## Introduction

Allelopathy is defined as any direct or indirect inhibitory or stimulatory effect by one plant on another through the chemical compounds that escape to the environment (Aldrich and Kremer, 1997). Allelopathy has been the subject of numerous modern research efforts. Putnam and Duke (1974) showed that some crops (wheat, barley, oats, rye, etc.) leach toxic substances from their alive and dead roots, which can reduce weediness of the field with its negative allelopathic influence. Shilling *et al.* (1995) reported that when soybean and sunflower grow in dried green rye, without soil tillage, the mass of lambsquarters, ragweed and redroot pigweed are reduced by 99, 92 and 96%, respectively. Chung and Miller (1995) observed that alfalfa residue mixed with sand at rates of 1 to 2 g kg<sup>-1</sup> reduced growth of lambsquarters, pigweed, crabgrass and velvetleaf. Sati *et al.* (2004) reported that during early phase of decomposition wheat biomass released more phytotoxins than at later stages of decomposition. The objective of this work was to investigate the influence of the plant residues on the number, height and biomass of green weed seedlings.

## Materials and methods

The effect of plant residues of wheat (*Triticum aestivum*), barley (*Hordeum vulgare*), rye (*Secale cereale*), potato (*Solanum tuberosum*), tomato (*Solanum lycopersicum*), and mugwort (*Artemisia vulgaris*) on germination and growth of quackgrass (*Agropyron repens*) and cleavers (*Galim aparine*) was investigated. 10 grams of dry plant residues mixed with about 2.7 kg of sandy clay soil were put in a pot and 50 germinable weed seeds were sown at a depth of 1.5 cm. A check without plant residues was included in the experiment. A randomized block design with four replicated was used. Three different experiments were carried out: 1) with wheat, rye and barley residues, 2) with tomato and potato residues, 3) with 10 and 15 g dw pot<sup>-1</sup> of *Artemisia*. Each experiment lasted for one month and were carried out two times (except that with mugwort residues). Experimental data were statistically analysed by ANOVA and means compared by LSD. The average of two experiment were shown in the tables.

## Results and discussion

The wheat straw inhibited the biomass of *A. repens* seedlings by 32.1%, while rye residue significantly diminished height and biomass of quackgrass by 12.4 and 40.2%, respectively (Tab. 1).

Potato and tomato plant residues diminished the investigated parameters of quackgrass (in average): the number by 20% and 22%, the height by 11% and 14%, and the mass by 25% and 34%, respectively (Tab. 2).

Residues of *A. vulgaris* stimulated only the number of germinated of *A. Repens* seedlings. When 10 grams of mugwort residues were placed in pot it caused stimulation by 18%, while 15 grams did it by 19% (Tab. 3).

The straw of wheat, rye, and barley (Tab. 4) did not inhibit the investigated parameters of *Galium aparine*. But, only rye caused stimulation of height and biomass of *G. aparine* seedlings (by 9.3 and 15.7%, respectively)

From the previous results it can be concluded that plant residues of potato and tomato showed allelopathic potential against *A. repens*. In this work a strong allelopathic potential against *A.*

*repens* was not obtained by rye, wheat and barley. Cleavers, one of the most important weeds in small grains, were stimulated when germinated and sprouted with rye straw. Kazinczi *et al.* (1998) found that shoot residues of *G. aparine* promoted the development of winter wheat.

Tab 1. The effect of residues of wheat, rye, and barley on the number, height and biomass of *Agropyron repens* seedlings (in percent respect to the check)

Plant residues	<i>Agropyron repens</i> seedlings		
	No. per pot	Height (cm)	DW biomass (g per pot)
Wheat	100.3	90.9	67.9*
Rye	93.3	87.6*	59.8**
Barley	91.5		83.2

\*\*significant at P=0.01; \*significant at P=0.05

Tab 2. The effect of residues of tomato and potato on the number, height and biomass of *Agropyron repens* seedlings (in percent respect to the check)

Plant residues	<i>Agropyron repens</i> seedlings		
	No. per pot	Height (cm)	DW biomass (g per pot)
Potato	80**	89*	75**
Tomato	78**	86*	66**

\*\*significant at P=0.01; \*significant at P=0.05

Tab 3. The effect of residues of mugwort on the number, height and biomass of *Agropyron repens* seedlings (in percent respect to the check)

Mugworth residues (g)	<i>Agropyron repens</i> seedlings		
	No. per pot	Height (cm)	DW biomass (g per pot)
10 g	118*	106	115
15 g	119*	101	113

\*significant at P=0.05

Tab 4. The effect of residues of wheat, rye, and barley on the number, height and biomass of *Galium aparine* seedlings (in percent respect to the check)

Plant residues	<i>Galium aparine</i> seedlings		
	No. per pot	Height (cm)	DW biomass (g per pot)
Wheat	103.7	96.2	
Rye	104.7	109.3*	115.7*
Barley	111.7	106.9	95.9

\*significant at P=0.05

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# Gene flow between oilseed rape (*Brassica napus* L. *oleifera*) and black mustard (*Brassica nigra* L.)

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## Introduction

Over the past decade, plant genetic engineering techniques have been developed where specific characters (genes) can be introduced into a plant in a relatively straightforward manner, provided the genes coding for the character have been identified. Oilseed rape crops (*Brassica napus* L. subsp. *oleifera*) can suffer severe yield loss due to weed infestation. Since *Brassica* species can be readily transformed, and specific genes have been identified for herbicide resistance, it is no surprise that genetic engineers have targeted herbicide resistance in oilseed rape (Moloney et al., 1989). A major concern of introducing herbicide resistant crops into agriculture is the spread of the engineered gene, particularly by pollen, to related weed species. The opportunity for gene escape via hybridization depends upon the presence of wild relatives capable of crossing with the crop under natural conditions (Ellstrand, 1988). There is substantial recent literature on intergeneric crosses within the *Brassicaceae* family, and an even larger number on hybridization between different *Brassica* species. Most of these hybrids do not produce mature seed; however normally incompatible interspecific hybridization can spontaneously produce a few seeds which usually yield true F1 plants as a result of unexpected ploidy changes (Nashiyama et al., 1991) This phenomenon has been documented between *B. napus* and *B. nigra*. The objectives of the present study were to determine the feasibility and frequency of gene flow between oilseed rape and a related weed species (*B. nigra*) by means of ISSR (Inter Simple Sequence Repeat) markers.

## Materials and methods

Two oilseed rape cultivars (Ceres and Talent) were selected from a collection kept at the Department of Agroenvironmental Science and Technology, University of Bologna, Italy. Oilseed rape and *B. nigra* were intercrossed and used in gene flow studies. *B. napus* and *B. nigra* plants were grown in the following conditions: 1) growth chamber with insect-mediated pollination (*Osmia cornuta*); 2) growth chamber with free pollination; 3) greenhouse with free pollination; 4) greenhouse with pollination under tissue/no-tissue cages. Total DNA was extracted from leaf tissue samples as described by Shagai-Maroo et al. (1984), spectrophotometrically quantified and used for molecular analysis. Thirty ISSR primers were screened for polymorphic loci and for accession-exclusive fragments in all five *Brassica* genotypes.

## Results and discussion

To obtain an estimate of the level of detectable polymorphism, 30 ISSR primers were screened on a subset of bulked DNA samples made up of a mixture of 13-20 individuals per each accession. Among the tested primers seven produced 89 reproducible and polymorphic fragments. The total number of bands (TNB), number of polymorphic bands (NPB) and percentage of polymorphic bands (P%), obtained per each primer are shown in Table 1.

Four ISSR bands (Figure 1) were uniquely detected in oilseed rape genotypes and were thus chosen for gene flow evaluation between *B. napus* and *B. nigra*. F1 individuals highlighted an average percentage of hybrids carrying at least one of the selected *B. napus* markers equal to  $1.6 \pm 0.2\%$ . No significant differences were found in relation to the growing environmental conditions (growth chamber =  $1.6 \pm 0.1\%$ ; greenhouse  $1.6 \pm 0.1\%$ ) and pollination systems (bees =  $1.4 \pm 0.3\%$ ; free =  $1.7 \pm 0.3\%$ ).



Even if exclusively performed in controlled environments, the results of this study suggested that hybridization can occur between *B. napus* and *B. nigra*. Obtained values were in general agreement with literature data about the potential hybridization between *B. napus* and other diploid *Brassica* species. It is still to assess whether the observed gene flow can actually occur in open field and the effective fitness of triploid hybrid individuals generated from the cross between *B. napus* and *B. nigra*.

Table 1: Total number of bands (TNB), number of polymorphic bands (NPB), percentage of polymorphic bands (%P) obtained per each ISSR primer.

Primer code	Nucleotide sequence (5'→3')	TNB	NPB	%P
LOL1	(CT) <sub>8</sub> AC	17	16	94
LOL2	(CT) <sub>8</sub> GC	16	15	93
LOL8	(GT) <sub>6</sub> CC	14	10	71
BR5	(AG) <sub>6</sub> GC	17	10	59
BR6	(CA) <sub>6</sub> GA	16	14	87
BR8	(AGC) <sub>3</sub> TC	17	11	65
PHV2	(GACA) <sub>4</sub>	18	13	72
Total		115	89	77

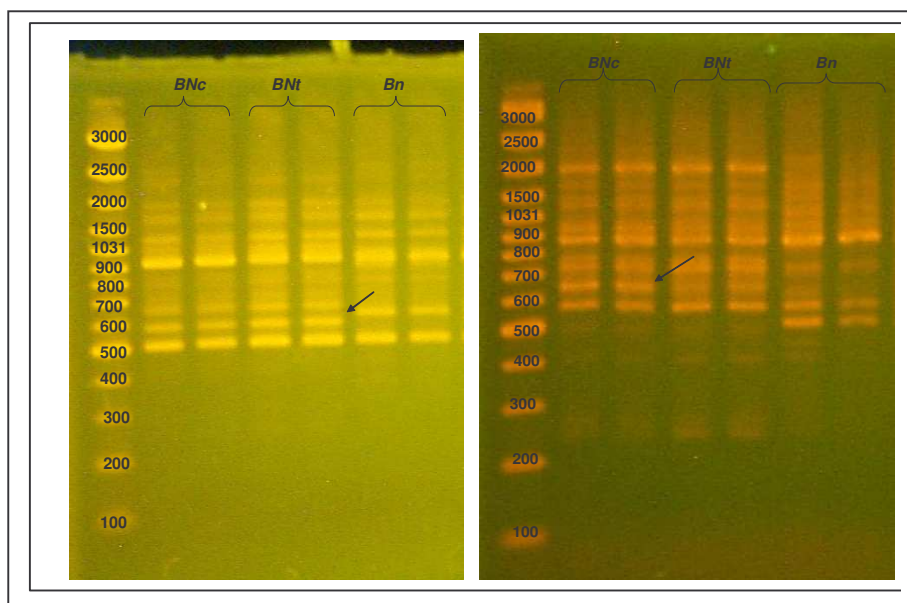


Figure 1: Arrows indicate ISSR fragments obtained with primer BR6 (on the left) and primer LOL8 (on the right) and used for gene flow evaluation between *B. napus* and *B. nigra*. Bnc: *B. napus* cv. Carinata; Bnt: *B. napus* cv. Talent; Bn: *B. nigra*

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# Weed community in soyabean monoculture: effect of soil fertility and content of phenolic compounds in the soil

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## Introduction

Monoculture system of plant cultivation determines microbiological soil properties and level of phenol compounds (Barabasz & Smyk 1997; Barabasz et al., 1998, Wójcik-Wójtowski et al., 1990). The system also shifts weed species composition, leading to develop high domination of one or two species (Jedruszczak et al., 2000 and 2004). The question arises if the status of the soil under monoculture is related also with the weed community shift.

## Material and methods

Weed community was assessed in 2001, in the eighth year of long term of field experiment including four types of the soybean monocultures: typical (M), with soyabean straw (MS), white mustard biomass (MM) and rye biomass (MR) buried into the soil (rich lessive loess) by ploughing through seven years. Both mustard and rye were sown after soybean harvest in August/September and their biomass was incorporated into the soil by ploughing before winter. In the same year (2001) soil fertility according to Myśków's index (bacteria to fungi ratio) and phenolic compounds content were assessed in the soil. It was done three times during soybean vegetation (Myśków 1987; Swain & Hillis 1959). The traits of the weed community were measured prior to soyabean harvest. The results were elaborated statistically by means of ANOVA; difference amongst means were assessed by Tukey's test.

## Results and discussion

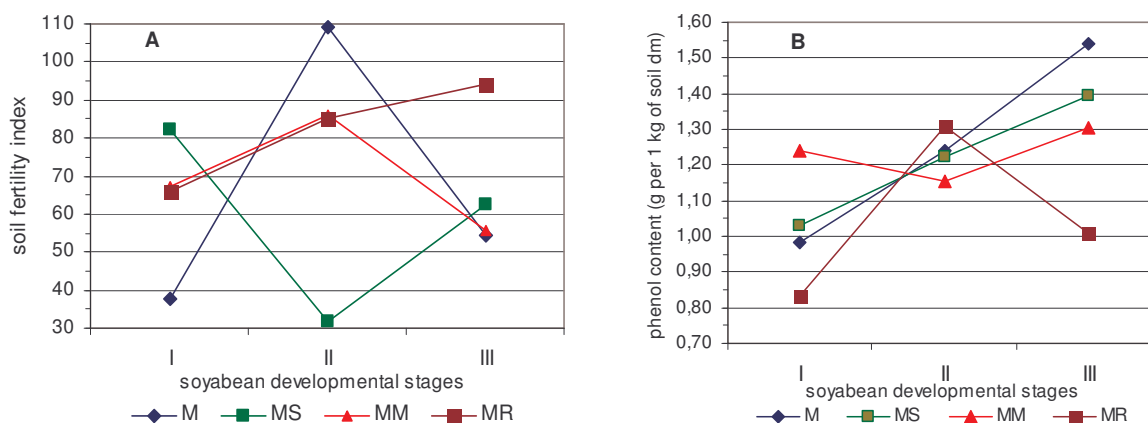


Fig. 1. Myśków's soil fertility index (A) and content of phenols ( $\text{g kg}^{-1}$  of soil dry matter) (B) in soyabean developmental stages: I-vegetative, II-flowering/pod formation, III-full seed ripeness under different monoculture types. LSD ( $P = 0.05$ ): (A): 32.9; (B): 0.14.

The monoculture types essentially influenced the soil fertility index value in soyabean stages (Fig. 1A): in the progress of soyabean development, substantial differences occurred under M and MS while they were not observed in MM and MR monoculture. The index was essentially lower under M in comparison to other monocultures in the I stage while higher than in MS only in the II stage. Content of phenol compounds essentially increased with progress of soyabean developmental stages, excluding MR, where decreased after II stage (Fig. 1B).

Table 2. The traits of weed communities growing soyabean canopy in different monocultures (SE in brackets)

Traits	Type of monoculture			
	M	MS	MM	MR
Total weed density (plants m <sup>-2</sup> )	32.3 (8.91)	33.9 (5.63)	31.4 (2.93)	32.0 (3.97)
Total above-ground dry weight (g m <sup>-2</sup> )	121.2 (33.27)	116.1 (23.20)	84.4 (23.32)	105.8 (11.33)
Number of species (as total)	13 (0.38)	11 (0.81)	8 (0.31)	10 (0.80)
Mean dry weight of one individual (g)	3.8 (0.65)	3.4 (0.38)	2.7 (0.63)	3.3 (0.69)
% of main species in weed density:				
<i>Echinochloa crus-galli</i> L.P. Beauv.	31.0 (10.58)	42.2 (9.61)	44.6 (7.11)	33.4 (9.14)
<i>Galinsoga</i> spp.	44.6 (11.64)	39.2 (7.57)	38.2 (8.32)	28.1 (6.28)
Sum	75.6 (2.34)	81.4 (8.22)	82.8 (5.93)	61.5 (5.64)
% contribution of perennial spp. in weed density	12.4 (2.92)	10.9 (2.82)	8.6 (5.21)	20.9 (4.33)

The weed density and their dry weight m<sup>-2</sup> (Tab.1) were not statistically different amongst monocultures. However, lowering trend of 20-30% of dry weight under MM in comparison with other monoculture types was noted. There were also some differences in the other traits amongst the communities. Soyabean + w. mustard (MM) was distinguishable by lowest weedy parameters, such as total air dry weight, individual weight, perennial individual contribution and species number, in relation to M, MS, and MR. It could be due to specific limiting influence of white mustard, probably along with the highest phenol status in the soyabean vegetative stage (I), on emergence and further growth of weeds (Duer 1996). The dominant species were *E. crus-galli*, *G. parviflora*, and *G. ciliata*. It should be underlined that the main species provided from over 61 % to almost 83 % of the all community individuals. They are thermophilous and nitrophilous species very common in soyabean crops in Poland (Jędruszczak et al. 2004 and 2000).

Summarising, it should be pointed that the soil, in the eighth year of soyabean monoculture enriched with organic matter from different plant residues (i.e. soyabean straw, green white mustard, green rye) differs in fertility and level of phenol compounds. It seems that the soil fertility index could not be responsible for differences found amongst the weed communities while white mustard residues combined with the highest phenols content through soyabean vegetative stage (I) could contributed to that under the MM monoculture type.

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# Driving forces for the development of *Cirsium arvense* in arable farming systems

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## Introduction

*Cirsium arvense* (L.) Scop. is a dioecious, self-sterile, insect-pollinated species, which furthermore possesses a highly effective system for root propagation and persistence. Thus, the species is characterised by efficient clonal propagation together with sexual reproduction. Vegetative spread and reproduction via seeds is possible and both seeds and root fragments can establish populations in arable farming situations.

The importance of the weed species in arable farming systems increases whenever agricultural inputs like fertilisers and pesticides are reduced, e. g. under the conditions of Organic farming. Besides the obvious difference in the use of herbicides, Organic farming systems differ in more instruments: generally the soil is more intensively cultivated and crops are less competitive, since they are less fertilised with mineral nitrogen.

In this paper we address to possible driving forces for increasing infestations of arable fields with *Cirsium arvense*. We therefore combine results from different experimental approaches with respect to the visible incidents patches, spread of root fragments and seeds and conclude on the influence of soil cultivation and the competitiveness of the crops as driving forces. The experimental work was done at the Research Center Agriculture and the Environment and the Institute of Plant Pathology at the University of Goettingen.

## Patch development

In an annually ploughed long term experiment (1988-99) with the crop rotation sugar beets - winter wheat - winter barley, the cereal crops were not N-fertilised in one treatment (Dau & Gerowitt, 2002). While occasional thistle sprouts appeared in the N-fertilised plots, thistle patches developed continuously in those plots where mineral N-fertilisation was skipped in the cereal crops. Thistle patches developed in six years in the not fertilised cereal crops, while this development was completely suppressed in the fertilised plots - without any use of herbicides. Intensive soil cultivation could not prevent the build-up of severe thistle patches.

Starting in 2000, the plots with the dense thistle patches were N-fertilised under the cereal crops (135 kg/ha N incl. Nmin) and a grass-clover mixture replaced the beet crop. Thistle patches were reduced from 353 sprouts/28m<sup>2</sup> in 2000 to 18 sprouts/28m<sup>2</sup> in 2003 (Dau & Gerowitt 2004). However, a crucial period were the intercropping periods in autumn. Sprouts cutted by the combine harvester recovered very rapidly and formed vigourous plants investing all ressources into their root system.

## Vegetative spread

Soil cultivation is recommended to restrict the population establishment. Under favourable conditions even small root fragments of *C. arvense* establish new sprouts – therefore soil cultivation can also support vegetative spread of root fragments. In order to estimate vegetative spread, thistle sprouts were mapped and collected on eight fields of a farming systems experiment (Hettwer & Steinmann 2002). While four fields were annually ploughed during the last 10 years, the other four were only shallow cultivated during this period. The genetic diversity was investigated based on 30 sprouts randomly taken per field. REP-PCR revealed high values for genetic diversity. Diversity indices and genetic distances per field indicate, that the field populations could not be established via vegetative spread. No evident difference appeared for the two soil cultivation regimes.

Ploughing can move root fragments over short distances within the direction of tillage. Genotyping thistle sprouts makes it also possible to check this. Within a field which was annually ploughed, five patches were mapped – one central patch and four surrounding patches. Two of them lay in the direction of tillage to the central patch while the other two lay beside (Hettwer & Gerowitt 2004). The genotypes of the plants were examined by using repetitive enterogenic primer (REP) and inter simple sequence repeats (ISSR) analysis. Differences in genotypes between neighbouring patches indicated that root fragment dispersal via soil cultivation was of minor importance.

### **Establishment of seedlings**

Data on genetic diversity indicate that *Cirsium arvense* is entering arable fields via seedlings more frequently than expected (Hettwer & Gerowitt 2004). *Cirsium arvense* requires light and high temperatures for germination. The influence of different light intensities on germination and development of *Cirsium arvense* was studied in a model experiment in two years. In 2002, shading was created with winter wheat at different seeding rates resulting in 291 resp. 211 ears/m<sup>2</sup>. In 2003, shading was created with different shading cloths and was adjusted to the changing light intensities in differently farmed winter wheat fields. In both years, thistle seeds were sown directly into the plots (Dau et al. 2004).

In 2002, seedlings numbers in the shaded treatments were lower in the unshaded control. In the second experimental year, seedlings number were highest in plots with rather low light intensities. After the removal of wheat or shading cloths in August, a second peak of germination in the formerly shaded plots led to approximately equal seedling numbers in all treatments. Very low numbers of root sprouts were produced in the shaded treatments. In these treatments, biomass of sprouts and roots were also considerably reduced in both years. Seedlings of *Cirsium arvense* investigated in an oat crop in 2002 developed differently depending on the crop stand: In an area with high crop density, all seedlings died until early July. Although about 50% of the seedlings survived in a thin crop, none of the plants were able to develop root sprouts.

### **Conclusions**

Thistle frequently enter an arable environment by seedlings. However, these seedlings are poor competitors, it can happen during the whole period from spring to autumn. Seedling establishment appears rather “secret”, farmers as well as researchers have difficulties to observe it. If the young thistle plant receives enough light, as in non-fertilised cereal crops or on cereal stubbles, they will be able to establish their effective root system. Therefore the periodic sequence of rather open conditions seems to be very important for the development of thistles in arable farming systems. There can be more of these favourable periods for thistle in Organic farming: thin cereals, other crops with a late canopy closure and early harvest dates of cereals.

The risk of spreading root fragment by intensive soil cultivation seems to be rather low. However, intensive soil cultivation can not prevent the long-term development of patches when the crops are poor competitors. Hence, the most important driving force for thistle development seems to be the “cover management” of arable fields.

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# Effect of field bindweed (*Convolvulus arvensis*) and pinto beans (*Phaseolus vulgaris*) densities on growth and yield of pinto beans in greenhouse

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## Introduction

Field bindweed is a serious perennial weed in most provinces of Iran including Fars province. It is most troublesome in cereals, legumes, and potato crops, causing up to 50% yield reduction (Farahbakhsh, 1983).

Weeds, especially those competing in the first 4 to 5 weeks of the growing season, are known to cause severe reductions in bean yield (Fennimore et al., 1984). Research indicated that up to 9 weeks of weed free maintenance after crop emergence is required to prevent bean yield losses (Blackshaw, 1991). Replacement series experiment is a method of studying crop-weed competition (Radosevich, 1997). It includes pure stands as well as mixtures in which the proportion of the two species studied is varied. The total plant density is a constant over all treatments in such experiments. Competition experiments using replacement series showed that bean germinated earlier than black nightshade (*Solanum nigrum*) and barnyardgrass (*Echinochloa crusgalli*) and caused significant reductions in weed height, leaf area, and dry weight (Fennimore et al., 1984). The impact of bean upon itself was always greater than the effect of either weed species.

This greenhouse study was conducted to evaluate the effects of different densities of field bindweed and pinto beans on growth and yield of pinto beans.

## Materials and methods

The experiment was performed in a completely randomized design with four replications using replacement series in which field bindweed and pinto beans were planted in different ratios of 4:0, 3:1, 2:2, 1:3, and 0:4 plants per pot. Field bindweed and pinto bean seeds were planted 2 and 5 cm deep, respectively, in 30 cm pots.

Plants were harvested from the soil surface 60 days after seeding. Plant materials were kept in oven at 75°C for 48hrs. Measurements included plant height, leaf area, shoot and root dry matter, number of bean pods and bean yield. Relative crowding coefficient (RCC), relative yield (RY), and relative yield total (RYT) were calculated using the following formula:

$$RCC = \frac{YA_{mix}}{YB_{mix}} \bigg/ \frac{YA_{mon}}{YB_{mon}}$$

$$RY = \frac{Y_{mix}}{Y_{mono}}$$

$$RYT = \sum_{i=1}^n RY$$

where  $YA_{mix}$  and  $YB_{mix}$  are average yield per plant of A and B grown in mixture, respectively.  $YA_{mon}$  and  $YB_{mon}$  are average yield per plant of B grown in monoculture, respectively.  $Y_{mix}$  and  $Y_{mono}$  are yields in mixture and monoculture, respectively. Means were compared using Duncans multiple range test (P=0.05).

## Results and discussion

Relative yield total (RYT) was more than 1 in all mixture ratios (Table 1). This showed that pinto beans and field bindweed were exploiting the resources in different ways or somehow benefiting each other. It is possible that the root systems of these two species penetrate different soil depths.

Relative crowding coefficient (RCC) of pinto beans to field bindweed in 3:1 (3 pinto beans and one field bindweed) was greater than in other ratios. RCC of field bindweed to pinto beans in the same ratio (3:1) was lowest among all ratios. This showed that pinto bean plants were engaged in intraspecific competition, while field bindweed plants were under the influence of interspecific competition.

Pinto beans plant height and leaf area increased and its shoot and root dry matter, number of pods and yield decreased as the number of field bindweed plants per pot increased. There were highly significant negative correlations between the weed dry matter and pinto beans pod number ( $r=-0.926$ ) and shoot dry matter ( $r=-0.96$ ). RCC of pinto beans and field bindweed showed that pinto beans in this experiment was at least 3 times more aggressive than field bindweed (Table 1).

Table 1. RYT and RCC of pinto beans and field bindweed in different ratio mixtures

Ratio		Competition indices		
Pinto beans	Field bindweed	RYT	RCC*	RCC**
1	3	3.00 a	2.06 b	0.52 a***
2	2	2.15 b	2.02 b	0.53 a
3	1	1.40 c	3.55 a	0.29 b
Mean		2.18	2.54	0.45

\*RCC of pinto beans to field bindweed

\*\*RCC of field bindweed to pinto beans

\*\*\*Means in each column followed by the same letter are not significantly different (Duncan 0.05).

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## Evaluations of multi-species weed competition in wheat

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### Introduction

Weed competition for moisture, nutrients and light, reduces wheat yield. Weed density is one of the most effective factors in competition. Most studies consider interference between single weed species and crop, while there are several competing species in the field; therefore, results from most of the studies on weed-crop competition can not directly applied in weed management programs. Additionally, many interference studies are conducted under conditions favoring weed establishment and cropping practices that would reduce growth and competitiveness of weeds are generally not used (Swinton et al., 1994). The specific objective of this study was to evaluate effect of multi-species weed competition on wheat yield in field conditions.

### Materials and methods

The experiment was conducted in 2002 at Agricultural Research Station of Ferdowsi University of Mashhad (Lat 36° 18' N, Long 59° 36' E), Iran. A 15 m x 50 m area of a 15 ha wheat field was selected as the experimental site. This area was managed like all other parts of the field, except for the use of the herbicides. At the beginning of shooting stage, 30 points were sampled systematically using a 50 cm x 50 cm quadrat. In these quadrats, number of wheat plants and tillers and number of weeds (each species separately) were counted without any damage to the plants. To prevent wheat seed shattering, the plants were harvested 7 to 10 days before all spikelets and plants being completely dried. All weeds in the quadrat were counted separately, harvested, dried at 80 °C for 48 hours, and their dry weight measured. Besides, all the wheat plants in the same quadrat were harvested and number of plants, seed weight and total biomass were measured.

Inter- and intra- specific competition coefficients were estimated by fitting regression model to reciprocal wheat biomass ( $W_c$ ) as dependent variable and wheat ( $N_c$ ) and weed densities ( $N_w$ ) as independent variables (Rejamanek et al., 1989, Eq. 1).

$$1/W_c = b_{co} + b_{cc}N_c + b_{cw1} N_{w1} + \dots + b_{cw8} N_{w8} \quad (1)$$

where,  $b_{co}$  is the intercept of regression equation,  $b_{cc}$  is the intra-specific competition coefficient of wheat,  $b_{cwi}$  is the inter-specific competition of wheat and  $i$ th weed species with density of  $N_{wi}$ . Equation 1 was first used for wheat biomass against 8 weed species. Then the same equation was used with weed biomass as dependent variable and wheat and the other weeds densities as independent variables, this procedure was conducted for all weeds separately. In this case Eq. 1 will be changed as follow:

$$1/W_w = b_{wo} + b_{ww}N_w + b_{ww1} N_{w1} + \dots + b_{ww8} N_{w8} \quad (2)$$

where,  $b_{wo}$  is the intercept of regression equation,  $b_{ww}$  is the intra-specific competition coefficient of any selected weed species,  $b_{wwi}$  is the inter-specific competition of the selected weed and  $i$ th weed species with density of  $N_{wi}$ . In Eq. 2, wheat considered as a plant species, which presents in the field.

## Results and discussion

The weeds in the field were: *Avena ludoviciana* (AVELU), *Chenopodium album* (CHEAL), *Solanum nigrum* (SOLNI), *Stellaria holostea* (STEHO), *Fumaria* spp. (FUMSP), *Polygonum aviculare* (POLAV), *Sonchus* spp. (SONSP) and *Convolvulus* spp. (CONSP). 1/W regression model (Table 1) indicated that only *Avena ludoviciana*, *Convolvulus* spp. and *Chenopodium album* had a significant reduction effect on wheat yield. Cudney et al. (1989) reported that *Avena fatua* reduced wheat yield as high as 66%. According to Carlson and Hill (1985), 5-6 plants of wild oat per m<sup>2</sup> caused 20% yield loss in wheat. However, *Solanum nigrum* had positive effect on wheat yield as shown on its inter-specific competition coefficient with wheat in Table 1. To explain this effect, inter-specific competition coefficients of *Solanum nigrum* with other species were estimated by Equation 2. Results showed a significant negative effect of *Solanum nigrum* on *Stellaria holostea* and *Polygonum aviculare* (Bold underlined coefficients in Table 2). It seems that positive effect of *Solanum nigrum* on wheat yield is probably due to its depressing effect on other existing weeds e.g. *Stellaria holostea* and probably has led a reduction in competition ability of these weeds against wheat (Table 2).

Table1. Intra- ( $b_{cc}$ ) and inter- ( $b_{cwi}$ ) species competition coefficients estimated from Eq. 1

1/W	Plant density								
	Wheat	AVELU	STEHO	SOLNI	CHEAL	POLAV	FUMSP	CONSP	SONSP
Wheat	<b>0.0063</b> **	0.0088**	- 0.008 <sup>ns</sup>	<b>- 0.003</b> **	0.0017**	- 0.001 <sup>ns</sup>	- 0.0025 <sup>ns</sup>	0.008**	- 0.001 <sup>ns</sup>

Table2. Intra- ( $b_{ww}$ ) and inter- ( $b_{wwi}$ ) species competition coefficients estimated from Eq. 2

1/W	Plant density								
	Wheat	AVELU	STEHO	SOLNI	CHEAL	POLAV	FUMSP	CONSP	SONSP
AVELU	0.0026**	<b>0.0002</b> <sup>ns</sup>	- 0.0007 <sup>ns</sup>	0.0000 <sup>ns</sup>	- 0.0006 <sup>ns</sup>	0.001 <sup>ns</sup>	- 0.017**	- 0.0007 <sup>ns</sup>	- 0.003 <sup>ns</sup>
STEHO	0.022 <sup>ns</sup>	0.056 <sup>ns</sup>	<b>- 0.030</b> <sup>ns</sup>	<b>0.013</b> **	- 0.030**	- 0.041 <sup>ns</sup>	- 0.133 <sup>ns</sup>	- 0.082 <sup>ns</sup>	- 0.054 <sup>ns</sup>
SOLNI	0.117 <sup>ns</sup>	- 2.87 <sup>ns</sup>	0.344 <sup>ns</sup>	<b>0.147</b> *	- 0.352 <sup>ns</sup>	1.083 <sup>ns</sup>	- 1.534 <sup>ns</sup>	- 1.043 <sup>ns</sup>	- 0.100 <sup>ns</sup>
CHEAL	0.077 <sup>ns</sup>	0.602*	- 0.017 <sup>ns</sup>	- 0.019 <sup>ns</sup>	<b>0.167</b> **	0.078 <sup>ns</sup>	- 0.632 <sup>ns</sup>	- 0.396 <sup>ns</sup>	0.167 <sup>ns</sup>
POLAV	0.042**	0.048 <sup>ns</sup>	0.009 <sup>ns</sup>	<b>0.011</b> **	- 0.041 <sup>ns</sup>	<b>- 0.030</b> <sup>ns</sup>	- 0.277*	- 0.042 <sup>ns</sup>	0.129**
FUMSP	- 0.002 <sup>ns</sup>	0.042 <sup>ns</sup>	0.174**	0.007 <sup>ns</sup>	- 0.019 <sup>ns</sup>	- 0.018 <sup>ns</sup>	<b>0.329</b> *	0.098 <sup>ns</sup>	0.107*
CONSP	- 0.010 <sup>ns</sup>	- 0.167*	0.083**	- 0.002 <sup>ns</sup>	0.016 <sup>ns</sup>	0.022 <sup>ns</sup>	- 0.19 <sup>ns</sup>	<b>- 0.083</b> <sup>ns</sup>	- 0.059 <sup>ns</sup>
SONSP	0.014 <sup>ns</sup>	- 0.103*	0.235**	- 0.004 <sup>ns</sup>	0.029 <sup>ns</sup>	- 0.014 <sup>ns</sup>	0.015 <sup>ns</sup>	- 0.054 <sup>ns</sup>	<b>- 0.009</b> <sup>ns</sup>

\*\* Significant at P = 0.01; \* Significant at P = 0.05; ns non-significant

Highlighted coefficients are intra-specific competition coefficients

It was concluded that inter-specific competition between weed species may indirectly affect on crop yield enhancement and could be of great importance in multi-species weed studies.

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# Competition of *Lolium rigidum* and *Anacyclus clavatus* in barley

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## Introduction

*Lolium rigidum* Gaud. and *Anacyclus clavatus* (Desf.) Pers. commonly infest the cereal crops of the semi-arid areas of Toledo, Spain. These weeds have different emergence times: *L. rigidum* emerges from the sowing of the cereal until the end of winter while *A. clavatus* emerges during the winter and part of the following spring. Numerous reports exist (especially from Australia) of the damage that *L. rigidum* causes to cereal crops. Indeed, reductions in wheat yield can be predicted from the infestation density (Reeves, 1976). No report exists, however, of *A. clavatus* having caused such damage (Dorado et al., 1997). The aim of the present work was determine whether the competition offered by these weeds when growing separately among barley is different.

## Materials and methods

In a experimental plot receiving the standard fertilization in pre-sowing and spring time was sown in autumn with 150 kg ha<sup>-1</sup> of barley (*Hordeum vulgare* L.) cv. Reinette. When the barley was at the 3-tiller stage (in January), two 30 x 50 m subplots were established. These had the same soil and were infested almost exclusively by the weeds *L. rigidum* (LOLRI) and *A. clavatus* (ANACL), although in different proportions. At this time LOLRI was at the 2-tiller stage and ANACL at the seedling stage.

In the LOLRI subplot, ANACL plants and other isolated dicotyledonous species were removed by applying bromoxynil + mecoprop + ioxynil (0.225 + 1.125 + 0.225 L a.i. ha<sup>-1</sup> respectively, Oxytril M, Bayer) over the barley tillering stage. In the ANACL subplot, LOLRI plants were controlled by applying diclofop-methyl (0.630 L a.i. ha<sup>-1</sup>, Illoxan, AgrEvo) at the same barley growth stage.

When the barley grain was ripe, 36 samples of the vegetation in each subplot were taken using 0.50 x 0.50 m randomly-placed quadrats. The material obtained was dried in an oven at 60°C for 48 h in order to determine the dry biomass of the weeds and crop separately. The dry biomasses of the barley and LOLRI plants were obtained from the sum of their straw plus grain, while that of the ANACL plants was determined from the sum of the vegetative and reproductive parts (flowers plus seeds) of the senescent plant.

STATISTICA 6.0 software was used to produce and compare the curves relating the biomass of the weeds to barley attributes, using a simple and fitted model.

## Results and discussion

The biomasses were more strongly correlated to barley straw and grain yield, and to the number of barley ears, than was the number of weed plants per area.

Figures 1 to 3 show the response curves for the above barley attributes with respect to the increase in dry biomass of each weed species. Grain and straw yield and the number of barley spikes per square meter were significantly ( $P \leq 0.05$ ) more affected by the presence of LOLRI than ANACL. Other attributes of barley were all reduced as a consequence of the increase in weed biomass being irrelevant the type of infesting weed.

The greater ability of LOLRI plants to compete with the barley was owed to their earlier emergence: they therefore have more time to compete than ANACL plants. Further, since LOLRI belongs to the family Gramineae it probably requires resources similar to those needed by barley, for which both must compete. Since ANACL belongs to the family Compositae, the competition with the crop plants is probably less intense.

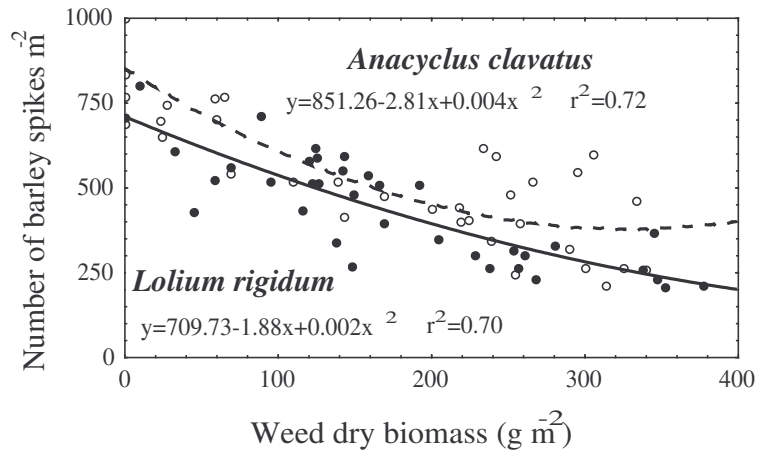


Figure 1. Relationship between weed dry biomass and number of spikes of barley

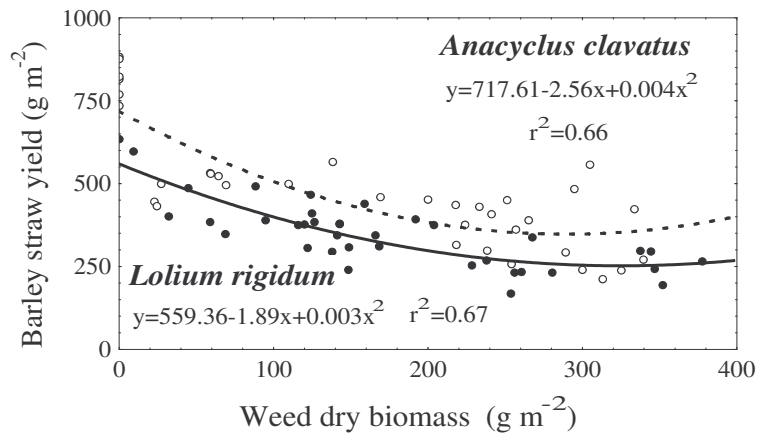


Figure 2. Relationship between weed dry biomass and barley straw yield

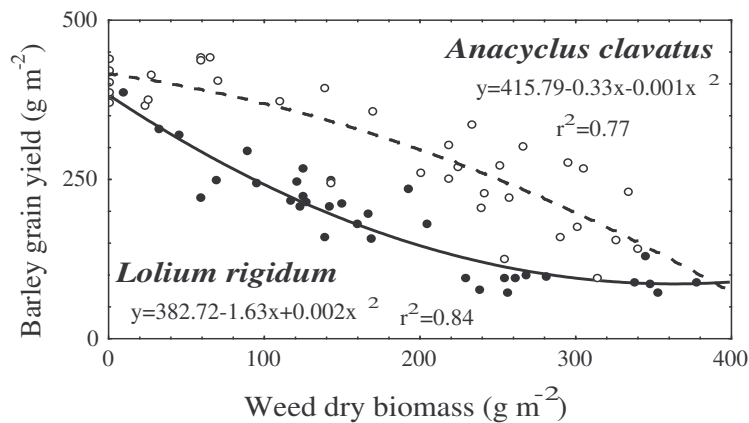


Figure 3. Relationship between weed dry biomass and barley grain yield

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# Competition of barnyardgrass with three spring-sown row crops

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## Introduction

During the last decades *Echinochloa crus-galli* L/PB has caused major damage including zero-yield-crops for spring-sown row crops, because of its strong colonizing potential (Barrett & Wilson, 1983). Therefore a long-term study about the interaction of this weed with relevant crops was initiated. At this moment its management can be improved, particularly if our eco-physiological understanding of the weed improves (Sagar, 1968; Norris, 1992; Labrada, 1995). A useful tool has been to define “the critical period of weed interference” that is the period during the crop cycle when weed species cause major injury to the crops (Labrada, 1995). New experiments about the ecology of weeds are important (Mortensen et al., 2000), as well as comparative studies, in which the competition of the weed with a number of different crops is compared. This paper shows a comparative study of weed-crop ecology for three major row crops: sunflower, maize and soyabean.

## Materials and methods

Sunflower – *Festiv* hybrid, maize – *Turda Super* hybrid and soyabean – *Atlas* variety were tilled on a podzol during 2001-2002. Individual plot size was 25 m<sup>2</sup> and each plot was replicated four times. The weed rapidly established from a natural population and the other weeds that emerged were controlled manually. The density of the weed was 140-160 plants m<sup>-2</sup>. The development of total biomass was determined through frequent harvesting: ten-day-intervals during the vegetative period, starting a week after emergence and five-day-intervals for the grain filling period, starting a week after flowering. At each harvest-day one square meter of each crop was harvested, cut and dried in the oven for 8 hours at 105° C. Total biomass and grain yield of the crop plants was measured both for weed-free and weed-infested conditions. Results of two years were averaged.

## Results and discussion

The data clearly represent the evolution of total biomass and grain yield of sunflower, maize and soyabean plants, both in monoculture and in competition with *E. crus-galli*. For sunflower and maize, total biomass in competition was approximately 50 % of that obtained under weed-free conditions, whereas for soyabean total biomass was reduced with about 70 % (Fig.1.A). The data highlight the specificity of the interaction between the crop plants and the weed, starting from the beginning (Wilson, 1988). Finally the grain filling process was disturbed. For sunflower and maize, seed production under competition was about 50 % of the weed-free control. For soyabean, grain yield under competition was reduced to 10 % of the control.

The grain filling periods of all crops, both with and without weed were also determined in great detail (Fig.1.B). Grain filling rate of sunflower was completely distorted, with a maximum of 4 g m<sup>-2</sup>d<sup>-1</sup> for the initial period, and very low rates towards the end. For maize the grain filling pattern was identical, but occurred at a much lower pace. Soyabean had a very long grain filling period, with a maximum of only 1 g m<sup>-2</sup>d<sup>-1</sup>.

These results indicate that competition from *E. crus-galli* affected the growth of the crops quite differently. All grain yields were seriously affected. The growth rate of dry matter into seeds was different and pointed out that grain filling of sunflower was abnormal (disturbed), of maize relatively normal and of soyabean nearly completely destroyed.

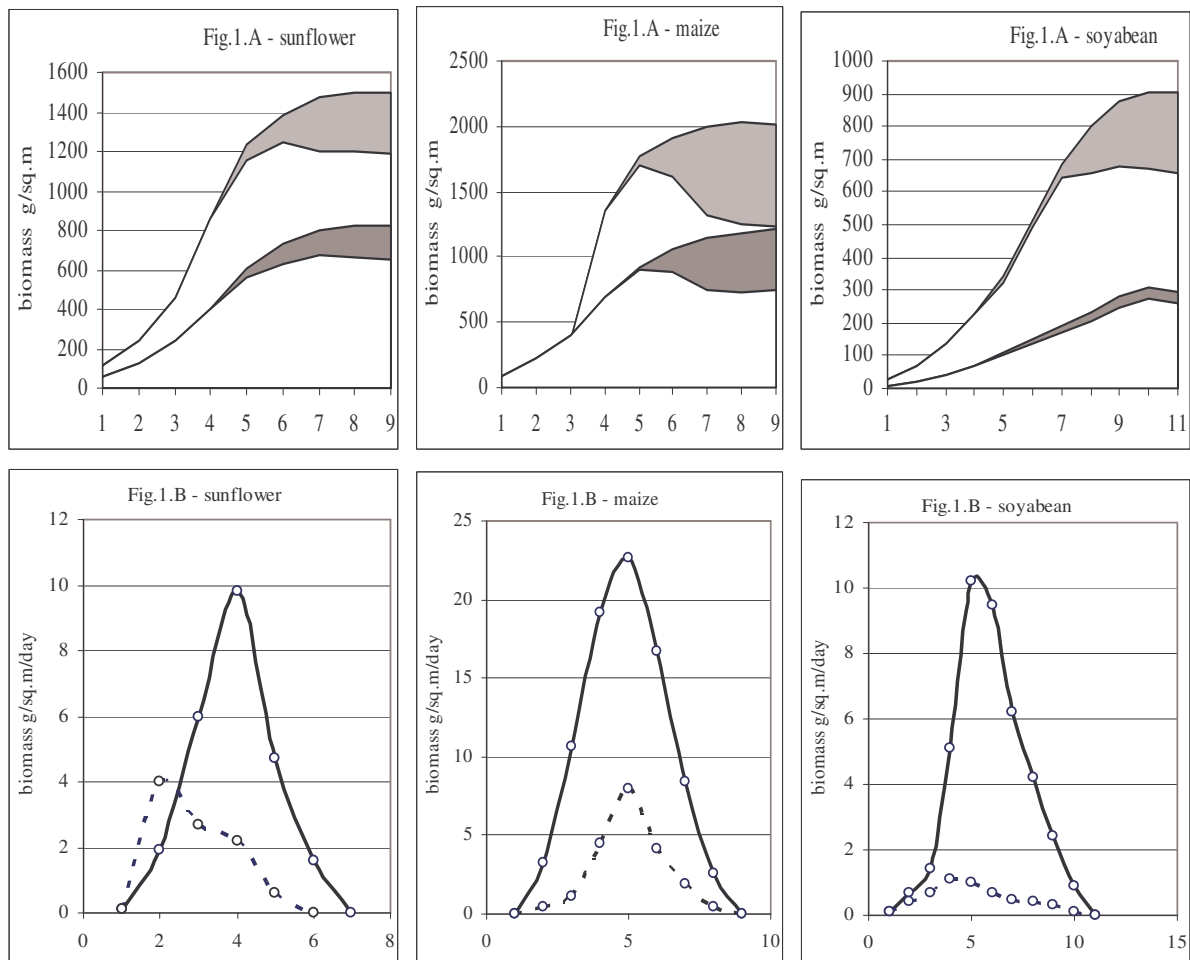


Figure 1. The evolution of biomass -total biomass and grain yield formation- (Fig.1.A) and grain filling rates (Fig.1.B) of three row crops: sunflower, maize and soyabean, as affected by the presence of *E. crus-galli*. The x-axis represents consecutive observations with time-intervals of 10 days for total biomass production and 5 days for grain filling rates.

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# **Investigation on the mechanisms of competition between barnyardgrass (*Echinochloa crus-galli*) and redroot pigweed (*Amaranthus retroflexus*) with dry bean (*Phaseolus vulgaris*)**

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## **Introduction**

Understanding the mechanisms of competition between species can increase our knowledge about the outcome of competition. This understanding might be useful for weed management and can be applied in crop breeding for improved weed suppression (Fontyn & Mahall, 1981). The success of weeds in farming systems depends on their growth strategy including: high growth rate, strong and efficient root system and good canopy structure (Maun, 1977). For example the species that have greater height, higher leaf area and more horizontal leaves are more successful in absorbing radiation than other species (Holt, 1995). This study was conducted to investigate the mechanisms of competition of barnyardgrass and red root pigweed with dry bean.

## **Materials and Methods**

An experiment was conducted in a randomized complete block design with 3 replications at Mashhad Experimental Station of Ferdowsi University in 2001. Treatments included three levels of redroot pigweed densities (4, 8, 12 plants m<sup>-2</sup>) and three levels of barnyardgrass densities (10, 20, 30 plants m<sup>-2</sup>) planted separately in bean (20 plants m<sup>-2</sup>). A weed free control crop was added to each block. Destructive sampling was done weekly and height, leaf area, dry matter and crop growth rate were measured for all species. Leaf area density in three canopy layers (0-25 cm, 25-50 cm and 50 cm up wards) was determined after canopy closure. To study competition for light, the INTERCOM model was used (Lantinga et al., 1999).

## **Results and Discussion**

Both weeds significantly ( $p < 0.01$ ) reduced total dry matter, growth rate, height and leaf area of bean. For all of the above parameters, redroot pigweed had a stronger effect on bean than barnyardgrass. Barnyardgrass growth index curves were lower than bean except at the end of the season (physiological ripening period of bean). Redroot pigweed growth index curves significantly exceeded that of bean and barnyardgrass except during the early growth period (data not shown). Redroot pigweed established 70 percent of its leaf area above the bean canopy throughout the season. Only at the end of the season, barnyardgrass established 40 percent of its leaf area above the bean canopy (Figure 1).

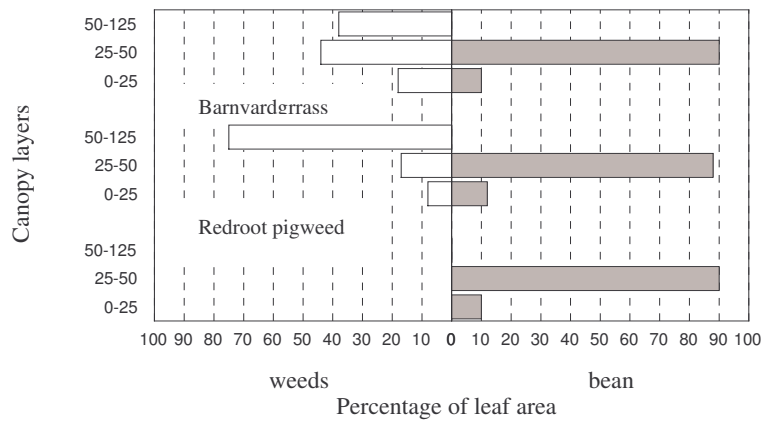


Figure 1. Distribution of leaf area in three canopy layers for each species after canopy closure

This indicates that the shoots of barnyardgrass had less effect on competition. Perhaps barnyardgrass imposed its competitive effects via partitioning of assimilates to the roots (Maun, 1977). For redroot pigweed it was shown that competition for light was one of the most important aspects in its competition with dry bean (Figure 2), which was due to its greater height, its better leaf area position (above the bean canopy) and its horizontal leaves (Holt, 1995). Results from the INTERCOM model also indicated that throughout the season redroot pigweed absorbed 60 percent of the radiation, whereas the share of dry bean was only 20 percent. Only at the end of the season barnyardgrass was able to absorb 45 percent of the total radiation.

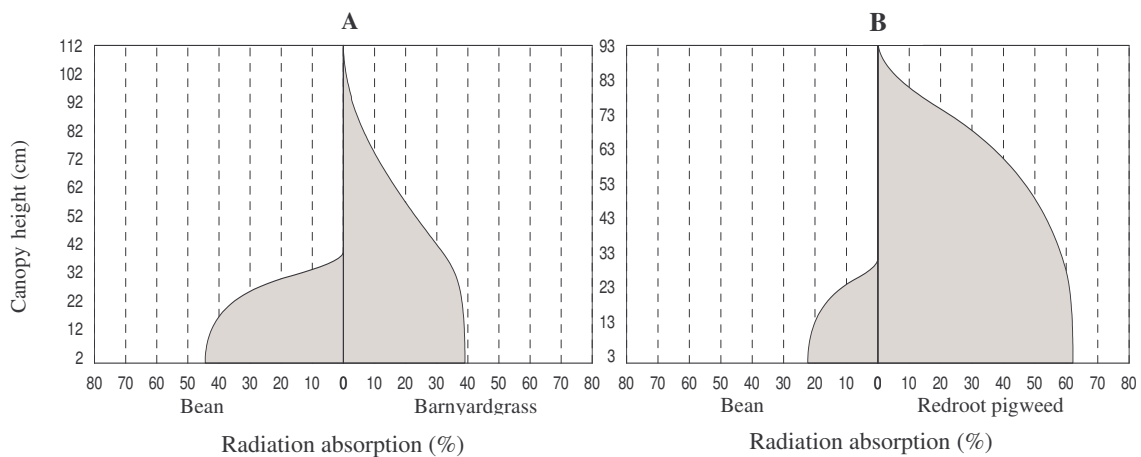


Figure 2. Cumulative radiation absorption in mixed canopies of bean and barnyardgrass (A), and bean and redroot pigweed (B), for each of the species after canopy closure

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# Stepwise rather than cyclic dormancy changes explain emergence of the summer annuals *Fumaria officinalis* and *Galeopsis speciosa*

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## Introduction

Seed dormancy is a seed characteristic that can prevent germination during conditions where a species would normally germinate (Vleeshouwers et al., 1995). Thereby, it can determine the timing of seedling emergence, and is a prevalent feature among annual weeds. Several species occurring as annual weeds are known to germinate readily when provided with favourable germination conditions after dormancy alleviation by stratification (e.g. Milberg & Andersson, 1997). Further, dormancy cycles are common; allowing extensive germination at a special time during a period with favourable circumstances after dormancy alleviation, and re-induction of dormancy if such a period fails to occur (e.g. Baskin & Baskin, 1985).

This study included two weedy species, *Fumaria officinalis* and *Galeopsis speciosa*. In Sweden, the emergence of *G. speciosa* in the field occurs during spring (Håkansson 1983), and the same has been observed for *F. officinalis*. Both species were studied with a focus on cold periods as a hypothetical dormancy alleviation factor.

## Material and methods

Seeds were collected in southern Sweden; *F. officinalis* in September 2001, 2002 (2 seed batches) and 2004 (3 seed batches) and *G. speciosa* in August 2001 and 2003 (2 seed batches each year). The experiments commenced after about one week at room temperature. At the time of writing, some experiments are still ongoing.

Seeds of all batches were sown on top of the soil in pots that were placed outside in slight shadow and kept moist; otherwise, these seeds were subjected to natural weather conditions. Pots were checked for seedlings weekly or biweekly, when snow free. Seeds of most batches were also subjected to various laboratory germination experiments. Of particular relevance were the experiments with the 2003 batches of *G. speciosa*, where seeds were subjected to alternating 0°C and 20/10°C (day/night) temperatures, where the 0°C period continued for either 8 or 16 weeks, and the 20/10 period for 8 weeks (Figure 1). Seeds were also subjected to continuous 0°C or 20/10°C. All seeds were provided light during 12 h per day.

## Results and discussion

Seeds of *G. speciosa* and *F. officinalis* subjected to a year of cold stratification and thereafter tested for germination at temperatures well covering spring temperatures occurring in Sweden indicated very modest dormancy alleviation. Seeds of both species sown outdoors produced

Table 1. Emergence of *Fumaria officinalis* during field experiments. Seeds were collected in southern Sweden in the autumn of 2001 (one seed batch) and 2002 (two batches) and were immediately sown on top of the soil in pots placed outside. Each year, emergence only occurred during a period of approximately two months in the spring.

Emergence year	Cumulative emergence (%)		
	2001 seeds (N=75)	2002 seeds (N=175)	2002 seeds (N=175)
2002	8	-	-
2003	17	1	0
2004	21	7	4

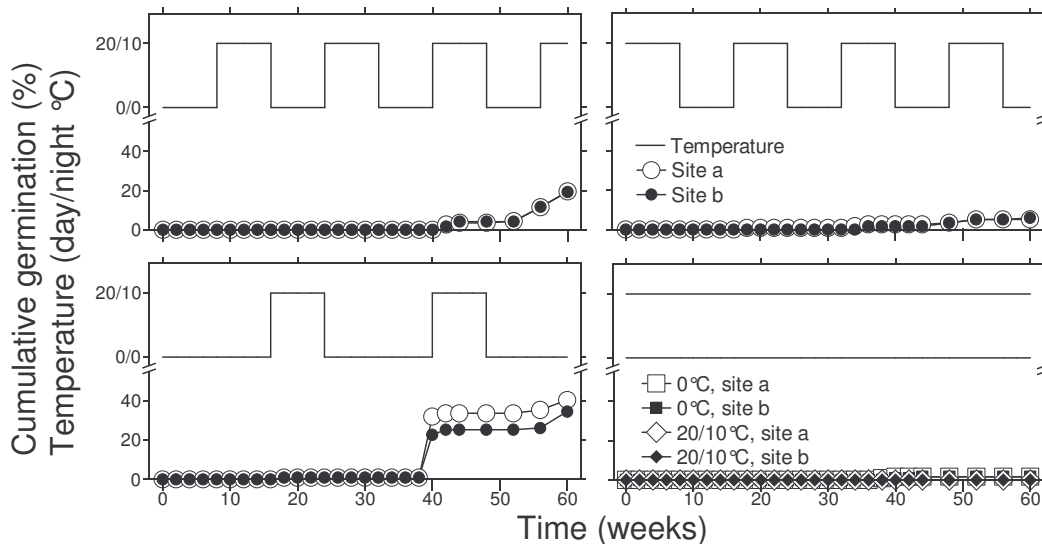


Figure 1. Cumulative germination of *Galeopsis speciosa* during laboratory experiment. Two seed batches were collected at different sites in southern Sweden on 10 August 2003 and subjected to different regimes of alternating or constant temperatures.

only a few seedlings each spring (Table 1). Furthermore, seeds of both species stratified outdoors and tested for germination at various temperature regimes, exhibited low germination. Germination occurred either in dishes outside during spring or, when tested late in winter, in the laboratory at a wide range of temperatures.

According to our germination results, *G. speciosa* and *F. officinalis* has some kind of physiological and morpho-physiological dormancy, respectively. This was expected because the two species have developed and underdeveloped embryo, respectively, and their dormancy were influenced by low temperatures. Nevertheless, none of the two species fitted well into the currently recognized template of dormancy classification (Baskin & Baskin 2004); even a full year in a refrigerator was not enough to completely break dormancy for any seed batch, whereas species that respond positively to cold stratification and are reported to have various sorts of dormancy classes do germinate nearly completely after less than four months of stratification.

We conclude that both species possess an innate inability to germinate to a high degree at any specific point in time. Instead, the cold period needed to completely break dormancy seems to be much longer than what occurs during one year in nature. This slow dormancy alleviation will allow part of a seed batch to germinate each spring and, as germination can occur over a wide range of temperatures (Figure 1), intermittent germination during cold stratification would prevent simultaneously germination. Warm periods did not re-induce dormancy; instead, cold periods alleviated dormancy with time, even when interrupted by warm periods (Figure 1). In the field, this leads to germination during several consecutive springs after dispersal. Therefore, the time of emergence is predictable, but the amount of germinated seeds from one cohort at any special time seems unpredictable but low. The emergence pattern is then a consequence of a deep dormancy being slightly alleviated each spring; not of an annual dormancy cycle, which could have been the expected for at least *G. speciosa* that is a typical summer annual with fully developed embryos.

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## Cucumber (*Cucumis sativus* L.) as test species in allelopathic research

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### Introduction

Cucumber (*Cucumis sativus* L.) as a sensitive test species is often used for detection of pesticide residues in the soil. It germinates rapidly and uniformly - within a few hours at a high percentage - and the length of its radicle can be easily measured. Therefore it is one of the best known acceptor (receiver) test species in allelopathic bioassay studies (Rice, 1984; Sterling et al., 1987; Narwal & Tauro, 1996; Kazinczi et al., 2001) although its allelopathic effect is also known (Wu et al., 1999). The aims of our investigations were to study the effect of water extracts of two allelopathic weed species on the germination and radicle length of cucumber.

### Materials and methods

*Cirsium arvense* and *Asclepias syriaca* shoots and roots were collected at Keszthely (Hungary) in July 2004. Plant parts were cut into small pieces and homogenized with a grinder. After grinding 100 ml distilled water was added to 25 g fresh homogenized plant parts and left for 24 hours (stock solution). Then the mixtures were filtered and diluted at two, five and ten times. So the mixtures were used at 25, 12.5, 5 and 2.5 % concentrations. Germination tests were carried out in Petri dishes in laboratory bioassays, at 22 °C in four replications, using 50-50 cucumber cv. 'Delicatessse' seeds in each Petri dish. Filter paper of the Petri dishes were wet with the plant extracts. Seeds germinated in distilled water served as a control (0% concentration). Germination was evaluated after three days and the radicle length of cucumber was also measured. Data were analysed by using log-logistic regression analysis (Streibig et al., 2003) and ED50 values (concentration of extracts causing 50% radicle length inhibition) were derived from dose-response curves and used for comparisons.

### Results and discussion

It has been seemed that the inhibitory effect of plant water extracts greatly depended on the donor species, the plant parts and the concentration of the extracts. The higher the concentration the stronger is the inhibitory effect due not only to its direct toxic effect, but to increased osmotic potential as well. In general, effects on seed germination were very slight and were visible only with the highest concentration rate (data not reported); thus, dose-response curves could not be estimated. In detail, *C. arvense* did not significantly affect germination of cucumber, but reduced radicle length, though the effect of root extracts was very slight and an ED50 value could not be estimated. Water extract of *C. arvense* shoots showed its ED50 at a concentration of 20.7 %, proving slightly more phytotoxic than water extracts of *A. syriaca* leaves that did not influenced significantly cucumber germination (not reported) but showed an ED50 value for radicle length at a concentration of 22.4 %. Water extract of *A. syriaca* stems at 25% concentration resulted in a significant 11% reduction in cucumber germination (not reported), while showed a stronger inhibitory effect on radicle length, with an ED50 value at a concentration of 18.3 %, which was significantly (see standard errors) lower than that of *C. arvense* (root and shoot) and of *A. syriaca* leaf. Root water extract of *A. syriaca* reduced significantly germination only at the highest concentration, while their phytotoxicity on radicle length of cucumber was very high, showing the lowest ED50 value (concentration of 5.8%). It has been concluded that water

extracts of *C. arvensis* and *A. syriaca* had more inhibitory effect on cucumber radicle length, than that on its germination (Table 1, Fig.1.).

Table 1. Parameters of dose response curves (Streibig et al., 2003) for radicle length (Tab. 1) *D* is radicle length of cucumber on the untreated control (higher asymptote; lower asymptote was not included in the model, to improve the estimate of ED50), *b* is the slope of the log-logistic response curve around the inflection point and ED50 is the concentration of extracts causing 50% radicle length reduction. Standard errors are in brackets.

Experimental treatment	D (mm)	b	ED50 (conc. %)
<i>C. arvensis</i> root	-	-	> 25
<i>C. arvensis</i> shoot	64.5 (4.65)	1.40 (0.534)	20.7 (4.35)
<i>A. syriaca</i> leaf	64.5 (1.57)	3.03 (0.557)	22.4 (1.17)
<i>A. syriaca</i> stem	61.3 (0.32)	4.85 (0.147)	18.3 (0.20)
<i>A. syriaca</i> root	41.9 (1.28)	2.88 (0.371)	5.8 (0.31)

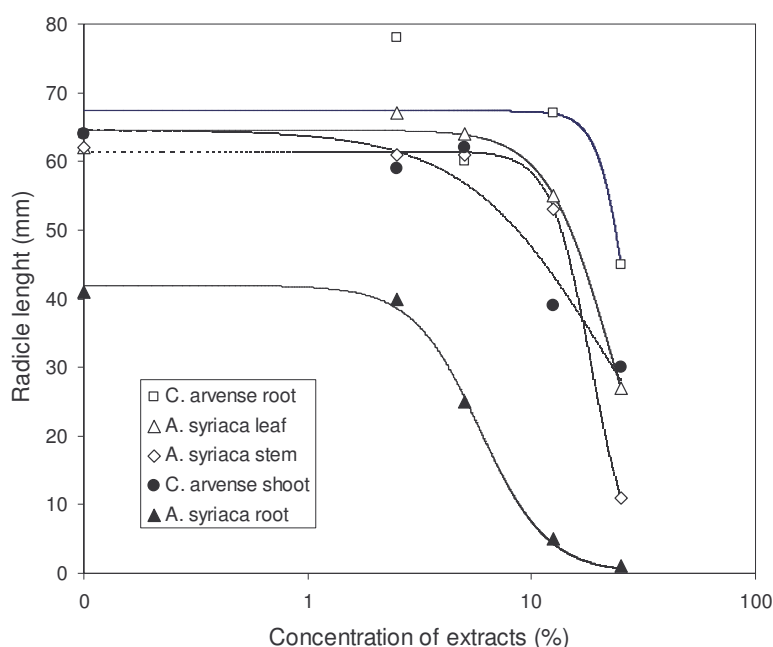


Fig.1. The effect of water extracts on the radicle length of cucumber

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# The role of weeds in the virus epidemiology

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## Introduction

Weeds can influence the quality and quantity of crops not only in direct way (e.g. by competing for nutrients and water), but also indirectly, as alternative hosts of various pests and pathogens. From a virological point of view, alternative weed hosts serve as food for the vectors of viruses, while the seeds and vegetative reproductive organs of certain weed species may also play important role in the epidemiology and allow the overwintering of economically important viruses. The positive side of the indirect effect is the so-called "bioherbicide strategy", when natural enemies of weeds (pests, pathogens) are used for suppression of a weed population. Although viruses are not used in the practice for biological weed control because of their poliphagous characteristics and great variability, biological decline of weeds due to virus infection may be considerable. Last years it has been proved that viruses in some weed hosts may reduce the growth, nutrient uptake, seed production, photosynthetic pigment content, drought-resistance and seed viability (Kazinczi et al., 1996, 1998, 2000, 2001, 2002a,b). Therefore they - in indirect way - may contribute to the reduction of a weed population. Recently a lot of weed-virus relations became known, which have a great virus epidemiological and ecological role (Kazinczi et al., 1999a,b; Kazinczi et al., 2002c; Magyar & Horváth, 2003). The aim of our present study was to detect new weed host-virus relations.

## Materials and methods

*Ambrosia artemisiifolia*, *Solanum nigrum*, *S. luteum*, *S. dulcamara*, *Chenopodium album*, *C. polyspermum*, *C. glaucum*, *C. ficifolium* and *Echinocystis lobata* plants were mechanically inoculated with 9 viruses (*Zucchini yellow mosaic virus*, ZYMV; *Cucumber mosaic virus*, CMV; *Potato virus Y*, PVY; *Sowbane mosaic virus*, SoMV; *Obuda pepper virus*, ObPV; *Tomato spotted wilt virus* TSWV; *Alfalfa mosaic virus*, AMV; *Melandrium yellow fleck virus*, MYFV; *Pepino mosaic virus*, PepMV) at 4-6 leaves stage in our virological vector free glasshouse. Reaction of plants to viruses was continuously evaluated on the basis of symptoms. Six weeks after inoculations back inoculation and DAS ELISA serological tests were also done.

Plant samples of some weed species (*Asclepias syriaca*, *Solidago gigantea*, *Mercurialis annua*, *C. album*, *A. artemisiifolia*, *Alisma plantago-aquatica*, *Datura stramonium*) showing virus symptoms and symptomless ones were collected from different parts of Hungary between 2002 and 2004. Virus infection of the samples were examined on the basis of symptoms, by biotest (Horváth, 1983), electronmicroscopical (Milne, 1984), DAS ELISA (Clark & Adams, 1977) and immunosorbent electronmicroscopical methods (Milne & Lesemann, 1984).

## Results and discussion

On the basis of inoculation some weeds as new artificial hosts of viruses became known (Table 1).

*A. artemisiifolia* showed high resistance to viruses, which can contribute to its rapid spreading in Hungary. PepMV was recently found and described in tomatoes grown under glasshouse in Hungary (Forray et al., 2004). It has a narrow host range, including the species belonging to

the botanical family of *Solanaceae* (Brunt et al., 1996). On the basis of our results *Solanum* weeds, occurring in tomato fields may be infection sources for tomato. *Chenopodium* species as local hosts of the most Tobamoviruses were known so far. *C. polyspermum* and *C. ficifolium* as local and systemic hosts of ObPV became known in our experiments for the first time. *C. glaucum* and *C. ficifolium* as latent host of SoMV became known. Symptoms were not seen, and serological tests also were negative, but back inoculation from the inoculated plants to the propagative hosts of SoMV (*C. quinoa*, *C. amaranticolor*) gave positive results. SoMV can be transmitted by pollen and seeds of *Chenopodium* species. ZYMV is one of the most important viruses of cucurbit crops in Hungary. *E. lobata* as host of ZYMV raises new epidemiological questions (Tóbiás et al., 1996).

Table 1. New artificial host-virus relations

Weed species	Viruses	Type of the host-weed relation
<i>Solanum nigrum</i>	PepMV	Local and systemic host
<i>S. luteum</i>	PepMV	Local and systemic host
<i>S. dulcamara</i>	PepMV	Local and systemic host
<i>Chenopodium polyspermum</i>	ObPV	Local and systemic host
<i>C. album</i>	SoMV, AMV	Local and systemic host
<i>C. glaucum</i>	SoMV	Latent host
<i>C. ficifolium</i>	SoMV ObPV	Latent host Local and systemic host
<i>Echinocystis lobata</i>	PVX ZYMV	Latent host Local and systemic host

On the basis of our surveys, economically important new natural weed host-virus relations are the followings: *A. syriaca*-Tobacco mosaic virus (TMV), CMV, AMV, TSWV; *S. gigantea*-CMV, Raspberry ringspot virus (RpRSV), Beet necrotic yellow vein virus (BNYVV); *M. annua*-SoMV; *C. album*-CMV, Potato virus S (PVS), Potato leafroll virus (PLRV); *A. artemisiifolia*-CMV; *A. plantago-aquatica*-SoMV, PVY; *D. stramonium*-Potato virus A (PVA), PVX, PVS, Potato virus M (PVM), CMV, TMV.

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# Systematic study of weedy *Setaria* species (L.) P. Beauv. in Iran

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## Introduction

The Genus *Setaria* (L.) P.Beauv. (Poaceae, Panicoiodae, and Paniceae) comprises three weedy annual species in Iran (*S. glauca*, *S. verticillata* and *S. viridis*). Weedy *Setaria* species compose one of the worst weed groups interfering with agriculture worldwide, and the species are also troublesome in other disturbed and managed habitats. Different characters like upper lemma texture, awn color and ligule shape are used to distinguish between the different *Setaria* species in Iran, but it seems there is a lot of morphological overlap between these species (specially a lot of variation is shown in *S. viridis*). This is the first systematic study of this species group in Iran.

## Materials and methods

Accessions of three species (*S. glauca*, *S. verticillata* & *S. viridis*) from different parts of Iran (40 populations) were collected (Figure 1). Morphological traits from vegetative and reproductive parts were studied for at least 10 individuals of each population. Anatomical structure of dorsal epidermis was studied by removing upper tissues and staining by methyl green. Transections of leaf blade were studied after following the staining combination method (carmine & methyl green). Quantitative and qualitative morphological and anatomical characters were analyzed by SPSS version 9.1.



Figure 1. Map of Iran, showing the different sites where accessions of *Setaria* species were collected.

## Results and discussion

Anatomical studies showed that the number and shape of different cells like guard and short cell, kind of micro and macro hairs, the ending point of long cells, the frequency of short cells and stomata in coastal and inter-coastal part of the leaf blade are diagnostic. Cluster analysis and dendrogram using Ward method showed that chosen quantitative and qualitative

characters were distinctive for *Setaria* species group in Iran. An identification key based on anatomical features is provided (Figure 2).

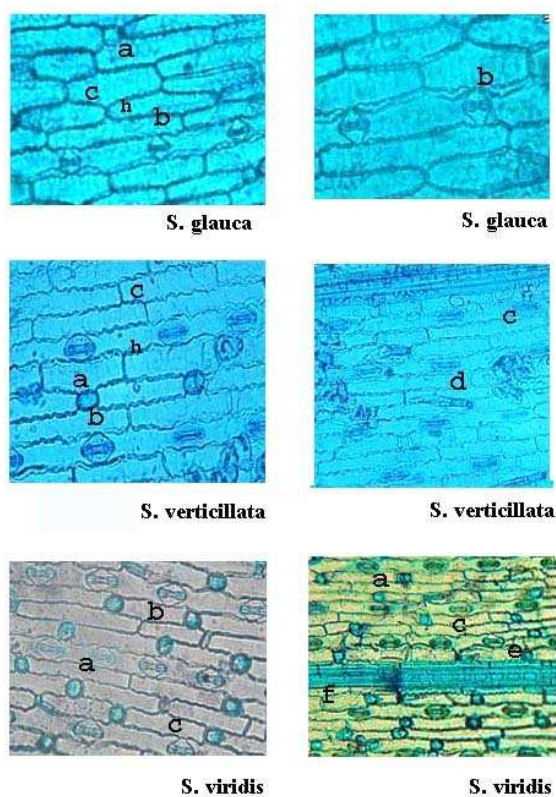


Figure 2. Anatomical differences of dorsal epidermis of weedy *Setaria* species in Iran. a) Short cell, b) Guard cells, c) Long cell, d) Cylindrical hairs, e) Coastal zone, f) Intercoastal zone, g) Silica cell, h) Long cells ending



# Study of vegetative reproduction and regeneration of *Lactuca tatarica*

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## Introduction

*Lactuca tatarica* belongs to the *Asteraceae* family and originates from south-east Europe and Central Asia. Secondly, it expanded to many European countries. It was spread from the east by railway to Czech republic. The species is able to grow in lowlands, but also in mountainous areas, up to 5000 metres above sea level. In the area of origin it occurs as a troublesome hard to control weed, which has expanded extensively. On arable land it reproduces mainly vegetatively. Jehlík (1998) described that the root system can be 400-500 cm long. Because the weed has a very good adaptability to the environment combined with a very high reproductive ability it is able to spread to new localities. In our experiments, we investigated the vegetative reproduction and regenerative ability of *Lactuca tatarica*.

## Materials and methods

Study of biomass production of plants: On 23 April 2004, single plants of 10 cm high were planted in pots with a diameter of 34 cm and 37 cm high. The pots were exposed to natural temperatures and regular watering was applied to avoid a moisture deficit in the substrate. Above and below ground biomass and the length of the root system were evaluated.

Study of regeneration from vegetative organs: In pots with diameter of 17 cm and 108 cm high five rootstock segments with a length of 5 cm and a diameter of 0,3 cm were put at different depth of soil: 10, 20, 30, 40 cm. The experiment was evaluated continuously and the number of emerging shoots was observed.

Study of regeneration of seedlings in different plant growth stages: In pots of 10x10 cm 20 seeds were sown. After plant emergence, the above ground mass was cut at different plant growth stages: 2–3 true leaves (9.4.2004), 3-4 true leaves (16.4.2004) and 5-6 true leaves (7.5.2004). Regeneration (above ground shoots) was evaluated on various dates: 16.4., 27.4., 7.5., 14.5., 21.5., 28.5., 4.6., 14.6.2004. Each variant had 5 replications.

## Results and discussion

Study of biomass production of plants: During the vegetation, the above and below ground biomass increased continuously until 24 September. At this date the above ground mass was 116 g, the below ground mass was 160 g and the length of the rootstock was 4297 cm. At the turn of September and October the temperatures decreased. The above ground biomass decreased, as was measured during the last evaluation. The plants already prepared for the end of the vegetation period. The above ground material got dry and plants concentrated their energy to production and preservation below ground mass. At the end of the vegetation period (around 7 October) *Lactuca tatarica* had produced a rootstock length of 5082 cm, 71 g of above-ground and 156 g of below-ground biomass in good water conditions. As in our previous research (Kneifelová and Mikulka, 2003), the perennial weed reacts to the wet period by production of a larger root system.

Study of regeneration from vegetative organs: New shoots emerged at different rates, depending on the depth at which root segments were buried in the soil (Fig. 1). Regrowth of the root segments from the depth of 10 cm was high and regeneration of the root segments from the deeper layers was low. In some cases, more shoots emerged from one root segment.

Study of regeneration of seedlings in different plant growth stages: Seedlings of *Lactuca tatarica* are not able to regenerate until the plant has reached the 2 true leaves growth stage. Plants of growth stage 3-4 true leaves kept a regeneration percentage of 60% by the whole time of evaluation. The best regeneration was obtained with plants cut in growth stage 5-6 true leaves. With these plants a regeneration of more than 100% was achieved for 14 days. In some cases, more shoots emerged from one seedling and that explains why the regeneration was higher than 100%. Plants of higher plant growth stages proceeded to have a regeneration as high as the plants in growth stage 5-6 true leaves.

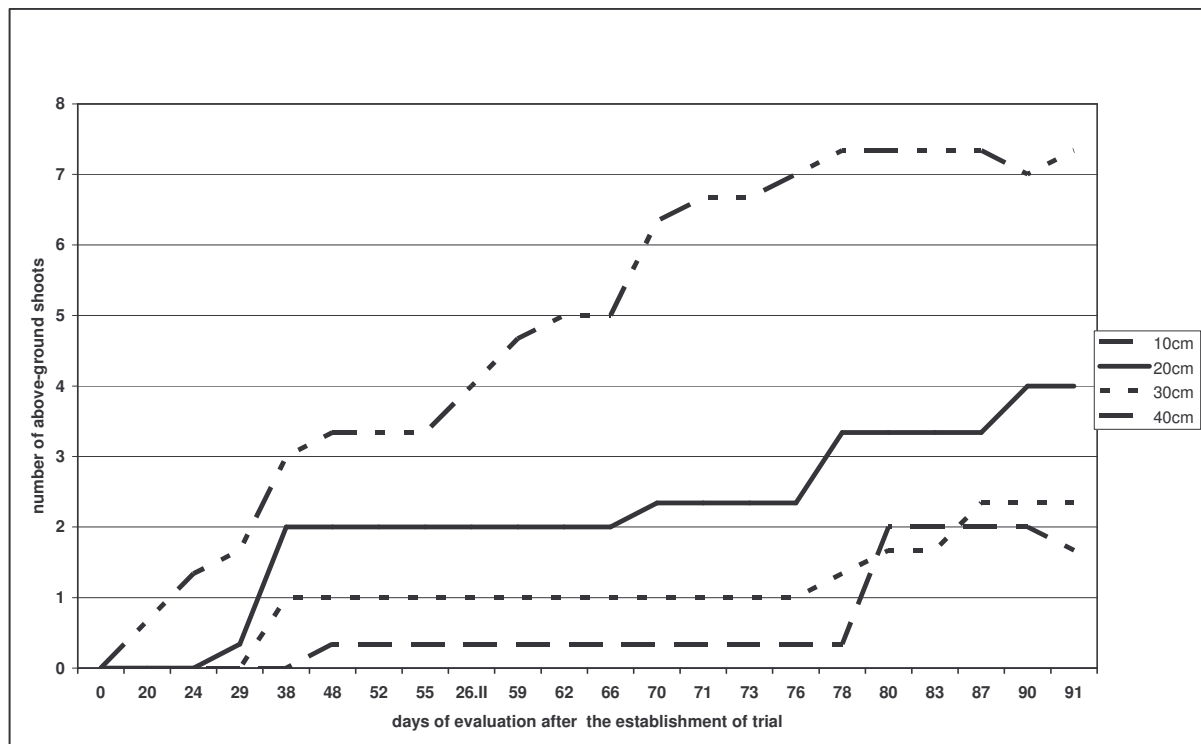


Figure 1. Regeneration of *Lactuca tatarica* from vegetative organs buried at different depths in the soil (10 – 40 cm)

**Acknowledgements:**

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# Competition for nitrogen between corn (*Zea mays* L.), common lambsquarters (*Chenopodium album* L.) and green foxtail (*Setaria viridis* L.) in early stages of growth

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## Introduction

Nitrogen (N) is the nutrient that is most often limiting in situations of crop-weed competition. Competition for nitrogen between corn (*Zea mays* L.) and weeds is influenced by N amount and weed species (Tollenaar et al., 1994). Prior to silking approximately 65-80% of the total N is taken up by corn (Rajcan and Tollenaar, 1999). Therefore, the evaluation of N competition in early stages of corn growth is very important. Increasing N can increase the ability of cereals to suppress weeds (Grundy et al., 1993), however the effect on individual weed species differs.

Common lambsquarters (*Chenopodium album* L.) and green foxtail (*Setaria viridis* L.) are two important weeds in corn production systems. Studies on the physiology of competition for N between corn and these two weeds are missing in the literature. The objective of this study was to investigate the effect of N supply on interspecific competition between corn, common lambsquarters and green foxtail in early stages of growth by measuring plant dry weight, growth traits, N content, N partitioning and N use efficiency (NUE) of these plants.

## Materials and methods

A pot experiment was conducted at the University of Guelph's weeds lab in a glasshouse with controlled environment. Corn, common lambsquarters and green foxtail seeds were planted in 6L plastic pots (25 cm diameter) containing a backed montmorillonate clay growth medium under 16 h photoperiod, 23/18  $\pm 2^{\circ}$  day/night temperature and 60% RH.

The experiment tested 21 treatments consisting of three levels of N supply (low= 1.5 mM N, medium= 5 mM N, high= 15 mM N) and seven species combinations (monoculture of each species, mixture of corn and each weed species, mixture of corn and both weeds species) that were combined factorially in a randomized complete block design. The experiment was replicated three times.

All plant parts were harvested individually when the corn (in monoculture and medium level of N treatment) reached the V8 stage of growth. Aerial plant materials were dissected into leaves, stems and roots. After measuring leaf area, the dry weight of all plant parts was recorded. The N percentage of these fractions was determined using auto analyser by Kjeldahl method. The effects of N supply and competition were determined by analysis of variance on the data recorded for each separate species. The relative biomass (RB) of each species was calculated as biomass per plant in mixture / biomass per plant in monoculture.

## Results and discussion

At the low level of N, the weeds had low dry weight during the early stages of growth, so they had no significant effect ( $P < 0.05$ ) on corn when competing separately. At low resource levels, interspecific competition between individuals is less intense, perhaps because under such conditions individuals are growing too slow to affect neighbours strongly. Lack of competition may also be related to the small size of seeds of common lambsquarters and green foxtail compared to corn seed. Davis and Liebman (2001) noted that minimized nutrient availability at early stage of growth may limit the growth of small-seeded weeds without

compromising the growth of crops with larger seeds. Competition of weeds at the medium level of N decreased the height, dry weight and LAI of corn. The effect of weed competition on these traits was higher at medium than at high N. The height, LAI and shoot dry weight of corn at medium level of N were 80%, 63% and 60% lower in mixture than in monoculture, respectively. At high level of N, reductions of 9%, 29% and 25% were found for these respective traits. It has been suggested that, if two species compete for a given resource, e.g. N, addition of that resource will reduce its deficiency and so reduce the intensity of competition (Wilson, 1988).

The significant interaction between weed competition and N supply on corn dry weight suggests that N was an important factor in competition. The relative competitive ability of corn and weeds were influenced by N supply. At medium and high levels of N, common lambsquarters was more competitive than green foxtail, but the whole plant relative growth rate was similar between the weeds species at low level of N. Increasing N (from medium to high) increased the RB of corn, but the RB of weeds was not influenced by N supply.

Competition decreased the N content of both corn and weeds, but in mixtures N content of weeds was higher than that of corn. Specific leaf N of all plants was not influenced by competition and N. Competition for N increased the root / shoot ratio of corn just at medium level of N, but had no effect on partitioning of N between root and shoot. Several studies have indicated that soil N availability, although strongly altering shoot growth, does not significantly affect the root dynamics (Gastal and Lemaire, 2002). It probably relies on the increase in root / shoot ratio with decreasing N supply and competition for N.

The NUE of corn in monoculture decreased as N supply increased, but was not influenced by N supply in mixture. The NUE of weeds, in contrast with that of corn, was influenced by N supply in monoculture and mixture. These observations suggest that common lambsquarters and green foxtail may uptake more N compared to the corn in mixture. Although the growth of both weed species was affected by N deficiency and competition, there was no significant interaction between competition and N for growth traits, N content and NUE of the weeds.

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# Dry matter production and partitioning of redroot pigweed (*Amaranthus retroflexus* L.) in pure stands and in mixed stands with corn (*Zea mays*)

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## Introduction

Mechanistic models of competition between crops and weeds are generally based on crop growth models which explicitly represent the processes of radiation interception, dry matter production and distribution (Rohrig & Stutzel, 2001). The assimilate partitioning among different plant organs is important for process based modeling of plant growth, however there is often not enough information on biomass partitioning of weeds under different levels of competitive stress. Most simulation models apply partitioning coefficients which change based on plant phenology. Allometry is another approach to separate the effects of treatments from ontogeny and to estimate the effects of competition on partitioning (Gunn et al., 1999). Allometric relationships are a simple and effective way to describe the intensity of competition. As little information on dry matter production in relation to light interception is available for *A. retroflexus* this aspect was studied in a field experiment, where also the biomass partitioning of this weed was determined.

## Materials and methods

The experiment was conducted at an experimental field of the Ferdowsi University of Mashhad in Iran during the 2001 growing season. A split – plot design in 3 replications was used. Main plots consisted of two planting dates (May 21 and June 6), whereas subplots contained a pure stand of *A. retroflexus* and mixed stands of this weed with corn, where the crop was sown at either 7.1 or 9.5 plants per m<sup>2</sup>. In all treatments *A. retroflexus* was planted at 9.5 plants per m<sup>2</sup>. Corn was planted in rows with a distance of 70 cm between the rows and an in-the-row distance of either 15 cm (high density) or 20 cm (low density). In order to avoid nutrient and water limitations, all treatments were ample fertilized, based on soil analyses, and well watered.

For both planting dates, measurements were conducted at three stages during the growing period, at 20, 45 and 75 days after planting. Around solar noon, the fraction of transmitted photosynthetically active radiation (PAR) was determined with a line quantum sensor (LI-COR 191 SA) below and a point sensor (LI-COR 190 SA) above the canopy. Five measurements were taken per plot. Simultaneously with light interception, total above ground dry matter of weed and crop were determined. At the end of the growing season the allocation of shoot biomass between stems and leaves was investigated using the allometric growth equation  $LnW_s = a + bLnW_L$  (equation 1), where  $W_s$  (gm<sup>-2</sup>) and  $W_L$  (gm<sup>-2</sup>) represent stem and leaf biomass, respectively and  $a$  and  $b$  are coefficients. Equation 1 implies a fixed ratio,  $b$ , between the relative growth rates of stems and leaves. This parameter is also called “allometric growth coefficient” and can be considered a measure for the intensity of the differential variation between the growth of the two plant organs. If treatments have an effect on partitioning, this will become evident from differences in estimated  $b$ -values among treatments (Rohrig & Stutzel, 2001).

## Results and discussion

Our results showed that at both planting dates, *A. retroflexus* dry matter decreased with an increased corn biomass (Fig.1a). For both planting dates total weed dry matter in pure stands was positively correlated with the amount of intercepted PAR (Fig.1b). It is most likely that

the reduction of weed biomass in mixed stands relative to its production in pure stand was due to a reduced light interception, as in all treatments an ample supply of water and nutrients was provided. McLachlan et al. (1993) reported that the radiation transmitted through the corn canopy declined when corn density increased, which may explain the reduction in *A. retroflexus* dry matter.

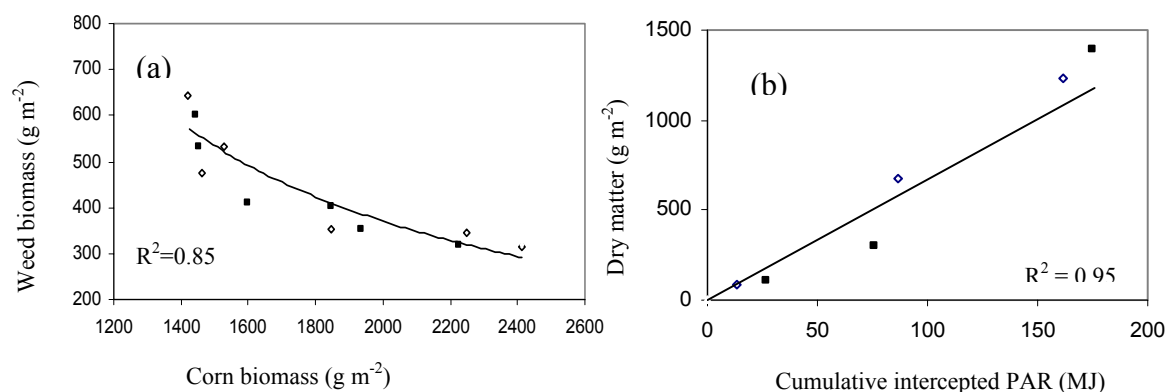


Figure 1. Relationship between corn and weed biomass at the end of the growing period in mixed stand treatments (a), and relationship between total dry matter and cumulative intercepted PAR by *A. retroflexus* (b), at planting date 1 ( $\diamond$ ), and planting date 2 ( $\blacksquare$ ).

Table 1 shows a close linear relationship between the natural logarithms of stem and leaf dry weight as calculated by equation 1. In the presence of corn, and particularly at higher corn density when light availability down the canopy is less, partitioning was characterized by a higher *b* coefficient, indicating that in competition situations *A. retroflexus* partitioned relatively more biomass to its stem.

Table 1. Equation coefficients, their corresponding standard error (SE) and the coefficient of determination ( $R^2$ ) of the allometric relationship between *A. retroflexus* leaf and stem biomass:  $\ln W_S = a + b \ln W_L$ .

Treatments		a	SE	b	SE	$R^2$
Planting date 1	Pure <i>A. retroflexus</i>	-1.77	1.06	1.624	0.23	0.94
	<i>A. retroflexus</i> in low corn density	-2.090	0.78	1.708	0.19	0.96
	<i>A. retroflexus</i> in high corn density	-2.490	0.36	1.770	0.08	0.99
Planting date 2	Pure <i>A. retroflexus</i>	-2.049	0.69	1.566	0.15	0.97
	<i>A. retroflexus</i> in low corn density	-2.345	0.78	1.731	0.19	0.96
	<i>A. retroflexus</i> in high corn density	-2.753	0.76	1.827	0.19	0.96

Even though photosynthetic area is reduced, this adjustment in dry matter partitioning can be regarded as a survival strategy of *A. retroflexus* to intercept more light high up in the corn canopy. This growth pattern matched the strategy of shade avoidance in which stem elongation is favored over leaf area (Caton & et al., 1997).

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## Multispecies competition effects of weeds on wheat

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### Introduction

Most economic threshold studies are based on weed population present at crop maturity at the end of growth season (Coble & Mortensen 1992; Coble et al., 1981), whereas weed control decisions based on weed numbers are made very early in the crop season. In addition, these studies consider interference between single weed species and crop, while there are several species in the field. The objective of this study was to determine economic damage threshold of dominant weeds on wheat based on weed populations present at early season and to study the effects of multispecies competition of weeds on yield and yield components of winter wheat in the field conditions.

### Materials and Methods

The experiment was conducted in 2002 at Agricultural Research Station of Ferdowsi University of Mashhad. At the beginning of shooting stage, 30 points were sampled systematically using a 50 cm x 50 cm quadrat. Then number of wheat plants and tillers and number of weeds (each species separately) was counted without any damage to the plants. At the end of growth season, all weeds in the quadrats were counted, separately. Besides, all the wheat plants in the same quadrat were harvested and number of plants, seed weight and total biomass were measured. LnW regression model was used to determine the relative fractions of inter- and intra-specific competition (Hashem et al., 1998; Equation 1).

$$\ln W = b_{co} + b_{cc}N_c + b_{cw1}N_{w1} + \dots + b_{cw8}N_{w8} \quad (1)$$

where, W is the biomass or yield per wheat plant,  $b_{co}$  is the intercept of regression equation,  $b_{cc}$  is an intra-specific competition coefficient of wheat,  $N_c$  is the wheat density,  $b_{cwi}$  is the inter-specific competition of wheat and weed species i and  $N_{wi}$  is the density of the ith weed species. Through Equation 1 a weed species indicated as effective weed when its inter-specific competition coefficient was statistically significant by a t-test.

To determine the economic damage threshold, Swinton (1994) method was used. At first, extended rectangular hyperbola model was fitted to wheat yield and the density of effective weeds on wheat yield, to estimate A and  $I_i$ .

$$Y = Y_{wf} \left[ 1 - \frac{\sum I_i w_i}{100 \left( 1 + \frac{\sum I_i w_i}{A} \right)} \right] \quad (2)$$

where  $I_i$  is the percentage yield loss associated with the weed species i,  $W_i$  is the density of weed species i, A is the maximum percentage of yield loss asymptote and  $Y_{wf}$  is the yield in weed free crop.  $I_i$  coefficients can be converted into the kind of competitive index values associated with the total competitive load (TCL) by dividing through by the largest  $I_i$  (that of the most competitive weed) as follows:

$$TCL = w_1 + \frac{I_2}{I_1} w_2 + \frac{I_3}{I_1} w_3 + \dots + \frac{I_n}{I_1} w_n \quad (3)$$

$W_1$  is the density of the most competitive weed,  $I_1$  is the percentage of yield loss associated with the weed species 1 and  $I_2, W_2, I_3, W_3, I_n, W_n$  are the coefficients of the other effective weeds.

Percentage of yield loss in different quadrats was calculated as:

$$YL(\%) = \frac{(Y_{wf} - Y)}{Y_{wf}} \times 100 \quad (4)$$

where  $Y_{wf}$  was estimated from equation 2.

Using TCL and substituting it for density in two-parameter hyperbolic model, the new A and I were calculated.

$$YL(\%) = \frac{I(TCL)}{1 + \frac{I(TCL)}{A}} \quad (5)$$

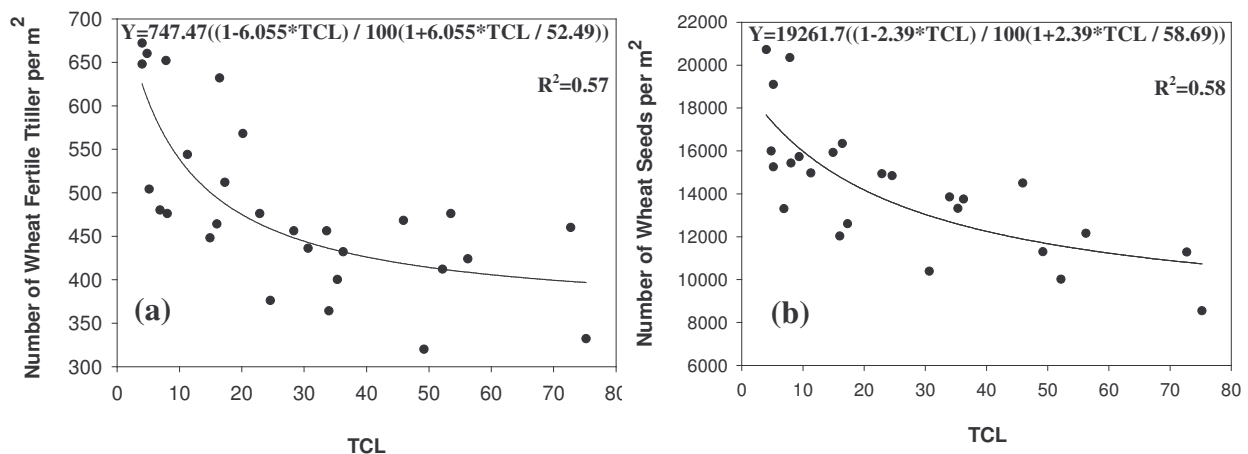
Using new A and I (estimated by equation 5), economic damage threshold was calculated by O'Donovan (1991) equation:

$$D = \frac{1 - \left[ \frac{YP - H}{YP} \right]}{\frac{I}{100} + \frac{I}{A} \left[ \frac{YP - H}{YP} - 1 \right]} \quad (6)$$

where D is the economic damage threshold density according to TCL, Y is the yield in weed free crop (kg.ha<sup>-1</sup>), P is the price of wheat (0.13 € per kg) and H is the chemical control costs (15 € per ha).

## Results and discussion

The weeds presented in the field were *Avena ludoviciana*, *Chenopodium album*, *Solanum nigrum*, *Stellaria holostea*, *Fumaria* spp., *Polygonum aviculare*, *Sonchus* spp. and *Convolvulus* spp. LnW regression model indicated that only *Avena ludoviciana*, *Convolvulus* spp. and *Chenopodium album* had a significant reduction effect on wheat yield. (data not shown). The economic damage threshold of effective weeds was determined about 5.23 TCL; that was equivalent to 5.23 plants per m<sup>2</sup> of *Avena ludoviciana* or 12.20 plants per m<sup>2</sup> of *Convolvulus* spp. or 9.65 plants per m<sup>2</sup> of *Chenopodium album*. Regression analysis showed that weed density based on TCL reduced number of fertile tillers (Fig.1a) well in line with results found by Morishita et al. (1991) in barley. Unusual low R<sup>2</sup> values, in spite of their significance, could be mainly due to heterogeneous nature of multi-specific field data.



**Fig.1.** Relationship between TCL and a) number of fertile tiller and b) wheat seeds.

Number of wheat seeds per m<sup>2</sup> was considered as an index for effect of weed competition on wheat yield. In this study, increasing of weed density based in TCL was led to reduction in wheat seed per m<sup>2</sup> (Fig.1b). However, TCL had no effect on harvest index and kernel weight of wheat (data not shown).

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# Generative and vegetative reproduction of *Bolboschoenus laticarpus* and *Bolboschoenus planiculmis*

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## Introduction

In the Czech Republic *Bolboschoenus laticarpus* and *Bolboschoenus planiculmis* have rapidly spread on arable land mainly in the last 15 years (Hroudová *et al.*, 1999). Both are harmful weeds especially in sugar beet, maize, sunflower, potatoes and field vegetables (Cure & Acock, 1986). Both species occur either separately or in mixed populations (Mikulka & Kneifelová, 2004). The species mainly reproduce vegetatively, through tubercles, but generative reproduction through seed has also been proven. A significant increase in the occurrence of these species was caused by minimum tillage technology and the use of herbicides to which the two species are tolerant.

## Materials and Methods

Plants of *B. laticarpus* (Pardubice locality) and *B. planiculmis* (Čejč locality) were chosen for experiments. Plants were grown in pots 45 cm in diameter and 50 cm in height. Potting substrate consisted of a 1:1:1 mix of clayey topsoil, sand and peat substrate. Tubercles of similar size were taken from maternal plants. One tubercle was planted per pot to a depth of 5 cm. The number of replications was five. Planting was carried out on April 24. Pots were regularly watered during the vegetation period. Evaluations were made on May 31, June 25, July 16, August 23 and September 24. We determined root and shoot biomass, number of tubercles and shoots, and at the end of the vegetation period we evaluated number of inflorescences, number of seeds per inflorescence and total seed production per plant.

## Results and Discussion

The increase in shoot biomass, root biomass, shoot number and tubercle number was determined in monthly intervals. On both species, shoots were formed relatively quickly in a short time after planting. Tubercles were formed as soon as two months after planting. *B. laticarpus* plants were more sizeable and produced a higher amount of shoot (75.1 g) and root (275.6 g) biomass compared to *B. planiculmis*, which produced 50.3 g of shoot and 125.8 g of root biomass. A decrease in the weight of both species was found during the last evaluation as the end of the vegetation period approached. Despite the lower weight, *B. planiculmis* plants formed a higher number of shoots, as earlier described by Hroudová *et al.* (1999). For both species the highest increment in tubercles was recorded in June and July. Tubercles of *B. laticarpus* were larger than those of *B. planiculmis*. One plant of *B. laticarpus* had on average four flowering stalks with an average number of 85.9 seeds per inflorescence. Total production was 391 seeds per plant. One plant of *B. planiculmis* had a markedly higher number of flowering stalks (16.8) than *B. laticarpus*. Average number of seeds per inflorescence was 48.6. Total number of seeds per plant was 755. Seed production per inflorescence was higher in *B. laticarpus* compared to *B. planiculmis*. But the latter species had a substantially higher number of flowering stalks.

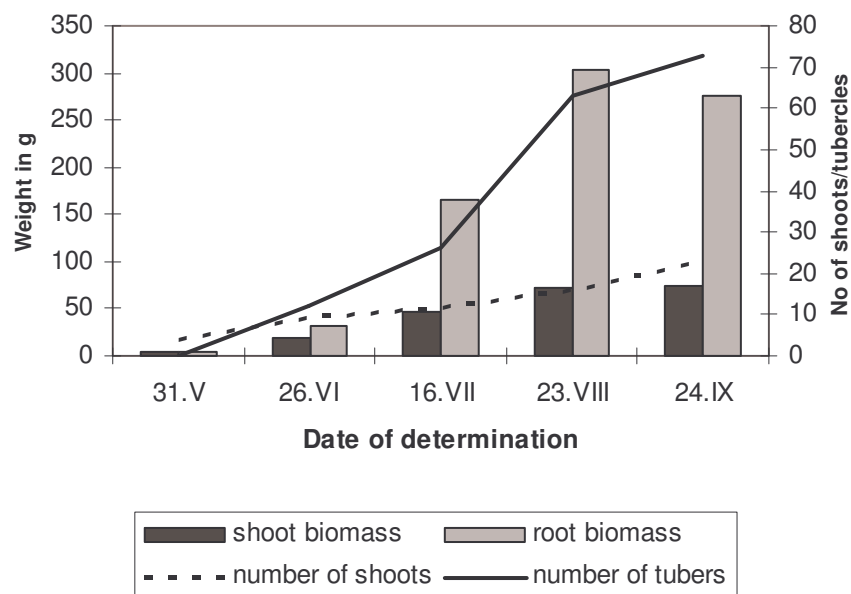


Figure 1. Production of root and shoot biomass, number of shoots and number of tubercles in *Bolboschoenus laticarpus*

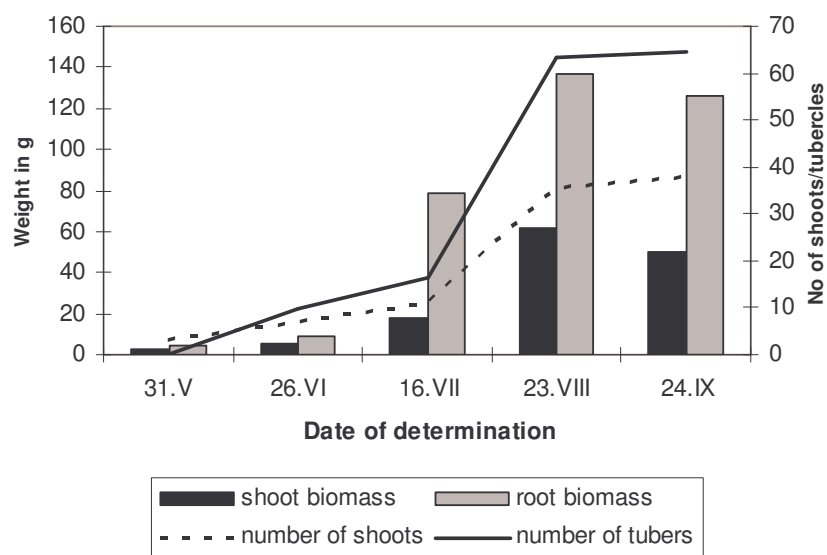


Figure 2. Production of root and shoot biomass, number of shoots and number of tubercles in *Bolboschoenus planiculmis*

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# Nutrient uptake of *Convolvulus arvensis* throughout the vegetation period

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## Introduction

*Convolvulus arvensis* is one of the most undesirable agricultural weeds of the world (Holm *et al.*, 1977). It is native to Europe. On the basis of the IV<sup>th</sup> - Hungarian National Weed Survey 1996-1997, it takes the sixth place in the dominance sequence, with an average coverage of 1.66 % (Hunyadi *et al.*, 2000). The fast-growing and twining *Convolvulus arvensis* is one of the most important perennial weeds of agricultural fields, vineyards, gardens and roadsides because it difficult to eradicate. This weed has a vertical and a horizontal root system, and is able to root to a depth of 2 m or more. Stems are 20-100 cm long. Although initially dispersed by seeds to new sites, it also can reproduce successfully and vigorously by underground rootstocks (Swan & Chancellor, 1976). Fragmentation of rhizomes is one of the primary mechanisms by which it disperses and persists in cultivated fields (Buhler *et al.*, 1994). Germination of seeds is slow, due to the hard seed coat and consequently seeds are able to remain in the soil for long periods. Its deep rhizomes provide another important dormancy mechanism for survival.

Bindweed root systems apparently do not utilize only the same soil-water and nutrient resources as most of the cultivated crops. However, under conditions of water stress field bindweed can be a better competitor than most cultivated crops. In the interest of effective weed control we were interested in knowing the biological characteristics and nutrient uptake of weeds in detail (Lehoczky *et al.*, 2003; Nádasy *et al.*, 2004). Our aim was to follow changes in nutrient content in shoot and root of *Convolvulus arvensis* throughout the growing season.

## Materials and methods

*Convolvulus arvensis* plants were collected from the fields in the surrounding of Keszthely in Hungary from April to December 2002. The type of soil was Ramann's brown forest soil. Nutrient content of the soil was examined. Main characteristics of this soil were: humus 1.88 %, mineral-N 11.07 mg kg<sup>-1</sup>, AL-P<sub>2</sub>O<sub>5</sub> 152 mg kg<sup>-1</sup>, AL-K<sub>2</sub>O 168 mg kg<sup>-1</sup>, and a pH<sub>(H<sub>2</sub>O)</sub> 7.05. Fresh- and dry- weights of root and shoot samples were measured. Nitrogen concentration was determined by Kjeldahl method, phosphorus concentration by spectrophotometer, potassium and calcium concentration by flame photometer.

## Results and discussion

Growing season 2002 was a really dry period with hardly any rain. Nitrogen content in the shoot dry matter altered highly between 1.7-3.7 % (Fig. 1.). At the beginning of May the N-content decreased as a consequence of vigorous growth accompanied by dilution of nutrients. After this time the content rapidly rose to 3.7 %, and later decreased gradually. N-content of root material varied between 1.1-2.2 % and changed less than in shoots. Phosphorus content altered opposite to nitrogen. P-content between 0.18-0.97 % in shoots and between 0.15-0.84 % in roots was measured. In autumn N- and P-content of roots increased gradually because of nutrient storage.

According to our observations *Convolvulus arvensis* can take up potassium in great quantities during flowering (Fig. 2.). Potassium content in roots and shoots was highest at the beginning of September and decreased rapidly after that period. Shoots contained more potassium than roots. Calcium content was lower than potassium. In shoots it altered between 0.5-1.3 %. In contrast to potassium, roots contained more calcium (0.8-1.4 %) than shoots.

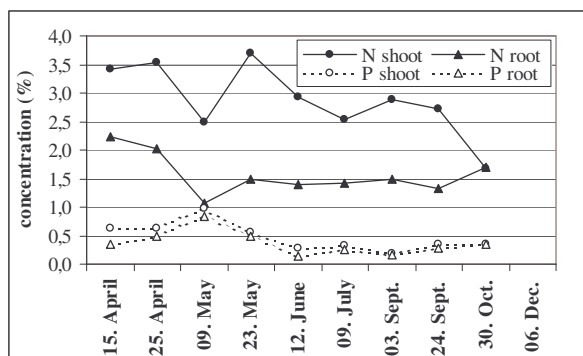


Figure 1. Nitrogen and phosphorus content in shoots and roots of *Convolvulus arvensis*

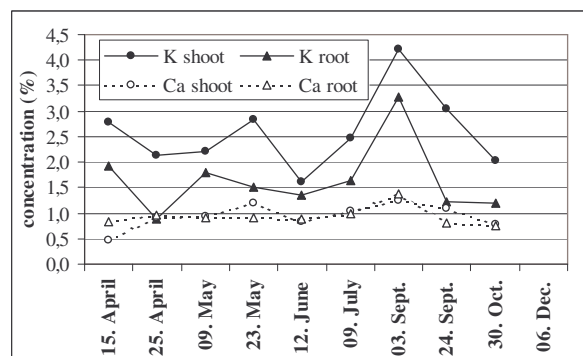


Figure 2. Potassium and calcium content in shoots and roots of *Convolvulus arvensis*

The rootstocks serve as nutrients storage for the plants. The high competitiveness of the species is due to its extensive and deep root system, which is mainly a network of underground rootstocks. Changes in nutrient content of shoots and roots would connect with physiological processes of the plant. Intensive nutrient uptake of *Convolvulus arvensis* has an important role in its considerable competitive capacity. Nitrogen, phosphorus and calcium concentration of shoots were higher than roots during the actual vegetation period, and became more or less identical at the end of October. Potassium content of shoots remained higher than in roots throughout the examined period.

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# Effect of length of storage, light and temperature on germination behaviour of eight tropical weed species

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## Introduction

Seed germination in controlled conditions seems to be one way of helping to understand the emergence and the chronology of emerging species in the field. Within the frame of this work, the influence of length of storage, light and temperature on germination capacity and dormancy was evaluated in order to understand some ecological requirements of the seeds of eight dominant weed species of millet and groundnut crops in the “Bassin Arachidier” (Groundnut Basin) of Sénégal. The species were: *Spermacoce chaetocephala*, *Hibiscus asper*, *Dactyloctenium aegyptium*, *Indigifera hirsute*, *Eragrostis tremula*, *Mitacarpus villosus*, *Sesbania pachycarpa* and *Cassia obtusifolia*.

## Materials and methods

Three year old stored (from 1993 to 1996) and freshly harvested seeds were placed on blotting-paper in petri dishes and put in four air-conditioned enclosures with constant temperatures of 20, 25, 30 and 40 °C. The surrounding walls were divided into 2 compartments; one in continuous light and one under continuous darkness. The duration of the tests was 21 days. The statistical analysis of the rates of germination was carried out by analysis of variance (ANOVA).

## Results and discussion

The length of storage had a significant effect on the germination rate of the eight species. This effect was positive for seeds of *Spermacoce chaetocephala*, *Cassia obtusifolia*, *Eragrostis tremula*, *Dactyloctenium aegyptium*, *Mitracarpus villosus* and *Hiscus asper* and negative for seeds of *Indigofera hirsuta* and *Sesbania pachycarpa*. This result expresses an increase in the germinability of seeds with time and demonstrates the difficulty of freshly harvested seeds to germinate. These difficulties are related to dormancy (e.g. unripe embryo's).

Without dormancy related to seed storage condition:

- seed germination rate of most species was increased with increased temperatures up to 30 or 40 °C. An exception was *Sesbania pachycarpa*;
- light promoted the germination of seeds of *Eragrostis tremula* and *Mitracarpus villosus*; this response is identical to that of species with photosensitive seeds.

Table 1 : ANOVA table indicating the response of various weed species to storage length, temperature and light. (\*\*\*) Very significant,  $p < 0.0001$  ; (\*\*) significant  $0.001 < p < 0.01$  ; (\*) not very significant  $0.01 < p < 0.1$  ; (NS) not significant  $p > 0.1$

	<i>Sperma</i>	<i>Hibis</i>	<i>Dactylo</i>	<i>Indigo</i>	<i>Erag</i>	<i>Mitra</i>	<i>Sesb</i>	<i>Cassia</i>
Storage length	***	***	***	***	***	***	***	***
Temperature	**	**	***	***	NS	NS	NS	*
Storage * temperature	NS	**	***	***	*	NS	NS	*
Light	NS	NS	NS	NS	***	***	NS	NS
Storage * Light	NS	NS	NS	NS	***	***	NS	NS
Temperature * Light	***	NS	NS	**	***	***	NS	*

Table 2 : Germination rates (%) of eight different weed species as affected by storage length (Year), temperature (Temp) and light (L/O)

Year	Temp	L/O	<i>Sperm</i>	<i>Hibis</i>	<i>Dactylo</i>	<i>Indigo</i>	<i>Erag</i>	<i>Mitra</i>	<i>Sesb</i>	<i>Cassia</i>
1996	20	L	0.0 <sup>c</sup>	0.5c	0.25d	23.0defg	0.50de	2.00def	8.5cb	7.5h
1996	20	O	0.5c	0.0c	0.00d	45.0b	0.00 <sup>c</sup>	5.25bcd	9.0cb	12.0fgh
1996	25	L	1.0c	0.5c	0.00d	22.5defg	2.75d	11.25b	12.0b	6.5h
1996	25	O	0.5c	0.5c	0.00d	10.0h	1.00de	0.00f	5.5cb	6.5gh
1996	30	L	3.0c	0.5c	0.00d	30.0bcde	0.25de	6.50bc	8.0cb	8.5gh
1996	30	O	0.0c	0.5c	0.00d	27.0def	0.25de	0.50ef	7.5cb	9.5gh
1996	40	L	0.5c	0.5c	0.25d	48.5a	3.00d	3.25cde	39.0a	11.5fgh
1996	40	O	0.5c	0.0c	0.00d	43.0abc	0.50de	0.00f	43.0a	8.5gh
1993	20	L	54.0b	2.5c	24.25a	14.0fgh	49.50b	1.00ef	4.0cb	21.0f
1993	20	O	91.5a	1.5c	14.00b	12.0gh	2.50de	1.00ef	4.0cb	18.0fg
1993	25	L	85.5a	1.0c	19.00ab	29.0cde	58.50b	3.00cde	2.5cb	33.5 <sup>c</sup>
1993	25	O	93.5a	3.5bc	19.75ab	25.0defg	1.75de	17.75a	1.5c	53.5d
1993	30	L	96.0a	9.0ab	26.50a	24.5defg	90.25a	17.25a	3.0cb	64.0c
1993	30	O	93.5a	4.5bc	21.50ab	16.0efgh	2.00de	9.75b	4.0cb	77.5b
1993	40	L	95.5a	13.5a	4.75c	33.0bcd	14.75c	3.00cde	7.5cb	92.0a
1993	40	O	61.5b	9.0ab	4.25c	35.0abcd	0.25de	0.25f	3.0cb	87.0a

Taking all of these results into consideration, it appears that among the three factors (storage length, temperature and light) storage length seemed the most prominent factor determining the seed germination of *Spermacoce chaetocephala*, *Hibiscus asper*, *Dactyloctenium aegyptium*, *Indigofera hirsuta*, *Sesbania pachycarpa* and *Cassia obtusifolia*. On the other hand, light was found to be the most preponderant exogene factor affecting seed germination of *Eragrostis tremula* and *Mitracarpus villosus*.

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# Allelopathic effect of potato tuber shoots on germination and seedling growth of some crops

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## Introduction

Allelopathy is defined as the direct or indirect harmful or beneficial effect by one plant (including micro-organisms) on another through the production of chemical compounds excreted into the environment (Rice, 1984). The interactions “weeds against weeds” and especially “crops against weeds” could be used as an alternative ecological method for weed control. The different allelochemicals could be extracted from fresh or dry leaves, stems, roots, fruits, seeds by water or organic solvent extracts, leachedes from leaves, roots or seeds, shoot and root residues or exudates and soil around the allelopathic plants (Qasem, 2001; Oudhia, 2000; Caamal-Maldonado et al., 2001). The main toxic substances having an allelopathic effect are phenolic acids and related compounds, organic derivatives, alkaloids, tannins, glycosides, fatty acids and esters of acids (Patterson, 1981; Chon et al., 2003; Bertholdsson, 2004). The aim of this study was to determine the effect of water extracts of potato tuber shoots on germination and seedling growth of bean, soyabean, lentil, maize and wheat.

## Materials and methods

Cut into fine pieces fresh shoots of potato were allowed to decay for 72, 120, 168, 240 and 336 h in normal water in the ratio of 1:10 w/v in a refrigerator at 6°C. These water extracts were applied on the crop seeds put on a filter paper in Petri-dishes (10 seeds per dish). At the 3<sup>rd</sup>, 5<sup>th</sup>, 7<sup>th</sup> and 10<sup>th</sup> day after sowing (DAS) the percentage of germinated seeds was determined. At the 14<sup>th</sup> DAS height, root length, fresh and dry weight of seedlings was recorded. The data were statistically analyzed by ANOVA followed by *t*-test.

## Results and discussion

Nearly all extracts reduced seed germination (Table 1). The differences between the control and each one of the extracts were analyzed by ANOVA followed by *t*-test at  $p < 0.05$ . For soyabean, lentil and maize nearly all extracts caused a significant reduction in germination. For lentil and maize the lowest negative effect on germination was obtained with the 120-h extract, for bean and wheat with the 168-h extract and for soyabean with the 240-h extract. For seedlings growth the strongest effects were found on soyabean and lentil (Table 2). For maize the mildest effects on seedling growth were found with the 72 and 120-h extracts, for bean and lentil with the 120-h extract, for wheat with the 120 and 168-h extracts and for soyabean with the 240-h extract. The findings on the allelopathic effects of extracts of potato shoots on crops could be used in follow-up experiments for the evaluation of such effects on relevant weed species.

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Table 1. Germination percentage of seeds of various species at 3, 7 and 10 DAS as affected by extracts of potato shoots.

D	C	Control		72-h		120-h		168-h		240-h		336-h	
		M-n	SE	M-n	SE	M-n	SE	M-n	SE	M-n	SE	M-n	SE
3	B	50.0	10.0	16.7	6.67	40.0	10.0	60.0	5.77	23.3	3.33	0.0*	0.00
	S	46.7	6.67	0.0*	0.00	0.0*	0.00	0.0*	0.00	70.0	10.0	53.3	3.33
	L	86.7	8.82	16.7*	8.82	56.7*	3.33	30.0*	11.6	0.0*	0.00	0.0*	0.00
	M	86.7	8.82	53.3*	6.67	43.3*	8.82	36.7*	6.67	36.7*	6.67	36.7*	6.67
	W	63.3	6.67	16.7*	3.33	40.0	11.6	53.3	6.67	10.0*	5.77	6.7*	3.33
7	B	63.3	8.82	10.0*	5.77	50.0	10.0	30.0	15.3	13.3*	6.67	0.0*	0.00
	S	70.0	0.00	0.0*	0.00	0.0*	0.00	0.0*	0.00	30.0	15.3	53.3*	3.33
	L	100.0	0.00	0.0*	0.00	56.7*	3.33	36.7*	17.4	13.3*	3.33	0.0*	0.00
	M	100.0	0.00	66.7*	3.33	60.0	15.3	60.0*	10.0	56.7*	12.0	53.3*	8.82
	W	66.7	3.33	16.7*	3.33	20.0	20.0	70.0	5.77	36.7	18.6	20.0*	5.77
10	B	63.3	8.82	13.3*	6.67	23.3	18.6	26.7	12.0	13.3*	6.67	0.0*	0.00
	S	76.7	6.67	0.0*	0.00	0.0*	0.00	0.0*	0.00	30.0*	15.3	76.7	12.0
	L	100.0	0.00	0.0*	0.00	56.7*	3.33	10.0*	5.77	13.3*	8.82	0.0*	0.00
	M	100.0	0.00	66.7*	3.33	60.0	15.3	60.0*	10.0	56.7*	12.0	60.0*	5.77
	W	66.7	16.7	13.3*	3.33	20.0	20.0	70.0	5.77	50.0	0.00	46.7	24.0

\* p<0.05 df = 1, 4 F<sub>theoretical</sub> = 7.71. M-n – mean; SE – standard error; D – DAS; C – crops; B – bean, S – soybean, L – lentil, M – maize, W – wheat.

Table 2. Growth characteristics of seedlings of various species at 14 DAS as affected by extracts of potato shoots.

C	S.	Control		72-h		120-h		168-h		240-h		336-h	
		M-n	SE	M-n	SE	M-n	SE	M-n	SE	M-n	SE	M-n	SE
B	H	3.75	1.89	1.35	0.68	2.99	2.69	0.87	0.24	0.30	0.15	0.00	0.00
	R	1.75	0.73	0.92	0.46	1.23	1.19	0.22	0.12	0.15	0.08	0.00	0.00
	Fr	573.0	0.05	148.*	0.74	512.3	2.89	285.*	0.42	90.0*	0.45	0.00*	0.00
	Dr	120.7	0.00	15.3*	0.08	81.67	0.42	63.7*	0.06	13.7*	0.06	0.00*	0.00
S	H	3.52	0.77	0.00*	0.00	0.00*	0.00	0.00*	0.00	1.41	0.78	0.61*	0.61
	R	1.49	0.56	0.00	0.00	0.00	0.00	0.00	0.00	0.39	0.31	0.06	0.06
	Fr	317.0	0.68	0.00*	0.00	0.00*	0.00	0.00*	0.00	150.3	0.87	76.67	0.77
	Dr	56.0	0.21	0.00*	0.00	0.00*	0.00	0.00*	0.00	29.33	0.22	17.67	0.17
L	H	19.10	2.08	0.00*	0.00	7.94*	0.80	0.28*	0.19	0.14*	0.08	0.00*	0.00
	R	5.91	1.28	0.00*	0.00	3.03	0.59	0.14*	0.10	0.09*	0.08	0.00*	0.00
	Fr	334.7	0.57	0.00*	0.00	225.0	0.14	14.3*	0.10	10.0*	0.06	0.00*	0.00
	Dr	45.67	0.18	0.00*	0.00	22.67	0.00	2.00*	0.00	2.33*	0.00	0.00*	0.00
M	H	10.63	2.38	5.56	1.31	3.93	1.11	1.67*	0.39	0.85*	0.45	2.48*	0.27
	R	5.81	1.90	4.49	1.81	3.15	1.17	0.93	0.24	0.61	0.35	2.61	0.14
	Fr	693.3	1.77	442.7	2.21	355.3	0.75	194.3	0.53	108.*	0.58	285.0	0.49
	Dr	158.7	0.48	90.67	0.50	100.0	0.25	55.00	0.13	30.67	0.18	82.33	0.10
W	H	5.03	0.33	2.12*	0.28	3.12	3.12	2.79	0.99	0.00*	0.00	1.71*	0.90
	R	2.11	0.26	0.80*	0.15	2.00	2.00	1.39	0.49	0.00*	0.00	0.68*	0.34
	Fr	81.00	0.10	37.0*	0.06	19.7*	0.20	53.33	0.06	0.00*	0.00	37.33	0.19
	Dr	9.00	0.00	4.33*	0.00	7.33	0.07	9.67	0.03	0.00*	0.00	7.33	0.04

\* p<0.05 df = 1, 4 F<sub>theoretical</sub> = 7.71. M-n – mean; SE – standard error; Cr – crops; B, S, L, M, W – as in Table 1; S. ch. – seedling characteristics; H – height (cm), R – root length (cm), Fr – fresh weight (mg), Dr – dry weight (mg).

# Inter- and intra-species competition for applied nitrogen

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## Introduction

In plant communities, individual plants compete for nutrients. In communities of cultivated plants the competition between species (crop and weeds) and between individual crop plants or crop rows may be affected by the distribution pattern of the applied nutrient source. Direct injection of nitrogen (N), either applied as mineral N or slurry N, may improve the competitiveness of a cereal crop in relation to weeds compared with uniform distribution of the applied N within the soil volume (e.g. Kirkland & Beckie, 1998; Rasmussen, 2002). In addition, the effect of mechanical as well as chemical weed control on weeds may be enhanced by applying N in bands (e.g. Rasmussen *et al.*, 1996; Rasmussen, 2002). As direct injection deposits the applied N in concentrated bands with minimal soil contact, immobilisation will be reduced and more of the applied N is available to plants (Petersen *et al.*, 2004), causing an increased crop N uptake (Tomar & Soper, 1987). In stands consisting of both a crop and a population of weeds, the two populations will compete for the plant-available mineral N, irrespective of the application method. Thus, direct injection has a potential for increasing the plant-availability of applied N as well as improving the crop's ability to out-compete weeds. Three experiments were conducted with the objective to examine the inter-species (crop:weed) and the intra-species (between crop rows) competition for applied N as affected by the horizontal and vertical distribution pattern.

## Materials and methods

The experiments were carried out at Foulumgård, Denmark (56° 30' N 9° 35' E). A <sup>15</sup>N-labelled nitrogen source was applied for determination of recovery of the applied nitrogen in crop and weeds. Treatments were applied to 30×40 cm micro-plots replicated for weekly samplings during the elongation phase of spring-sown cereals. Each treatment was sampled 6-8 times in duplicate or in triplicate. The recordings of <sup>15</sup>N recovery in crop and weeds were fitted using a sigmoid growth model, and an interpretation of the estimated and derived parameters was made. The recoveries are reported as percentages of applied N, and differences between treatments are given as %-point.

In Exp. I and II pig slurry was applied, and in these organic farming experiments the spring cereals competed with the natural weed population of 1800 plants m<sup>-2</sup> dominated by *Galeopsis tetrahit* L., *Polygonum persicaria* L. and *P. aviculare* L., *Lamium purpureum* L., *Veronica arvensis* L. and *Sinapis arvensis* L. (Petersen, 2003, 2005a). In Exp. III ammonium nitrate was injected in bands at tillering of spring wheat grown in weed free conditions for investigation of inter-row competition (Petersen, 2005b).

In Exp. I, before sowing of the crop, the slurry was incorporated, or injected, at either 5, 10 or 15 cm depth (Petersen, 2003). Two rows of spring barley were grown with 12 cm inter-row distance, and the injected bands were located in the middle between the rows. In Exp. II the two slurry distribution patterns were combined with either crop density (one or two rows for the incorporated slurry), or distance between the slurry band and the crop row (one row seeded at either 2 or 12 cm distance from the slurry band) (Petersen, 2005a). In Exp. III two rows of spring wheat were sown with 12 cm inter-row distance (Petersen, 2005b). After emergence mineral N was injected in bands dividing the inter-row space in either 2-10, 4-8 or 6-6 cm sections, resulting in a total of five crop row–fertilizer band distances.

## Results and discussion

In Exp. I and II maximum recovery of applied N by weeds was 12% (Petersen, 2003) and 26% (Petersen, 2005b), respectively, making weeds a significant competitor for the applied N. Positioning of band-applied nitrogen close to the crop row (Exp. II) decreased the recovery by weeds and caused a corresponding increase in N-recovery of the crop (Petersen, 2005b). Similarly, the band position in Exp. III affected the distribution of recovered N between the two competing crop rows (Petersen, 2005a). These observations of complementary recovery in competing populations are in contrast to those of Blackshaw et al. (2002).

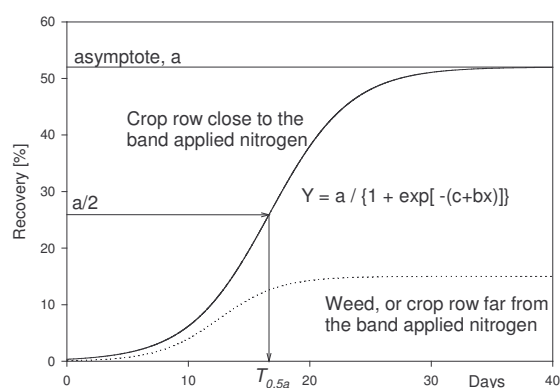


Figure 1. Typical plant recovery courses of applied nitrogen. Two important parameters of the sigmoid model are the asymptotic recovery,  $a$ , and the time until 50% of the final recovery is attained,  $T_{0.5a}$ .

In Exp. II, band-application increased the total plant recovery by 2.5%-point compared to broadcast applied nitrogen incorporated by harrowing (Petersen, 2005a). This might be due to a reduced immobilization due to reduced contact between applied N and the soil. Although more of the applied nitrogen becomes plant available, an even distribution of applied N may in some cases be preferred because of difficulties in obtaining the optimal geometric placing of nitrogen. For two competing crop rows in Exp. III the recovery decreased by 5.1%-point  $\text{cm}^{-1}$  (Petersen, 2005b), but for a single crop row competing with weeds in Exp. II the decrease was less than 2.2%-point  $\text{cm}^{-1}$  (Petersen, 2005a). In both cases the crop recovery course was delayed by 0.5 days per cm increase in crop row–fertilizer band distance. In comparison, the banding depth examined in Exp. I delayed the recovery course by 0.5-1.1 day per cm depth (Petersen, 2003). These figures underline that precise banding is required to obtain the advantage: nitrogen present in bands that facilitate a high N-recovery of the crop and simultaneously reduce the N-recovery of weeds. Thus, methods for N application may be part of a well-balanced weed control strategy. However, to obtain satisfactory weed control, N management needs to be supplemented with chemical or mechanical weed control.

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# The effect of NaCl and PEG (Polyethylene glycol 6000) induced water potentials and desiccation duration on the germination of *Glycyrrhiza glabra* L. rhizomes.

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## Introduction

Wild licorice (*Glycyrrhiza glabra* L.) is a rhizomatous perennial weed that spreads by seed and rhizome. Because of its deep and extended rhizomes, licorice competes severely with wheat, chickpea and alfalfa. An understanding of factors that influence the germination of licorice rhizome buds can help us find appropriate methods to successfully manage this weed. King & Oliver (1994) reported that the rate and percentage of germination increased as available soil water increased. Katembe et al. (1998) reported that higher concentrations of NaCl (-1.5 MPa) were more inhibitory to imbibition and germination of seeds of *Atriplex* species than iso-osmotic solutions of PEG. NaCl and PEG solutions with water potentials between -0.25 and -0.5 MPa did not significantly decrease the germination velocity of these species. At -1.5 and -1.4 MPa, both NaCl and PEG were inhibitory to germination of *Atriplex* seeds.

Hot and dry climates in many parts of the world can be used as a strategy to kill weed propagating organs like tubers, rhizomes and corms. No buds of *Cynodon dactylon* L. rhizomes survived when the rhizomes reached 50% of their original weight (Thomas, 1969). The regenerative ability of *Cynanchum laeve* root dried for 24 to 192 hours at 20°C (22% RH), and 30°C (30%RH) was completely lost (Soteres & Murray, 1982).

## Material and Methods

*NaCl and PEG experiments.* Wild licorice rhizomes were collected in 2003 from kamalshahr region, karaj. NaCl and PEG were used at five iso-osmotic concentrations corresponding to 0 (distilled water), -0.4, -0.8, -1.2 and -1.6 MPa. 10 Rhizome buds were placed in each Petri dish and then placed in a germinator under dark condition at 25°C. Numbers of buds germinated were counted at 5, 10, 15, 20 and 25 days and germination rate was calculated.

*Desiccation experiment.* This experiment was conducted in a complete randomized design with factorial arrangement of treatments and four replications. Factors included rhizome diameter at two levels (thin: 0.05-0.4 g/cm specific rhizome weight; thick: 0.4-1.6 g/cm specific rhizome weight) and desiccation duration (exposing rhizome fragments to sunlight over the soil surface) at six levels (0, 6, 8, 12, 24 and 48 hours). Percent germination of rhizome buds was measured by planting treated rhizomes in sand pots in a glasshouse.

## Results and Discussion

*NaCl and PEG experiments.* The effect of water potential concentration on germination was significant ( $P<0.01$ ) for both NaCl and PEG 6000. NaCl treatments from -0.4 to -1.2 MPa, drastically reduced germination and the rate of germination, whereas at -1.6 MPa no germination was observed (Fig. 1). Bajji et al. (2002) reported similar results for *Atriplex spp* seeds. Maximum rate of germination occurred at 10 to 15 days. No germination was observed in -1.2 and -1.6 MPa NaCl solutions. At all water potentials, NaCl was found to be more inhibitory to water uptake and germination than the corresponding PEG solutions. The same result was reported by Katembe et al. (1998).

*Desiccation experiment.* Desiccation durations significantly reduced rhizome bud germination (Fig. 2). However, rhizome specific weight did not have any significant effect on rhizome bud

germination. Germination decreased to 40% and 50% after a 24 hour desiccation period and no bud survived after 48 and 36 hours desiccation periods for thick and thin rhizomes, respectively. Complete inhibition of germination occurred when rhizomes, as a result of desiccation, were reduced to 42 and 70 percent of their original weight, for thin and thick rhizomes, respectively.

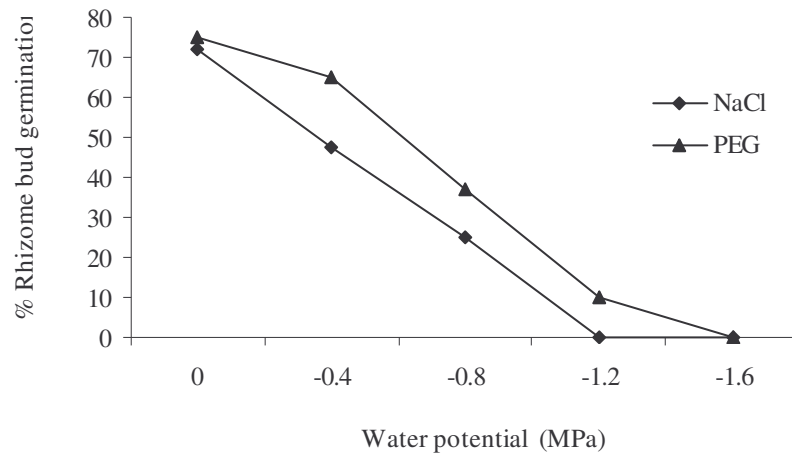


Figure 1. Comparison of rhizome bud germination in different NaCl and PEG induced water potentials.

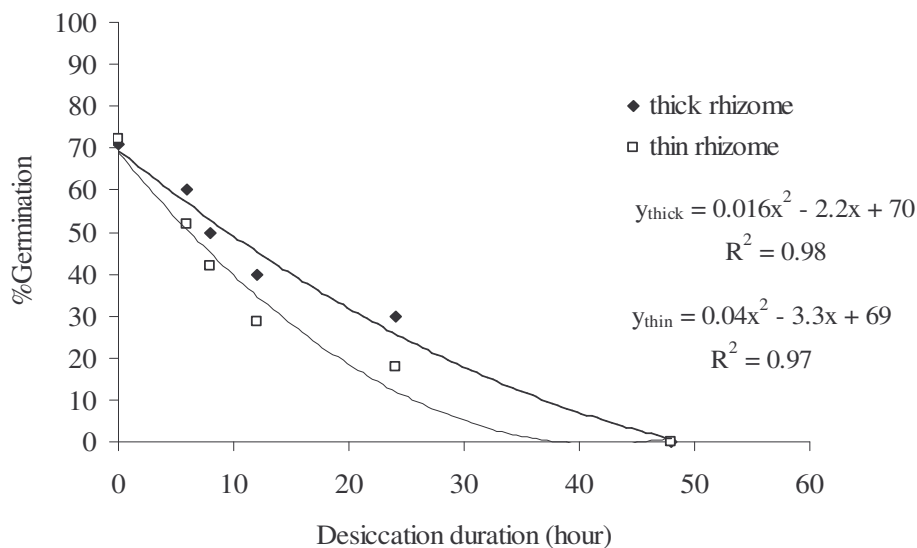


Figure 2. The effect of desiccation duration on germination of buds of thin and thick rhizomes.

This experiment suggests that rhizome sections exposed on the soil surface for a relatively short period of time would essentially lose their ability to produce shoots and develop into new plants.

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# Corn yield loss due to weeds as influenced by nitrogen and herbicide application

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## Introduction

Weeds compete with crops for water, light and nutrients. Yield loss is often closely related to weed infestation level (Hons and Saladino, 1995; Kapusta et al., 1994; Norris 1999; Swanton et al., 1999a, 1999b). Many factors such as nitrogen and herbicide application, because of their effects on weed density and weed community, have an influence on weed-crop interactions. The objective of this project was to determine the reduction in corn yield due to the presence of weeds as influenced by nitrogen and herbicide application. Attaining these goals will help us to make a better decision towards weed management in corn production.

## Materials and methods

This experiment was conducted in 2001 at Lods Agronomy Research Center, Macdonald campus, McGill University in Montreal. The type of design was a split plot with 6 replications, in which herbicides (4 levels) and nitrogen (3 levels) were allocated to main plots and subplots, respectively. Corn seeds were planted at 4 cm depth in May 10. The size of the plots was 10 × 10 m with rows 75 cm apart. Corn population was based on 76,000 plants ha<sup>-1</sup>. Nitrogen at rates of 60, 120, and 250 kg ha<sup>-1</sup> were split and applied at planting time and on July 3. Nicosulfuron (Ultim, 9 g ha<sup>-1</sup>) and mineral oil (Agral 90, 25% v/v) were used to control grassy weeds. Dicamba (Banvel II, 1.2 L ha<sup>-1</sup>) was used for control of broadleaved weeds. Mixture of Ultim plus Agral 90, at above rates and Banvel II (0.6 L ha<sup>-1</sup>) were used to control both broadleaf and grassy weed species. Weedy check with no herbicide application was also present. Determination of corn plant dry weight was done on June 25, 19 days after herbicide application and on July 25, 3 weeks after top dressing of nitrogen. Biological yield and seed yield of corn were determined at the end of the growing season. ANOVA was performed on corn data by using General Linear Model (GLM) procedure of SAS. Additionally, means were compared by using the Least Significant Difference of means (LSD;  $p = 5\%$ ).

## Result and discussion

Nitrogen rate had a significant effect on corn biomass at 25 June and at harvest time (Figure 1). At the intermediate sampling on 27 July no significant differences were observed. Increasing nitrogen rate from 120 to 250 kg ha<sup>-1</sup> increased corn yield significantly. Comparing experimental treatments with calibration plots indicated that depriving corn from nitrogen resulted in pale green leaves of corn. Visual rating and biomass determination (data not shown) showed that the nitrogen application rate did not have any effect on weeds.

Significant differences among herbicide treatments were observed both on June 25 and on July 27 (Figure 2). The weedy check and the use of dicamba, the herbicide to control broadleaved weeds, produced the lowest corn dry weight. Surprisingly, the highest level of corn biomass on June 25 was observed in plots with control against grassy weeds only. The same trend was observed for biological yield and seed yield at harvest. Control of grassy weeds only increased corn yield even more than total weed control.

Treatment of plots with herbicides against broadleaved weeds only, resulted in enormous grassy weed infestation levels and this explains why corn biomass and seed yield were not significantly different from that of the weedy check. It is also noteworthy that controlling grasses with nicosulfuron and Agral had an even better result than total weed control. In

conclusion we might say that under Montreal conditions, corn responds effectively to herbicides and nitrogen rates. There was no interaction between nitrogen and herbicides.

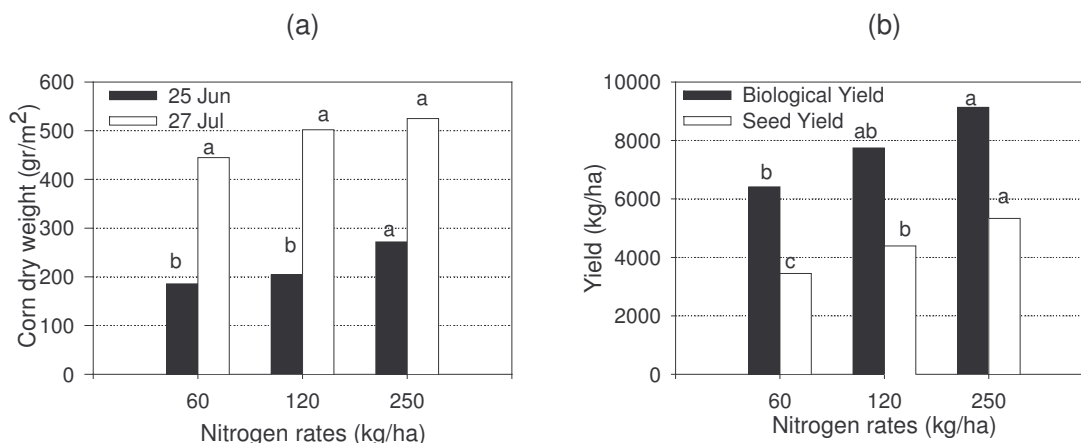


Figure 1. Effect of nitrogen rate on (a) corn dry matter at June 25 and July 27 and (b) biological yield and yield of corn at harvesting time. Values in each series not marked with the same letter are significantly different (LSD 5%).

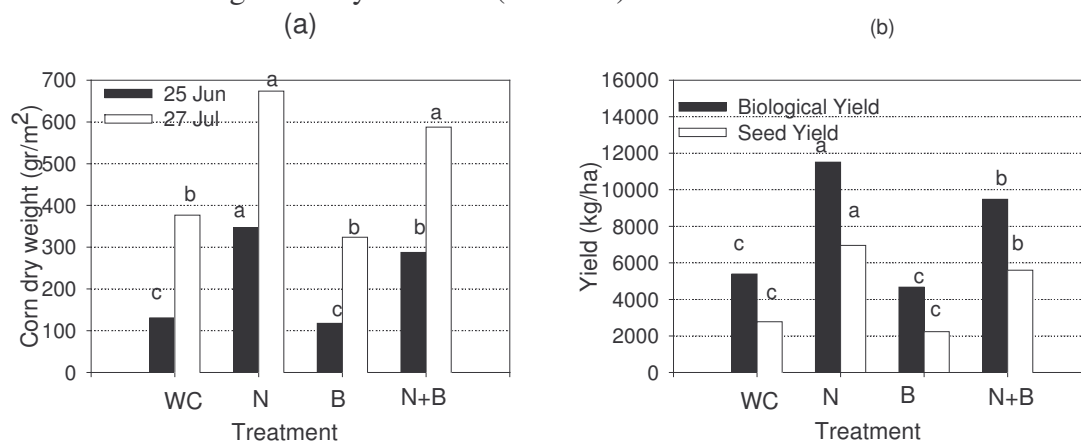


Figure 2. Effect of herbicides on (a) corn dry matter at June 25 and July 27 and (b) biological yield and yield of corn at harvesting time. WC: Weedy Check, N: Nicosulfuron, and B: Banvel. Values in each series not marked with same letter are significantly different (LSD 5%).

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# Wild mustard (*Sinapis arvensis*) seed production in response to amount and timing of nitrogen application

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## Introduction

Despite management efforts, many weeds escape control and produce seeds that replenish seedbanks and increase the potential future weed infestation (Kegode et al., 1999). High seedbank populations can ultimately lead to high weed densities. It may require several years of intensive management to minimize the problems associated with these densities (Kegode et al., 1999). Many cultural practices such as crop density (Wilson et al., 1995), crop rotation, tillage (Kegode et al., 1999), herbicide rate (Wille et al., 1998), spatial arrangement (Norris et al., 2001) and other management inputs may affect weed biomass and ultimately weed seed production. Also the intensity of these factors on weed seed production is related to weed – crop interactions (Van Acker et al., 1997). The objective of this project was to determine seed production of wild mustard (*Sinapis arvensis*) in response to amount and timing of nitrogen application.

## Materials and methods

This experiment was conducted in 2001 at the Research Farm of Mashhad College of Agriculture. A split plot design with three replications was used with a combination of weed density (0, 8, 16, and 32 plant m<sup>-2</sup>) and nitrogen fertilization rate (low = 100, optimum = 150, and high = 225 kg ha<sup>-1</sup>) as main plot. The sub plot included a nitrogen split pattern (P<sub>1</sub>=1/3 at planting time + 2/3 at tillering, P<sub>2</sub>= 1/3 at planting time + 1/3 at tillering + 1/3 at stem elongation). ANOVA were performed on data by using the General Linear Model (GLM) procedure of SAS. Additionally, regression analysis with regression models was used for data analysis. Seed production in dependence of density was fitted to a two-parameter hyperbolic rectangular function (Cousens et al., 1995) and an allometric function was used for determination of minimum biomass for seed production in wild mustard.

## Result and discussion

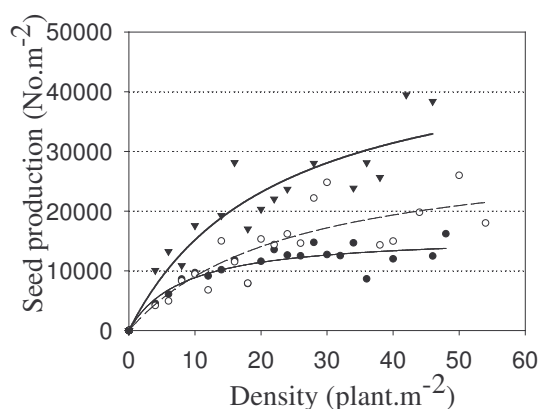


Figure 1. Relationship between density of wild mustard and its seed production per area in dependence of N-fertilization rate. Observed data were fitted to a two parameter hyperbolic rectangular function ( $S_w = PN_w/(1+PN_w/q)$ ). 100 (●), 150(○), 225(▼) kg N ha<sup>-1</sup>

According to the results, seed production of wild mustard increased with increasing wild mustard density and nitrogen rates, due to high wild mustard biomass production. Estimation of wild mustard seed production was 161, 311, and 488 million seeds ha<sup>-1</sup> in low, optimum and high nitrogen rates, respectively (Figure 1 & Table 1). The minimum biomass for seed production was determined 1.12 g per plant (Figure 2).

Table 1. Estimated parameter values of the rectangular hyperbolic function describing seed production of wild mustard at various nitrogen levels.

Nitrogen (kg ha <sup>-1</sup> )	P (No plant <sup>-1</sup> )	q (No m <sup>-2</sup> )	R <sup>2</sup>
100	1967 (421.10) *	16108 (1283.40)	0.91
150	1282 (283.75)	31105 (5700/25)	0.86
225	2200 (429.77)	48862 (7686.34)	0.91

\*Values in parenthesis are standard error (SE).

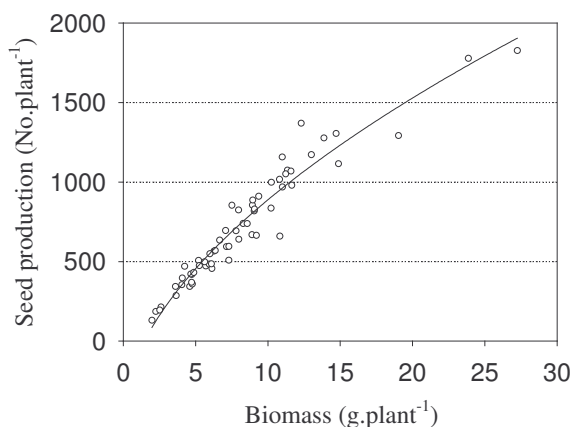


Figure 2. Relationship between wild mustard biomass per plant and seed production per plant fitted to an allometric function. ( $S = CW^k - a$ ).

Whereas density and nitrogen rates had a significant effect on wild mustard fecundity, nitrogen split pattern showed no significant effect on wild mustard seed production (Data not shown).

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# Effect of burial depth on the width of leaflets of *Oxalis latifolia*

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## Introduction

*Oxalis latifolia*, a weed with a world wide distribution, is highly affected by depth of burial. Marshall (1987) reported that the weed is able to grow from a depth of up to 30 cm. Esler (1962) observed that bulbs buried up to a depth of 25 cm were able to develop leaves, but rarely were leaves formed on bulbs that were buried at a greater depth and never from over 60 cm. Esler also found that depth increased the mortality of the weed, only 20 % of 0.66 cm diameter bulbs buried at 25 cm survived. Finally, this author also observed that leaves from deep buried bulbs were wider than those of shallow buried bulbs. López & Royo (2001a) compared bulbs buried at 1 and 12 cm and found that the latter developed wider leaves, that their growth was delayed, mortality increased, productivity decreased and that they never developed contractile roots. Finally, Royo-Esnal & López (2004) observed that a burial depth of 34 cm was able to kill almost any bulb, irrespective of size. In this study the size of the leaflets from common and cornwall bulbs buried to different depths was measured.

## Materials and methods

Five rows were disposed with six bulbs in each row. The bulbs were planted inside a tube, at 10 cm depth but without soil over them. There were tubes of 4.5 cm in diameter and 15, 20, 25, 35, 40 and 50 cm in length in each row covering the bulbs so that, for reaching the light, they would have to elongate their petioles as far as the length of the tube. An additional bulb buried at 1 cm depth and without tube was added to each row. In two of the rows common form bulbs were planted, whereas in the other three cornwall form bulbs were planted. Planting was done on 14 April 2001 and on 29 July, when the weed is in its most prolific growth stage (Royo, 2004). One leaflet from each bulb's three largest leaves was measured. In this way three measurements were obtained from each bulb.

## Results and discussion

The length of each petiole hardly surpassed the length of the tube with more than 2 cm. Table 1 shows the mean values of leaflet width in dependence of tube length for each of the two *Oxalis latifolia* forms, as well as the statistical result of the comparison between both forms. These values are also represented in Figure 1. For both forms, leaflet width increased until a petiole length of 35 cm. Bulbs of common form usually developed wider leaves than bulbs of cornwall form.

Table 1. Mean leaflet width in cm (followed by standard error) of bulbs of *Oxalis latifolia* for both common and cornwall form, as affected by tube length

Form	depth (cm)	Tube length (cm)						Wilcoxon
	1	15	20	25	35	40	50	
common	4.2 (0.36)	5.5 (0.12)	5.3 (0.32)	7.1 (0.32)	7.9 (0.16)	6.9 (0.30)	6.7 (0.56)	**
cornwall	3.9 (0.11)	4.8 (0.14)	5.3 (0.16)	5.4 (0.11)	5.8 (0.20)	5.4 (0.21)	5.0 (0.30)	

As leaflet width increased until a petiole length of 35 cm a linear regression between leaflet width and petiole length was made for bulbs of both forms that were put into tubes shorter than 40 cm. Results are represented in Figure 2. A significant and a very significant regression between petiole length and leaflet width was obtained for bulbs of common form and cornwall form, respectively.

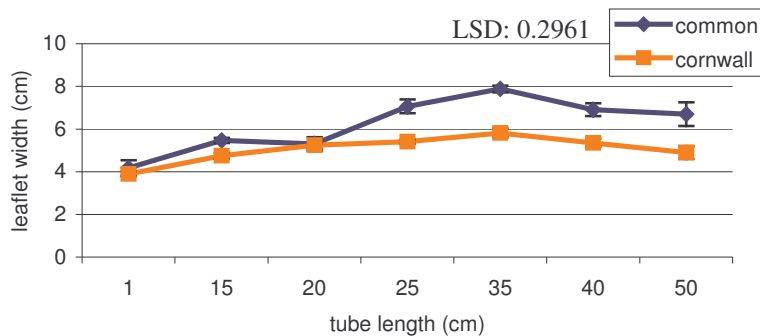


Figure 1. Relation between leaflet width and tube length.

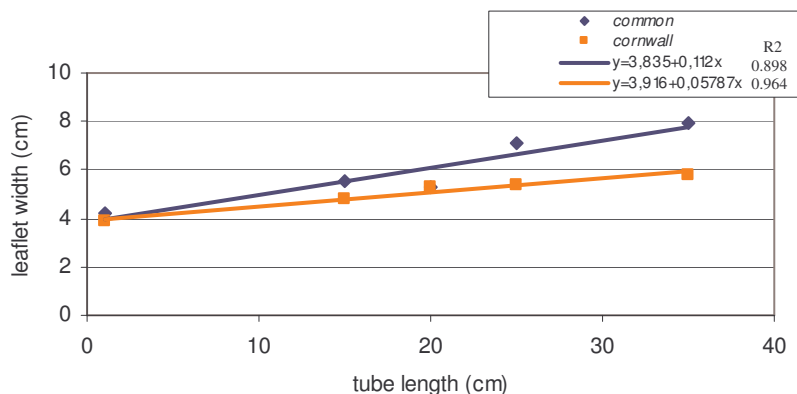


Figure 2. Regression between petiole width/tube length for tubes with a length up to 35 cm.

López & Royo (2001b) showed that *Oxalis latifolia*'s petiole grow from an intercalary meristem. The activity of this meristem lengthens the petiole, but also gives embryonic cells to the leaflets. The embryonic cells will not differentiate until the emergence of the leaf. The more time it lasts before leaves are exposed to the light, the greater the amount of cells in their leaflets, and consequently, the wider they will be when mature and unfolded. That explains why deep buried bulbs have wider leaves than shallow ones.

From a practical point of view, it is likely that wider leaves will absorb herbicides more effectively because of the greater area exposed. Chawdhry & Sagar (1974) and Parker (1966) pointed at the importance of a first defoliation of the weed before it develops four leaves. Due to an effective absorption of the chemical, wider leaves may help to use minimal doses for this first defoliation. So a burial depth of 30-35 cm will be beneficial in two ways in controlling *Oxalis latifolia*: it will kill many small bulbs as Royo-Esnal & López (2004) showed, and the surviving bulbs will develop wider leaves capable of a more effective herbicide absorption.

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## Spurge – a new dangerous weed in Siberia

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Spurge (*Euphorbia waldsteinii (virgata)*(Sojak) Czer.) is a species widely spread in Siberia. It can be often met on meadows, road sides, near electric mains and in other similar areas.

Spurge weed has become frequent in last decades of the XX century. At first it became abundant on fields in the steppe zone and afterwards it spread for several years all over the region. It grows on fields of Altai and Krasnoyarsk territories, Novosibirsk, Omsk, Kemerovo, Tyumen' regions. Total area of weedy land in Siberia & Kazakhstan is about 10 million ha.

In compliance with the type of the root system structure it is among dicotyledonous perennial offset weeds. Root system shows a good mechanical strength, what gave the name of the plant. Position of the horizontal part of root depends on inhabitancy. On meadows it goes 10-20 cm deep in the soil, in fallow field 20-40 cm with some offsets up to the depth of 60 cm. Deepening of the root system is influenced by its cutting by soil machinery. At the place of cutting additional short horizontal sprouts are produced.

Renovation buds are located both on vertical and horizontal parts of the root (Fig.1).

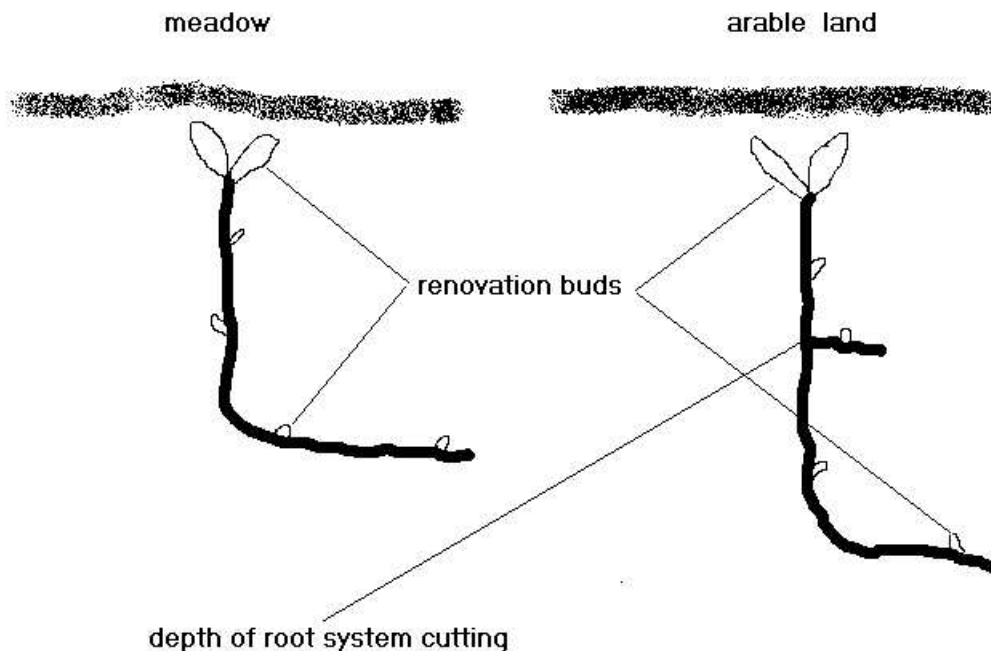


Figure 1. The structure of spurge root system at hibernation stage

In spring time regrowing starts from upper buds. These buds begin to grow very early just after thawing of the soil. Plants develop fast and they often can produce seed to date of chemical treatment of cereals. At this stage spurge is already resistant to herbicides.

Spurge can severely decrease cereal yield (Fig.2). At the density 10 sprouts per 1 m<sup>2</sup> losses reach ¾ of the yield. Development of crop protection measures against the weed in Siberia is a serious challenge.

It was established that the part of vertical root, which was cut in autumn by soil cultivators, died during winter. Sprouts from deep located renovation buds appear significantly later in the spring. Young sprouts of the spurge are sensitive to the majority of herbicides applied in cereals: 2,4-D derivatives, dicamba, sulfonyleurea family.

An integrated spurge management have been developed in Siberia as follows: 1) infested fields must be deeply tilled in autumn so that the truncated part of the root system perish during winter; 2) later on common post-emergence herbicides against dicotyledonous weeds should be applied in wheat. A combination of agronomic and chemical approaches can totally destroy spurge in two years.

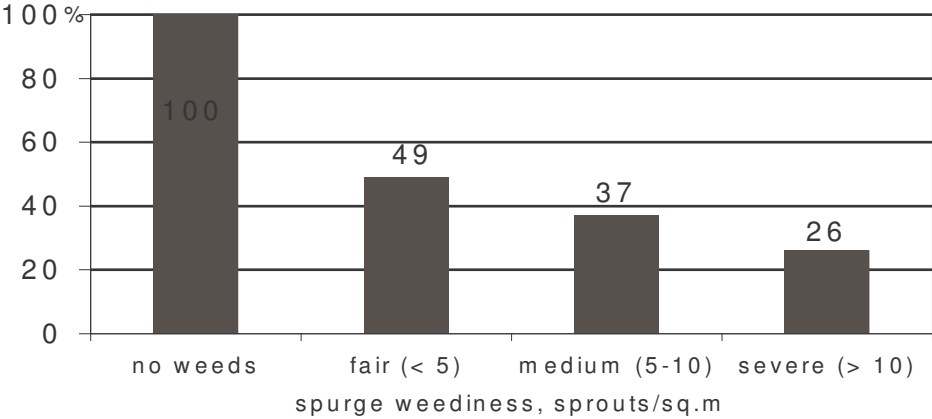


Figure 2. The effect of spurge weediness on the yield of spring wheat (% to clean crop)

## A study on wild oat populations in Iran

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### Introduction

Wild oats (*Avena spp.*) are the most abundant and problematic annual grass weeds in wheat in Iran. Both winter annual *A. ludoviciana* and summer annual *A. fatua* frequently occur in cereal crops. Seeds of this species can remain dormant in the soil for several years, so a long-term strategy is required for its control (Tottman & Wilson 1990). Dormancy is caused by both coat-imposed and embryo-imposed dormancy. Also, wild oat seeds normally require after-ripening under warm-dry conditions (Foley, 2000). Genetic variability has previously been demonstrated in wild oat populations (Armstrang & Adkins 1998). Also, variation in tolerance and resistance to herbicides has been found in several biotypes (Seefeldt et al., 1998; Andrews et al., 1998). Widespread variation in morphotypes has been observed, particularly in relation to seed color and hairiness, but this variation has not been documented (Medd, 1996). Information on seed germination and other aspects of biology such as competitive ability, phenology of growth, flowering and seed set (Andersson et al., 2001) are attributes contributing to the success of wild oats and are therefore of great importance for the development of weed management strategies. The objective of this study was to survey wild oat populations, identify the species and variation among and within species with respect to factors like germinability and seed production.

### Material and methods

A scouting was done to survey and collect wild oat seeds in wheat fields in different regions of Iran during 2003. During 2004, a field experiment was conducted in Malekshahi, Ilam, Iran. Seed samples of wild oats populations including *A. fatua* and *A. ludoviciana* were sown in a Randomized Complete Block Design with 3 replicates. Morphological differences, fraction germination, number of tillers, time to anthesis (DC 60) and ripening (DC 94) (Zadoks et al., 1974), number of spikelets, and seeds per plant were recorded.

### Results and Discussion

A variation in morphotypes was observed. According to the seed coat color (seed, lemma and palea), two types of *A. fatua* were discerned (brown and grey/black) and three types of *A. ludoviciana* (brown, yellow and grey/black). The regional survey showed that 69% of the samples collected and identified were *A. ludoviciana* and 31% were *A. fatua*, indicating that *A. ludoviciana* is apparently the most common wild oat species in Iran. One explanation is that winter wheat is usually grown in most of the area, which is favorable for *A. ludoviciana* infestation. *A. ludoviciana* responds to cooler temperatures and germinates in winter and early spring (Medd 1996). As it is frost tolerant, it is typically a weed of autumn-sown crops (Tottman & Wilson 1990). *A. fatua* tends to germinate from autumn through to spring.

In the field experiment populations were sown on 10 November. Two weeks after sowing most of the seedlings had emerged. Germination varied greatly between species and populations within species, resulting in a range from 3 - 73 %. Of the *A. fatua* seeds, on average 47.5 and 10% of the seeds germinated for the grey/black and brown types, respectively. Of the brown, grey/black, and yellow types of *A. ludoviciana* 44.7, 40 and 28.3% germinated, respectively. Individual plants produced between 66.7-1204 seeds in the first year. Among the *A. ludoviciana* populations significant differences in number of tillers, time to anthesis and ripening, number of spikelets and

seeds per plant were observed. In contrast, populations of *A. fatua* only differed in germination percentage and time to reach anthesis (Table 1).

Table 1. Mean germination percentage, number of tillers, time to anthesis and ripening, number of spikelets, and seeds per plant for populations of *Avena* species collected from different parts of Iran

Populations	Germination (%)	Tiller (No.)	Anthesis (Days)	Ripening (Days)	Spikelets (No.)	Seed (No.)
<i>Avena loduviciana</i>						
1	56.7 <sup>ab</sup>	14.0 <sup>a</sup>	168.7 <sup>b</sup>	191.3 <sup>c</sup>	39.7 <sup>ab</sup>	1124 <sup>a</sup>
2	56.7 <sup>ab</sup>	11.7 <sup>ab</sup>	169.7 <sup>b</sup>	191.0 <sup>c</sup>	32.0 <sup>abc</sup>	745.3 <sup>ab</sup>
3	60.0 <sup>ab</sup>	13.7 <sup>a</sup>	169.0 <sup>b</sup>	190.7 <sup>c</sup>	41.0 <sup>ab</sup>	1204 <sup>a</sup>
4	40.0 <sup>abcd</sup>	10.3 <sup>ab</sup>	168.7 <sup>b</sup>	192.7 <sup>c</sup>	17.0 <sup>cd</sup>	367.3 <sup>abc</sup>
5	23.3 <sup>cdef</sup>	6.7 <sup>abc</sup>	170.0 <sup>b</sup>	195.3 <sup>abc</sup>	23.0 <sup>bcd</sup>	365.0 <sup>abc</sup>
6	46.7 <sup>abc</sup>	5.3 <sup>bc</sup>	170.0 <sup>b</sup>	194.7 <sup>abc</sup>	9.7 <sup>d</sup>	161.3 <sup>c</sup>
7	70.0 <sup>a</sup>	7.7 <sup>abc</sup>	168.7 <sup>b</sup>	191.3 <sup>c</sup>	22.3 <sup>bcd</sup>	347.3 <sup>abc</sup>
8	16.7 <sup>edf</sup>	7.0 <sup>abc</sup>	171.0 <sup>b</sup>	192.0 <sup>c</sup>	15.7 <sup>cd</sup>	324.0 <sup>bc</sup>
9	10.0 <sup>ef</sup>	4.3 <sup>bc</sup>	179.7 <sup>a</sup>	196.7 <sup>abc</sup>	11.0 <sup>d</sup>	150.0 <sup>c</sup>
10	3.30 <sup>f</sup>	1.7 <sup>c</sup>	180.0 <sup>a</sup>	201.0 <sup>a</sup>	6.7 <sup>d</sup>	66.70 <sup>c</sup>
11	33.3 <sup>bced</sup>	8.0 <sup>abc</sup>	184.7 <sup>a</sup>	199.0 <sup>ab</sup>	48.7 <sup>a</sup>	785.3 <sup>ab</sup>
<i>Avena fatua</i>						
1	43.3 <sup>b</sup>	8.3 <sup>a</sup>	177.3 <sup>b</sup>	198.0 <sup>a</sup>	41.3 <sup>a</sup>	671.3 <sup>a</sup>
2	73.3 <sup>a</sup>	5.0 <sup>a</sup>	172.3 <sup>c</sup>	198.3 <sup>a</sup>	24.3 <sup>a</sup>	248.0 <sup>a</sup>
3	70.0 <sup>a</sup>	7.3 <sup>a</sup>	180.0 <sup>b</sup>	200.3 <sup>a</sup>	32.7 <sup>a</sup>	472.7 <sup>a</sup>
4	10.0 <sup>c</sup>	5.0 <sup>a</sup>	192.0 <sup>a</sup>	202.0 <sup>a</sup>	21.7 <sup>a</sup>	326.7 <sup>a</sup>
5	6.70 <sup>c</sup>	4.3 <sup>a</sup>	192.0 <sup>a</sup>	200.0 <sup>a</sup>	27.3 <sup>a</sup>	462.0 <sup>a</sup>
Species	**	**	**	**	ns	**

Means with the same letter are not significantly different at the 0.05 level as determined by Duncan's multiple range test; ns, not significant; \*\* significant at p<0.05

Furthermore, the survey showed that wheat monoculture, frequent use of herbicides targeted at broad leaved weed species, and growing of modern dwarf and semi dwarf wheat varieties instead of local varieties seem to be major factors responsible for infestation of wild oats in Iran. More investigations on biology and ecology of wild oat populations are required to be able to design adequate weed management strategies.

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## The critical period of weed control in soybean (*Glycine max* (L.) Merrill.) grown as a second crop in Adana

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### Introduction

In Adana, multiple cropping by growing two consecutive crops is successfully applied. If soybean is grown as a second crop, it is often highly affected by weed infestation, especially in the earlier periods of its growth (Arioglu, 1999). Depending on the variety and intensity of weeds, 10 to 86 % yield loss was detected in soybean due to the absence of weed control (Panneerselvam & Lourduraj 2000). Weed control in soybean is done by hoeing 2-3 times during the vegetation period or by using chemicals, such as applying herbicides during the in pre-sowing or pre-emergence stage (Tepe, 1997). In 2003, field trials were conducted to determine the critical period of weed control in soybean grown as a second crop.

### Materials and methods

Field trials were performed according to a randomized split plot trial design in four replications. Plot size was 16 m<sup>2</sup> and contained four crop rows of variety Nazlican, a middle early soybean variety. Treatments consisted of different weed-free periods (till 10, 20, 30, 40, 50, 60 and 70 days after crop emergence and always weed-free as control) and different weed-infested periods (till 10, 20, 30, 40, 50, 60, and 70 days after crop emergence and always weed-infested as control). To obtain weed-free plots, hoeing of weeds was performed in the earlier growing period, while collecting the weeds by hand was applied in the late growing period when the plants closed the distances between rows. Every ten days, the composition of the community and weed density was determined by random sampling, using four areas of 50 cm<sup>2</sup> per plot. Soybean yield was determined by harvesting the middle two rows of each plot. Analysis of variance combined with Duncan's multiple comparison test was applied to the results. The trial will be repeated next year.

### Results and discussion

In the vegetation period of soybean the following weed species were observed: *Xanthium strumarium* L., *Convolvulus arvensis* L., *Amaranthus retroflexus* L., *Echinochloa colonum* (L.) Link, *Cyperus rotundus* L., *Portulaca oleracea* L., *Setaria verticillata* (L.) P.B., *Chenopodium album* L., *Echinochloa crus-galli* (L.) Pal. Beauv., *Euphorbia serpens* Kunth., and *Digitaria sanguinalis* (L.) Scop. Total weed number right after emergence ranged from 19 to 65 plants m<sup>-2</sup> in each plot. At the last observation date, at 70 days after emergence, this number varied from 0.75 to 20 plants m<sup>-2</sup>. The initially high weed number had decreased due to the both hoeing practices and an increased soybean canopy in the later stages of crop development.

Soybean yield in dependence of the duration of the weed-free infested period is presented in Figure 1. Leaving the plots uncontrolled until 10 days after emergence of the crop did not result in a significant yield loss. Maintaining the weed-infestation for a longer period did result in serious yield losses. Controlling the weeds did result in an increase in yield. The data suggest that weed control should be continued until 30 days after emergence. Weed control after that period did not result in any yield advantage. The period between the moment yield

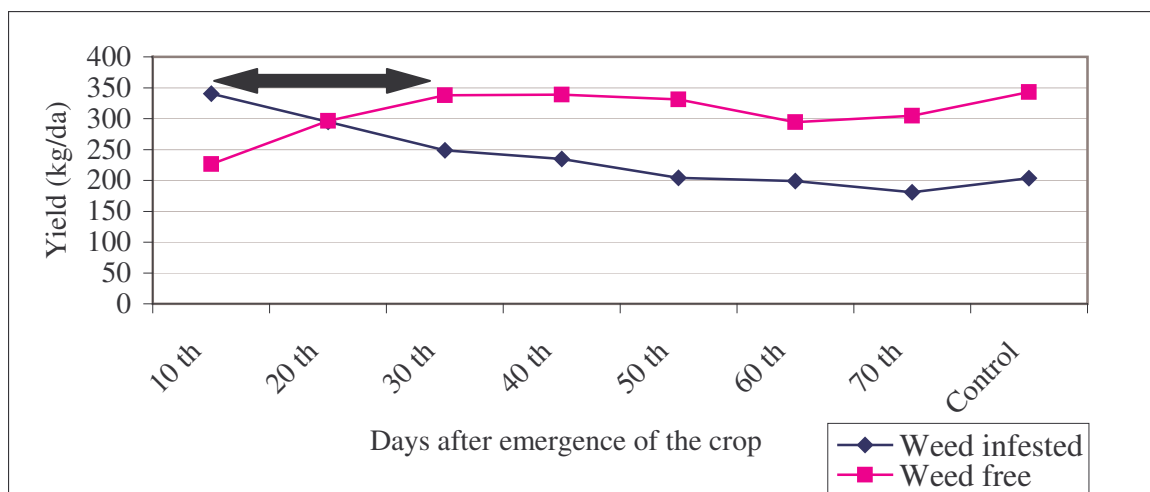


Figure 1. Soybean yield for different weed-free and different weed-infested periods. The critical period for weed control is indicated by the double-  
arrowed line.

started to decrease in the weed- infested plots and the point where the increase in yield in the weed-free plots reached the maximum yield level was taken as the critical period of weed control. It is in this time period that weed control has to be performed (Koch and Kunish, 1989). In this trial, the critical period of weed control was determined as the period between 10 and 30 days after emergence of soybean.

For soybean different critical periods of weed control have been reported. A critical period between the 1<sup>st</sup> and the 12<sup>th</sup> week after emergence in the rainy season and between the 1<sup>st</sup> and the 9<sup>th</sup> week in the dry season was reported by Suwanagul and Duangporn-Suwanagul (1998). Controlling the weeds from the 30<sup>th</sup> up to the 45<sup>th</sup> day after planting was suggested as the critical period by Chhokar and Balyan (1999). They observed a 74 % increase in yield if weeds were controlled until the 45<sup>th</sup> day. Varshney (1989) suggested a critical period of weed control for soybean till the 40<sup>th</sup> day after planting. Variability of critical periods in these studies resulted from differences in soybean variety, in soil type, weed community and weed infestation level. To obtain a more reliable critical period the current experiment will be repeated in the coming year.

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# Autumn growth of *Elymus repens*, *Cirsium arvense* and *Sonchus arvensis* as affected by climate change at high latitudes

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## Introduction

The global temperature is expected to increase by 1.4-5.8°C and the atmospheric CO<sub>2</sub> to above 500 ppm during the next 100 years (IPCC 2001). A much more variable climate during autumn and winter with more precipitation/clouds is expected in Norway (RegClim). The effects of these changes on winter survival of weeds, plant diseases, winter wheat and grasses, are the subject of a research programme started in 2004. The present study is part of that programme with the objective of examining the effects of temperature, CO<sub>2</sub> and irradiation on autumn growth of a northern (63°N) and a southern (59°N) ecotype of *Elymus repens* (L.) Gould, *Cirsium arvense* (L.) Scop. and *Sonchus arvensis* L. A change in the growth pattern of these perennial weeds in autumn may influence the infestation of the following crops and the need for control measures.

## Materials and methods

Pieces of rhizomes (*E. repens*, 3 nodes) and roots (*C. arvense*, *S. arvensis*, 5 cm) were planted in 10 L pots on 26 May 2004. Two pieces were planted per pot, in a mixture of 50% sand and 50% peat amended with balanced nutrients. One month after planting the pots were thinned to one piece/pot. The plants were grown outdoors at 58°N (Særheim) until the start of the experiment. Six days before the start (27 August 2004) plants were cut to about 20 cm height to simulate the effects of cereal harvesting. From 2 September to 1 November 2004 plants were exposed to six different environments; in field chambers with/without increases of temperature (+2-2.5°C compared to field chambers without heating) and CO<sub>2</sub> (ca. 370 ppm vs. ca. 550 ppm) and under outdoor conditions with/without shading (30% light reduction). There was a gradual decrease of temperature outdoors during the experimental period from about 15°C to 8°C. The field chambers without heating had about 0.3°C higher temperatures than outdoors. There were four replicates in the experiment. Every fortnight during the experimental period dry weight of above ground and below ground plant parts, root/rhizome length, leaf area and the developmental stage of the plants were assessed, and soil samples were taken to determine the nutrient status. To achieve nutrient conditions comparable to what is found in autumn stubble fields, a balanced nutrient solution was applied on 3 September and 4 October corresponding to 45 and 20 kg N/ha, respectively. Water was given as needed. The assessments were analysed by analysis of variance and if effects were significant, contrasts were used to detect differences between treatment means (CO<sub>2</sub>, temperature, CO<sub>2</sub> x temperature, chamber vs. outdoors, shading).

## Results and discussion

After cutting, *E. repens* regrew with new shoots and more leaf area until the end of the experiment (Fig. 1) while *S. arvensis* stopped growing and withered shortly after start of the experiment. Therefore, no results of this species are presented. *C. arvense* was in an intermediate position with a small regrowth on the existing shoots, but the overall leaf area decreased during the experimental period (Fig. 1). *S. arvensis* is known to become dormant in late summer/autumn (Fykse 1974, Fogelfors et al. 2003). For *C. arvense* some degree of dormancy is found (Fykse 1974), while *E. repens* is not becoming dormant and can grow as soon as the climatic conditions for growth are present (Håkansson 1974, Fogelfors et al. 2003). These species characteristics were thus confirmed by this study. During the experiment

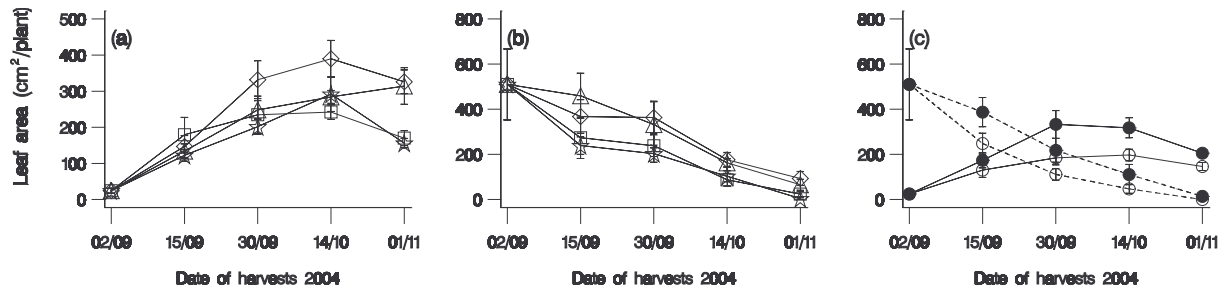


Figure. 1. Effect of temperature and CO<sub>2</sub> in field chambers on leaf area of (a) *E. repens* and (b) *C. arvensis* (☆=control, □= +CO<sub>2</sub>, △=+temperature, ◇=+CO<sub>2</sub> and temperature) and (c) effect of 30% shading outdoors (○=control, ●=+shading) on leaf area of *E. repens* (—) and *C. arvensis* (---). Values are averages over ecotypes and replicates (±SEM, n=8).

The below ground dry weight of *E. repens* increased, the root dry weight of *C. arvensis* and *S. arvensis* was a relatively constant, and the ratio between dry weight of above vs. below ground plant parts ('shoot/root-ratio') of all species decreased.

The southern ecotype of *S. arvensis* and *C. arvensis* developed more roots (dry weight and length) than the northern ecotype with a corresponding lower shoot/root-ratio. For the southern ecotype of *C. arvensis* also more leaf area was produced. The northern ecotype of *E. repens* developed more leaf area and shoot and root dry weight. Maybe the northern ecotype of *E. repens* was better adapted to the coastal climate, whereas the southern ecotypes of the other two species were better adapted to the day length at Særheim.

A raised temperature as well as increased shading in general increased the leaf area and the above ground shoot dry weight. Except for an effect of increased CO<sub>2</sub> on the root dry weight at one harvest (30 September), no effect of the three climatic factors on dry weight of below ground plant parts, length of rhizomes or propagating roots and shoot/root-ratio, was detected.

Looking at each species, only leaf area of *E. repens* and *C. arvensis* were significantly affected by increased temperature and shading (Fig. 1). Leaf area of *E. repens* was also affected by increased CO<sub>2</sub> (Fig. 1a). In principle the northern and southern ecotypes did not react differently on climate and therefore the results are presented as averages over ecotypes. *S. arvensis* did not have any regrowth. However, a tendency of a small delay in the cessation of *S. arvensis* with higher temperatures was observed.

Since the roots/rhizomes were little affected by the climate in autumn, the infestation next year may not be influenced either. However, large variation in plant size was observed and this might have hidden some of the effects.

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## Use of allelopathic crops from *Brassicaceae* family to control weeds

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### Introduction

*Brassicaceae* species have been known for their allelopathic effects on weeds (Rice, 1995). In Turkey, vegetable *Brassicaceae* crops such as radish (*Raphanus* spp.) and turnip (*Brassica* spp.) species are grown as well as rapeseed (*Brassica napus* var. *oleifera*). In a limited area, garden radish (*Raphanus sativus*) has been used by farmers when the population of johnsongrass (*Sorghum halepense*) increased. It has been reported in industrial crops such as cotton and maize, wheat fields, vegetables, fruit plantations and waste areas in Turkey (Ulug *et al.*, 1993; Zel, 1994; Uludag & Uremis, 2000). Despite earlier studies (Koseli, 1991), use of allelopathic crops has not expanded or improved. The main reasons for that are lack of data and limited radish marketing. In order to start a successful extension activity, possible usage of radish in cropping systems should be known. If different crops can be introduced for allelopathic purpose, farmers can get benefit of marketing them too. It is believed that this will help to expand the use of allelopathic crops. In addition, arousing weed control problems, an expanding demand for organic crops, and increasing public concerns on environmental issues require alternative farming systems which are less pesticide dependent or based on naturally occurring compounds (Singh *et al.*, 2003; Waller, 2004). The aim of our study was to find out how to use radish in cropping systems and to explore the possibilities of using other *Brassicaceae* crops to control johnsongrass.

### Materials and Methods

In order to improve the use of radish, field experiments were conducted in 2002 and 2003. In these experiments radish was grown and harvested and incorporated to a different extent. Treatments were all radish incorporated in the soil (cover crop); half of the crop harvested (intermediate); 3/4<sup>th</sup> of radish harvested (rotational crop); radish brought from another area; only above ground part of radish brought from another area. In addition two control treatments were included: untreated and haloxyfop applied. Cotton, which is an important crop of which johnsongrass is a common weed, was grown after treatments. Cotton yield and johnsongrass infestation level were measured.

To explore the possibilities of alternative allelopathic *Brassicaceae* crops, both field and laboratory experiments were carried out in 2002 and 2003. Five *Brassicaceae* crops, round white radish (*Raphanus sativus*), black radish (*Raphanus sativus* var. *niger*), little radish (*Raphanus sativus* L. var. *radicula*), turnip (*Brassica campestris* subsp. *rapa*) and rapeseed were compared with garden radish. Rhizome bioassays were done using *Brassicaceae* extracts at different concentrations in controlled conditions. Extracts were prepared using above ground parts of plants at early flowering stage. In field experiment, after *Brassicaceae* crops were harvested and crop residues incorporated into the soil, johnsongrass infestation level was determined up to the 60<sup>th</sup> day after incorporation. All experiments were analyzed using proper statistical method.

## Results and Discussion

In 2002, johnsongrass density was too low (only one plant m<sup>-2</sup> in control plots) to compare the effect of the various treatments on johnsongrass control. However, it gave us an opportunity to determine the side effect of these treatments on cotton. None of the treatments affected cotton yield adversely. In the second year, treatments significantly affected both johnsongrass control and cotton yield. Even before harvest the effect of radish on johnsongrass density was significant. Cover crop application gave the highest cotton yield followed by herbicide application and the intermediate application.

The laboratory experiments showed that *Brassicaceae* species had varying level of suppression rates on the sprouting of rhizomes. Suppression rate of shoot powder extracts of *Brassicaceae* species varied between 35.0 and 54.4%. However, for all species, germination inhibition increased with increasing extract rate. The suppression by rapeseed, black radish, and turnip was higher than that of garden radish, while little radish suppressed rhizome sprout as much as garden radish. In field studies, johnsongrass infestation level in control plots increased from 3% coverage to 95% from the 7<sup>th</sup> day to the 60<sup>th</sup> day after crop incorporation. At the 30<sup>th</sup> day after incorporation johnsongrass infestation was 80%. Johnsongrass control was greater in 2002 than in 2003. It might be due to the effect of environmental and climatic factors between years (e.g. Rosa *et al.*, 1997). In both years, all alternative crops controlled johnsongrass better than garden radish. Whereas suppression was significantly different among species in 2002, it was not in 2003. 60 Days after incorporation of plant residues to the soil, the time period required by summer crops such as corn, cotton or soybean to reach a sufficient canopy level to compete with weeds, johnsongrass was under suppression. It was concluded that other allelopathic *Brassicaceae* crops can be used as a substitute of garden radish.

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# Biological properties of *Cirsium arvense* (L.) Scop. and possibilities of control

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## Introduction

*Cirsium arvense* is widely regarded as a noxious and invasive weed in the agricultural areas of Serbia as well as in many other countries across the world (Vrbničanin, 2004; Ziska et al., 2004). This perennial herbaceous plant species occurs in all types of crops owing to its high capacity for vegetative reproduction and a broad ecological amplitude. The species is also frequently observed in non-arable lands. The high degree of aggressiveness of Canada thistle is connected with an exceptional regenerative performance of its underground creeping roots, forming up to 8.2 m long underground creeping roots with over 500 adventive buds per m<sup>2</sup> of arable soil (Donald, 1994). Its high seed production (up to 40,000 achenes per plant) provides this species with a remarkable genetic variability. Canada thistle is also characterized by a high population variability. Some morphological characters showing the various degrees of peripheral leaf protection, e.g. the cuticle, hairs, number of stomata per unit area, are assumed to affect the absorption of herbicides by the leaf surface (Solymosi and Nagy, 1998). This research aimed at identification of effective methods for control of Canada thistle in maize, wheat and stubble.

## Material and methods

Adult *C. arvense* plants were harvested from arable fields at different localities. Leaf characteristics (smooth vs. hairy; lobed vs. non-lobed) were used to classify the plants in four groups (P<sub>1</sub> – P<sub>4</sub>). The following morphological parameters were measured: 1) plant height, 2) head number, 3) leaf blade length, 4) maximal and minimal leaf blade width, 5) lobe number and 6) terminal spine length.

Various herbicides control treatments (Table 1) were carried out in maize, wheat and stubble fields populated with *C. arvense* populations classified as P<sub>1</sub> (smooth and lobed leaves) and P<sub>3</sub> (hairy and lobed leaves). Herbicide application was done by a knapsack sprayer with a boom and eight spraying system nozzles, using 200 L water ha<sup>-1</sup>. On stubble, prior to glyphosate treatment, one variant was subjected to shallow harrowing, while another one was left without harrowing. At the moment of treatment plants of Canada thistle were 10-20 cm tall.

Efficacy evaluation in maize and wheat was done on the 30<sup>th</sup> and 60<sup>th</sup> days following treatment, and after harvest. Evaluation in the stubble was done 60 days after treatment. All evaluations were carried out by visual examination of the aboveground parts, while the underground parts were dug out.

Table 1. Herbicidal control treatments applied to *C. arvense* populations in maize, wheat and stubble

Maize	Wheat	Non-harrowed stubble	Harrowed stubble
2,4-D DMA (1 kg ha <sup>-1</sup> )	2,4-D DMA (1.25 kg ha <sup>-1</sup> )	glyphosate (1.9 kg ha <sup>-1</sup> )	glyphosate (1.9 kg ha <sup>-1</sup> )
2,4-D esters (0.6 kg ha <sup>-1</sup> )	2,4-D esters (0.72 kg ha <sup>-1</sup> )		
clopyralid (0.2 kg ha <sup>-1</sup> )	clopyralid (0.2 kg ha <sup>-1</sup> )		
dicamba (0.35 kg ha <sup>-1</sup> )	dicamba (0.11 kg ha <sup>-1</sup> )		



## Results and discussion

Statistically significant differences in morphological properties (plant height, head number, leaf blade length, maximal and minimal leaf blade, lobe number and terminal spine length) were obtained for the four examined populations of Canada thistle (Table 1).

Table 1. Morphological variability among four different *C. arvensis* populations

Population	P <sub>1</sub>	P <sub>2</sub>	P <sub>3</sub>	P <sub>4</sub>	ANOVA	
	with smooth and lobed leaves	with smooth and non-lobed leaves	with hairy and lobed leaves	with hairy and non-lobed leaves		
Character	Mean and SE	Mean and SE	Mean and SE	Mean and SE	F	p
Plant height (cm)	135,42±2,59	124,87±1,08	116,57±2,36	117,56±1,41	19,1	0,0
Head number	24,33±1,27	41,53±1,70	18,10±1,13	54,08±2,03	50,0	0,0
Leaf blade length (cm)	10,48±0,19	12,19±0,14	10,67±0,30	11,36±0,11	19,0	0,0
Max. leaf blade width (cm)	2,39±0,06	4,03±0,08	2,02±0,11	3,72±0,05	126,3	0,0
Min. leaf blade width (cm)	1,83±0,03	2,31±0,04	1,86±0,11	1,90±0,03	22,1	0,0
Lobes number	1,35±0,10	2,40±0,07	0,37±0,06	2,87±0,03	206,0	0,0
Terminal spine length (cm)	1,72±0,08	1,70±0,05	1,11±0,06	1,29±0,04	22,4	0,0

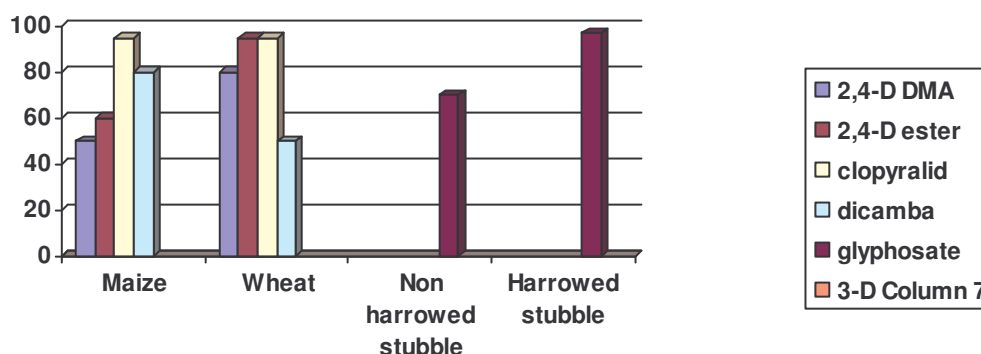


Figure 1. Efficacy of various herbicidal treatments applied to populations of *C. arvensis*

Glyphosate applied in the stubble in combination with shallow harrowing gave the best efficacy. Relatively good efficacy was also achieved by clopyralid in maize and wheat and 2,4-D esters in wheat. A moderate to good efficacy was obtained with dicamba in maize and 2,4-D DMA in wheat. In order to achieve the most effective control of *C. arvensis* shallow harrowing should be carried out after harvest, so as to provoke regeneration of adventive buds. Through this, the plants are prepared for maximum herbicide uptake and, consequently, destruction, which will ultimately reduce the potential weediness of the next crop.

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## Persistence of a new botanical type of *Alopecurus myosuroides* Huds. in Northern-Italian wheat fields.

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### Introduction

Within weed species different types of plants can frequently be distinguished. These different types can be characterized on the basis of phenological traits or ecological behaviour and become the so-called botanical forms or subspecies. This is what was for instance found in some Italian *Amaranthus* populations (Cacciato, 1996). In 2002 a particular type of *Alopecurus myosuroides* was found in populations infesting wheat fields in Emilia Romagna region in Northern Italy (Viggiani, 2005). This particular type shows all the phenological characteristics of the species, as reported in the literature (Clarke, 1980; Pignatti, 1982; Hubbard, 1984), except for the awn which is either absent or very short and at least completely hidden within the glumes. For this reason this form is henceforth called “*tectarista*”, while the normal type is denominated exerted (Figure 1). The aim of this work was to ascertain whether the *tectarista* plant type persists in the same site and whether it is present in other localities of the region.

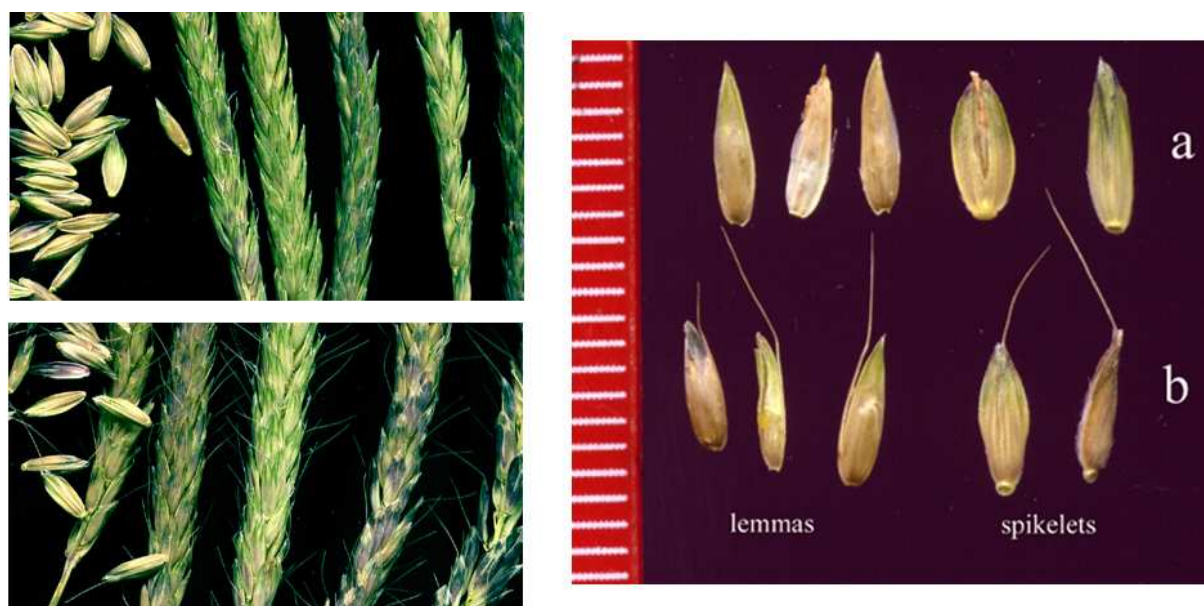


Figure 1. Panicle traits of the two types of *A. myosuroides*: a = *tectarista*; b=exerted

### Materials and methods

In June 2004, two years after the first findings, the presence of the *tectarista* form of *A. myosuroides* was assessed in four wheat fields with a predominant infestation of blackgrass. The fields had a clay soil, were 15 km apart from each other and in none of the fields herbicides were used. Two fields were located near Bologna (in Bentivoglio and Medicina) where wheat followed a three year alfalfa crop, and two fields were in the province of Ferrara, in Goro (maize as preceding crop) and Ariano Ferrarese (wheat following sugarbeet). In Medicina and Ariano *Triticum durum* Desf. was cropped (cv. San Carlo and Orobel,

respectively), whilst in Goro and Bentivoglio the crop was *T. aestivum* L. (cv. Bologna and Mieti).

The fields represent a wide area of the Po valley, where a similar weed control strategy is commonly used in wheat. Against narrow leaved weeds the most frequently used herbicides are pendimethalin, clodinafop-propargile and fenoxaprop-p-ethyl. In each field four sample areas (1 m<sup>2</sup> each) were chosen. The fields were similar regarding ecological condition and blackgrass density (about 150 culm m<sup>-2</sup>). In the selected areas two hundred *A. myosuroides* inflorescences were randomly collected and their morphology was determined. The *tectarista* was separated from the exerted type. From each field 100 mature heads of both forms were kept for further analysis in the laboratory.

## Results and discussion

In all *A. myosuroides* populations the presence of the *tectarista* type was detected. In the various fields this type accounted for 15 to the 22% of all plants.

As in the 2002-investigation, it was confirmed that single plants have only one kind of panicles. In the laboratory a significant variability in the panicle morphology of the two types was found. The average panicle length of both types was 10, but panicle differed according to diameter, unit spikelet weight and number of spikelets per panicle (Table 1). The obtained results indicate that other morphological characteristics can be associated with the difference in awn length. Under field conditions, the *tectarista* type is mainly identifiable in a mixed *A. myosuroides* infestation because of the shorter awn length.

Table 1. Plants traits of the two botanical types of *A. myosuroides* (mean  $\pm$  standard error)

Parameter	<i>tectarista</i>	<i>exerted</i>
Number of spikelets/panicle	146.9 $\pm$ 7.8	133.2 $\pm$ 10.0
Spikelet unit weight (mg)	2.02 $\pm$ 0.09	1.77 $\pm$ 0.05
Panicle diameter (mm)	4.12 $\pm$ 0.06	4.62 $\pm$ 0.12

The survey revealed that the *tectarista* form of *A. myosuroides* was still present in the same location as observed in 2002 and also in other regional fields. This presence and persistence can be of great importance not only from a vegetational point of view but also agronomically. Because the *tectarista* type is not mentioned in the current literature regarding the variability within *A. myosuroides* species probably it is spreading as a weed in Italy only recently. This spread can be probably related to the herbicide resistance problems which are frequently reported for this species all over the world. To verify this hypothesis it would be interesting to measure the heritability of the *tectarista* character and its competitive power in herbicide treated crops. Studies on these topics, using the collected material, have recently been started in our institute.

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# Sugar beet Kautsky curves and photosystem II inhibitors

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## Introduction

Photosystem II (PSII) inhibiting herbicides dramatically change the shape of the Kautsky curve, because they inhibit the transfer of electrons from  $Q_A$  to  $Q_B$  (Hiraki *et al.*, 2003). The shape of the Kautsky curve is also affected by herbicides with other modes of action (Christensen *et al.*, 2003; Abbaspoor & Streibig, 2005). Metamitron is an herbicide for weed control in sugar beets and is rapidly degraded to non-phytotoxic compounds. Terbutylazine is a potent herbicide used in for example maize; and sugar beet has a limited capacity to metabolize it to non phytotoxic compounds.

The objectives of this study were to determine how rapidly metamitron and terbutylazine affect the Kautsky curve, defined by maximum quantum efficiency ( $F_v/F_m$ ); and whether recovery from PS II damage can be mirrored in the shape of the Kautsky curve as a function of time after exposure. Maximum quantum efficiency was related to the final biomass at the termination of the experiments.

## Materials and methods

Sugar beets were grown in greenhouse in an aerated nutrient solution (hydroponics culture) from January-February 2004 (first experiment) and from March-May 2004 (second experiment). Sugar beet seeds were germinated in vermiculite, and two weeks after germination, four plants per pot were transplanted into the nutrient solution. Solution was renewed once a week before herbicide treatment and renewed three days after treatment (DAT) with untreated media. Subsequently it was renewed once a week until harvest 17 DAT in the first and 12 DAT in the second experiment at the ten-true-leaf stage. A completely randomized design with three replications was used; the untreated pots had six replications. The dose ranges of metamitron were (0-280 mg a.i.  $l^{-1}$ ) and dose ranges of terbutylazine were (0-10 mg a.i.  $l^{-1}$ ).

The chlorophyll fluorescence was measured by a portable chlorophyll fluorometer, which emits a light of 650 nm wavelengths with an intensity of 2500  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$  for 10 seconds. The fluorescence measurements were taken at two hours after treatment and one, two, three DAT and two, four and six hours after renewing the solution and subsequently once a day until harvest. The Kautsky curves, obtained by the BIOLYZER program (Rodriguez & Strasser, 2002) for different doses and time intervals were visually examined for the effects of time and dose. The parameter derived from the Kautsky curve was:  $F_v/F_m$ , maximum quantum efficiency of PSII, ( $F_v/F_m = (F_m - F_0)/F_m$ ), where  $F_0$  is the fluorescence at the ground state value and  $F_m$  is the maximum value.

## Results and discussion

In unstressed plants, the value of  $F_v/F_m$  is close to 0.83, independent of plant species (Hiraki *et al.*, 2003). Therefore, maximum quantum efficiency,  $F_v/F_m$ , is a good indicator of the physiological status of the plant. Terbutylazine affected  $F_v/F_m$  (figure 1, right) irreversibly in low doses (1 mg a.i.  $l^{-1}$ ) while plants recovered from metamitron injury even at the highest doses (140 mg a.i.  $l^{-1}$ ) (Figure 1, left)

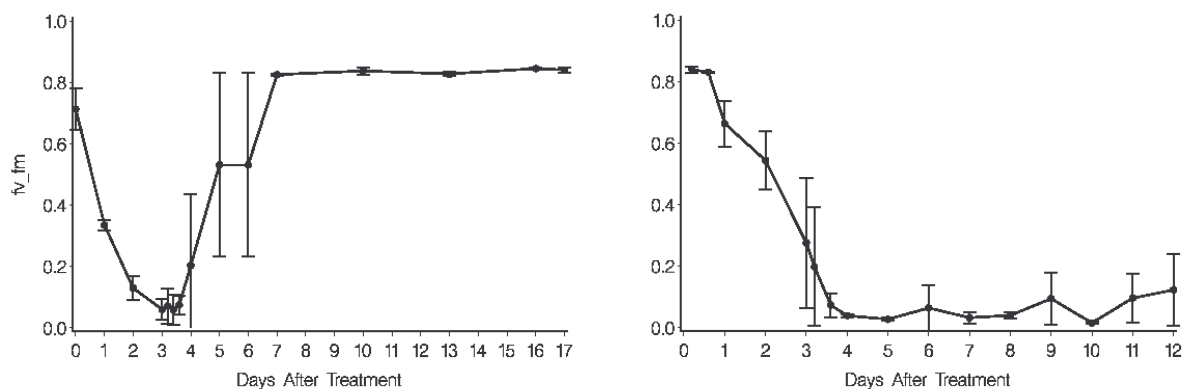


Figure 1. Recovery of maximum quantum efficiency of PS II ( $F_v/F_m$ ) from exposure to metamitron (left, dose=140 mg a.i. l<sup>-1</sup>) in the first experiment and terbuthylazine (right, dose=1 mg a.i. l<sup>-1</sup>) in the second experiment.

Dose-responses of dry matter on metamitron and terbuthylazine showed sugar beet dry matter decreased significantly at much lower doses of terbuthylazine than metamitron (data not shown). Even though the obvious recovery of the PSII damage took place about seven DAS, four days after discontinuation of the metamitron exposure (Figure 1, left), the final dry matter yield was still affected negatively. The PSII damage with terbuthylazine was so severe that discontinuation of terbuthylazine exposure did not matter.

There was well-defined linear relationship between  $F_v/F_m$  and dry matter for both herbicides at the termination of the experiments (data not shown). This linear relationship has been reported for bentazone, another PS II inhibitor (Christensen *et al.*, 2003). In a previous study we also find linear relationship between some fluorescence parameters and dry matter for ACCase inhibitors for barley and oat (Abbaspoor & Streibig, 2005).

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# Carryover effects of propoxycarbazone-sodium on winter and spring oilseed rape and sugar beet

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## Introduction

The herbicide propoxycarbazone-sodium is characterized by a high efficacy against grass weeds, including *Agropyron repens*, as well as some broadleaf weed species in winter wheat (Feucht et al. 1999, Scogan et al. 1999, Adamczewski et al. 2000). Some sulfonylurea herbicides can persist in the soil for quite a long time, and hence can present a potential threat to following crops (Paradowski 1994, Adamczewski et al. 1998). According to Amman et al. (2000) propoxycarbazone-sodium degrade quickly in the soil and does not constitute a problem to following crops.

The aim of the experiments were to determinate the carryover effects of propoxycarbazone-sodium on winter and spring oilseed rape and sugar beet.

## Material and methods

Biological activity of herbicide propoxycarbazone-sodium (Attribut 70 WG, 700 g kg<sup>-1</sup>, Bayer CropScience) was assessed in field and greenhouse experiments. In the field experiment propoxycarbazone-sodium was applied at two doses with four replications in the middle of April in a winter wheat crop. Following harvest of winter wheat the soil was cultivated and at the end of August winter oilseed rape was drilled. After germination plant development was assessed. In the following spring oil seed rape was drilled after shallow soil cultivation. In the spring oilseed rape emergence was assessed and prior to harvest 30 plants from each plot were sampled and length, weight and number of pods were determined.

In the greenhouse experiments soil in pots was spray with 3 doses of propoxycarbazone-sodium and four months latter 25 seeds of winter oilseed rape was sown. Forty days after emergence number of plants was counted and plants fresh weight was recorded. One day after harvest 10 seeds of sugar beet and 10 seeds of spring rape were sown in the same pots. Forty days after emergence number of plants was counted and plants fresh weight was determined. Soil moisture was kept at 60-70 % of field capacity throughout the experiment.

## Results and discussion

Propoxycarbazone-sodium used alone and with adjuvants was evaluated for broadleaf and grass weed control by Scogan et al. (1999) and Adamczewski et al. (2000). There are reports on the influence of propoxycarbazone-sodium on the environment (Amman et al. 2000). The first reported symptoms on sugar beet and winter oil seed rape leaves were similar to those caused by sulfonylurea compounds (Paradowski 1994). In the field experiment germination of oilseed rape was normal but 10 days later injury symptoms became visible and a reduction in the number of emerged plants was observed (Table 1). About 35-45 days after germination all plants had died. In the following spring germination of spring oilseed rape was normal but 20 days later injuries was observed. The injury of spring oilseed rape was manifested mainly by leaf deformations, height reduction and a reduction in the number of pods per plants.

In the greenhouse experiments germination was highest in the in the control treatments. The number of plants and fresh weight were lower when the highest herbicide rate was used. One day



after harvest of winter oilseed rape spring oilseed rape and sugar beet was sown in the same pots. Irrespective of dose propoxycarbazone-sodium reduced plant density and fresh weight statistically significant compared to the control. Deformation of plants was also noticed. The symptom of propoxycarbazone-sodium damage was reduction of plant growth, leaf deformation (ribbon like leaves) and deformation of the apex. The degree of deformation depended on herbicide dose. The most severe damage occurred when propoxycarbazone-sodium was applied early at a high dose. Carryover effects on spring oilseed rape were similar to those observed in the autumn on winter oilseed rape. Carryover effects on sugar beet were less severe than on spring oilseed rape.

The results obtained from the field and greenhouse experiments showed a very strong carryover effect of propoxycarbazone-sodium on winter and spring oilseed rape as well as sugar beet. The negative effects were manifested as a reduction in the number of emerged plants, reduction in plant density and leaf deformations. On spring oilseed rape a marked reduction in the number of pods was also noted. These observations suggested a relatively slow soil degradation of propoxycarbazone-sodium. The results obtained in the present experiment are different from the results reported by Amman et al. (2000).

Table 1. Carryover effects of propoxycarbazone-sodium on oilseed rape (field experiment)

Dose	Winter oilseed rape, 2002			Spring oilseed rape, 2003			
	Plants m <sup>-2</sup>			Plants m <sup>-2</sup>	Height cm	Fresh weight g	Pods plant <sup>-1</sup>
	20.09	10.10	25.10				
Control	54	54	54	84	98	81	49
60 g a.i. ha <sup>-1</sup>	35	17	0	83	85	69	10
100 g a.i. ha <sup>-1</sup>	22	4	0	82	72	65	2

Table 2. Carryover effects of propoxycarbazone-sodium on oilseed rape and sugar beet (greenhouse experiment)

Dose	Winter oilseed rape		Sugar beet		Spring oilseed rape	
	Plants pot <sup>-1</sup>	Fresh weight g plant <sup>-1</sup>	Plants pot <sup>-1</sup>	Fresh weight g plant <sup>-1</sup>	Plants pot <sup>-1</sup>	Fresh weight g plant <sup>-1</sup>
Check	20,5	3,5	9,0	3,5	9,5	2,3
60 g a.i. ha <sup>-1</sup>	17,5	2,3	8,0	3,1	7,7	1,1
100 g a.i. ha <sup>-1</sup>	15,8	2,1	7,8	2,8	7,3	0,3
140 g a.i. ha <sup>-1</sup>	14,0	1,3	7,5	1,0	6,5	0,2
LSD(0,05%)	1.41	0.25	0.75	0.39	1.26	0.17

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## Chemical and mechanical weed control in white head cabbage – a comparison

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### Introduction

Many studies have been recently focused on non-chemical weed control methods. Herbicides provide an easy and effective way to manage weeds infestation. Mechanical cultivation is an integral part of the weed management practices available as an alternative to herbicides. It is an environmentally friendly option of weed control and it can lead to reduction in herbicide use (Rasmussen, 1996, Baumann & Slembrouck 1994). The use of herbicides is still the main method of weed control in cabbage, although this crop can be grown under good cultural practices with mechanical inter-row cultivation and intra-row hand hoeing as the only weed control measures. Late head cabbage grown without herbicides usually requires 2-3 mechanical soil cultivation, depending on weed species and weather conditions, mainly rainfall. To reduce the chemical input in cabbage weed management can be based on preplant or post-planting application of a herbicide, supplemented with mechanical cultivation and hand hoeing. It is, however, still an open question, how additional mechanical and manual weeding, used after herbicides application, influence the weed population and the crop. The present studies were undertaken to evaluate the influence of weed management systems combining herbicides with mechanical cultivation and hand hoeing on growth and yield of white head cabbage.

### Materials and methods

The trials were conducted in 2002-2004 at Skierniewice, to compare the effect of weed management systems including herbicides and additional mechanical cultivation and hand hoeing in cabbage. In experiments carried out on a pseudopodsolic, sandy soil (1.2-1.6 % of organic substances, pH 6.5), cabbage (cv. Transam F<sub>1</sub>) was planted on 3 and 4 June. Trifluralin (0.96 kg a.i. ha<sup>-1</sup>) and oxyfluorfen (0.24 kg a.i. ha<sup>-1</sup>) were applied before planting of cabbage while metazachlor (0.8 and 1.0 kg a.i. ha<sup>-1</sup>) was applied 7-10 days after planting. Trifluralin was incorporated in to soil to a depth of 5-10 cm. Herbicides were chosen to achieve different levels of weed control. Additionally, one and two mechanical inter-row cultivation and intra-row hand hoeing in rows were performed and the results were compared to untreated control. In the experiments three treatments were studied: A) no hand and no mechanical weeding, B) herbicides followed by one inter-row mechanical cultivation and one intra-row hand hoeing and C) herbicides followed by two inter-row mechanical cultivations and two hand hoeings in the rows. The first mechanical weeding and hand hoeing was done 3 weeks after planting when weeds were small, while the second treatment was done after 5-6 weeks. Mechanical cultivation was performed to a depth of 2-3 cm depth using a hoe weeder. Shallow cultivation is preferred because it protects against crop root injuries. Small weed seedlings, cut by hoes, were exposed to the drying effects of the wind and sun and they quickly desiccated and died. Cabbage was harvested after 140-147 days.

### Results and discussion

Cabbage without weed control was strongly infested by weeds (Table 1). After 3 weeks of vegetation almost 120 weeds·m<sup>-2</sup> and after 5-6 weeks 194 weeds·m<sup>-2</sup> were recorded. At high-weed densities, even with the most effective use of direct weeding methods, some weeds are likely to survive (Rasmussen, 1993). In this case the mechanical treatment should be repeated. The herbicides oxyfluorfen applied before planting plus metazachlor applied after planting

provided excellent weed control. In contrast, trifluralin had little effect on the number of weeds and resulted in a higher secondary weed infestation. Post planting soil cultivation and hand weeding removed effectively weeds and has reduced secondary weed infestation in all treatments.

Herbicides were selective to cabbage. The yield of cabbage grown without herbicides increased by 15% following one hand hoeing and one mechanical weeding in comparison to no mechanical control. Increasing the number of mechanical and hand weedings to two had no effect on the yield. Effective herbicides enhanced cabbage yield more than less active herbicides. Additional mechanical cultivation and hand hoeing, performed 3 weeks after herbicides application, increased the yield of cabbage but repeating the treatment after 5-6 weeks resulted in a reduction in the yield. This tendency was observed with all herbicides. It can be concluded that the use of inter-row mechanical cultivation and hand hoeing for weed control in white head cabbage grown on a sandy soil is necessary in case of poor effect of herbicides. Following the use of effective herbicides no additional hand hoeing and mechanical weeding is required to ensure the yield.

Table 1. The response of weeds and white head cabbage to different weed control systems.

	Method of weed control*	Herbicide (kg·ha <sup>-1</sup> ) **			Untreated
		Trifluralin 0.96	Metazachlor 1.0	Oxyfluorfen + Metazachlor 0.24 + 0.8	
Number of weeds per m <sup>2</sup> :					
<i>3 weeks after planting</i>	A	104.1	11.8	2.4	119.5
<i>5-6 weeks after planting</i>	A	137.9	28.3	3.7	194.0
Chlorophyll content:					
<i>36-40 days after planting</i>					
<i>(mg per 100 cm<sup>-2</sup> of leaf surface)</i>	A	7.1	7.2	7.0	7.2
	B	6.8	6.8	7.0	6.7
	C	7.0	6.4	6.75	6.5
<i>110-120 days after planting</i>					
<i>(mg per 100 cm<sup>-2</sup> of leaf surface)</i>	A	7.8	7.8	7.1	7.3
	B	7.7	7.8	8.15	7.2
	C	7.3	7.7	7.55	7.7
Weed infestation before harvest (%)					
	A	11.4	6.2	2.4	23.7
	B	1.5	1.2	0.5	3.4
	C	1.8	0.9	1.4	4.0
Yield (t·ha <sup>-1</sup> )					
	A	93.8	98.6	102.7	85.0
	B	101.4	99.6	101.0	100.2
	C	93.2	96.7	94.7	97.1

\* A) herbicides; B) herbicides + one inter-row mechanical cultivation + one intra-row hand hoeing; C) herbicides + two inter-row mechanical cultivation + two intra-row hand hoeing,

\*\* trifluralin and oxyfluorfen applied before planting, metazachlor 7-10 days after planting,

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# Primisulfuron – methyl for weed control in maize under Lithuanian conditions

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## Introduction

Weeds compete with a crop for water, nutrients and light. This interference with crop growth often reduces yield, and may also causes secondary problems such as reduced grain quality, higher moisture content and harvest problems. The larger or the faster the weeds are growing, the greater the risks of crop interference. The key to successful weed management is controlling weeds before they can reduce crop yield. The product and dose required will depend on weed species and growth stage.

Maize is grown in rows and grows slowly making it very susceptible to weed competition. Weed control is required in virtually all maize fields every year. The critical period of weed interference in maize is from 3 to 8 weeks after planting (Hall et. al.1992, Adzgauskienė, 1996).

New sulfonylurea herbicides are being developed for a variety of uses. Their low application rates, broad weed spectrum and favourable toxicological properties have contributed to the success of this group of herbicides (James and Rahman, 1994). Primisulfuron-methyl is a sulfonylurea herbicide for the control of troublesome perennial grasses such as couch grass (*Elymus repens*) as well as annual grass weeds and many broadleaf weeds in maize (Maurer *et al.* 1987, Bhowmik *et al.* 1990). Surfactants have been reported to increase the efficacy of many post-emergence herbicides including the sulfonylureas (Fielding and Stoller 1990; Nalewaja et al. 1991).

The objective of the present study was to determine the effectiveness of primisulfuron-methyl for weed control in maize under Lithuanian conditions.

## Materials and methods

In total 8 field trials were conducted in 2000 and 2001 in different regions of Lithuania. The growing conditions were favourable for weeds, especially in 2000 when precipitation was 105 mm, 130% above the average mean, and temperature was close to average mean. In 2001 temperature was 1 – 3°C higher than the average mean and precipitation was close to average mean. Maize cultivation was the same in the all trials. Maize was fertilized with NPK before sowing, seed rate was 90,000 seeds per ha and row distance was 75 cm. Herbicides were applied post-emergence. The first application was made at BBCH 13 – 14 and the second application was made two weeks later. In all treatments with primisulfuron – methyl the surfactant Kemiwett was added at 0.2 l ha<sup>-1</sup>. Weed number per m<sup>2</sup> and weed biomass was assessed 6 weeks after application and percentage weed control was calculated on basis on weed number and weed biomass.

## Results and discussion

In total 40 weed species were found in the field experiments. *Chenopodium album* was found in all eight trials while *Elymus repens*, *Polygonum aviculare*, *Capsella bursa-pastoris* was found in six trials and *Galium aparine* and *Viola arvensis* in four trials. Weed density varied

from 400 and 600 weed m<sup>-2</sup>. Total fresh weight of the weeds varied from 1500 to 3500 g m<sup>-2</sup>. From 20 - 35 % of the total weed biomass was *Elymus repens*.

Primisulfuron-methyl was a very effective herbicide. The split application of 15 plus 15 g ha<sup>-1</sup> provided 100% control of *Capsella bursa-pastoris* and *Galium aparine*, 99 % control of *Elymus repens* and 94 % control of *Chenopodium album*. The efficacy against other weeds was lower. Against *Viola arvensis* the effect was about 70% and against *Polygonum aviculare* only 40 percent. The efficacy of the same herbicide dose as a single application was lower than of the corresponding split-application. Control of *Elymus repens* was 95%, of *Chenopodium album* 90% and of *Polygonum aviculare* only 21%.

A split application of 2,4-D amino salt + dicamba in the first spraying and primisulfuron-methyl in the second spraying was most effective against *Polygonum aviculare*.

Table 1. Efficacy of primisulfuron – methyl on weeds in maize. Average of 8 field trials

Treatment	Percentage effect					
	ELYRE	CHEAL	CAPBP	POLAV	GALAP	VIOAR
Untreated*	995,2	554,3	25,0	557,0	227,0	130,0
Primisulfuron (15 g ha <sup>-1</sup> )	88	75	100	16	90	46
Primisulfuron in split (15 g ha <sup>-1</sup> + 15 g ha <sup>-1</sup> )	99	94	100	40	100	71
Primisulfuron (30 g ha <sup>-1</sup> )	95	90	100	21	100	75
Primisulfuron (15 g ha <sup>-1</sup> )+2,4-D amino salt (344 g ha <sup>-1</sup> ) +dicamba (120 g ha <sup>-1</sup> ) in split	67	100	100	47	75	38
2,4-D amino salt (344 g ha <sup>-1</sup> )+ dicamba (122 g ha <sup>-1</sup> ) and primisulfuron (15 g ha <sup>-1</sup> ) in split	72	100	100	72	100	60

\* Fresh weight of weeds

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# The effect of herbicides on the regeneration of yellow nutsedge (*Cyperus esculentus* L.)

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## Introduction

*Cyperus esculentus* L. originates from the tropical and subtropical areas of North-Africa. Four *C. esculentus* varieties are known as weeds, while one of them (*C. esculentus* var. *sativus*, sweet almond) is a cultivated plant (Dancza et al. 2004). As a weed, in Europe *C. esculentus* var. *lepostachyus* is the most common (Schippers et al. 1995). *C. esculentus* is one of the world's worst weeds (Holm et al. 1977). It is found in several European countries, e.g. in Germany (Gieske et al. 1992), Switzerland (Schmitt & Sahli 1992), France (Bernard 1996), the Netherlands (Ter Borg 1998), Austria (Fertsak & Hain 2003). In Hungary, it was detected for the first time in 1993 (Dancza 1994). Out of the naturalized neophytes, *C. esculentus* is the only species, which has invasive and transformer characteristics in the Cyperaceae family. Transformer species is a part of the invasive species, which can change characteristic features, conditions, appearance or nature of the natural and/or agroecosystems (Richardson et al. 2000, Botta-Dukát & Balogh 2004). *C. esculentus* is dominant in maize and occurs on 3000 hectares, mainly in Somogy county in Hungary (Hoffmanné et al. 2004).

*C. esculentus* is a rhizomatous species. It develops tubers at the end of the thin rhizomes, which are ball-shaped, dark brown if mature and reaching 0.2-1 cm in diameter. One type of the tubers grow horizontally in the soil and develop above ground shoots. The other type produces new tubers. The aim of our study was to examine the effect of herbicides on the regeneration of *C. esculentus* from tubers.

## Materials and methods

Matured *C. esculentus* tubers were collected in maize stubble in Somogy county in March 2004. After cleaning the tubers were planted in plastic pots (11 cm in diameter) at 5cm depth (5 tubers in a pot). Pots were placed in a glasshouse at 23 °C. Presowing, pre-emergence and post-emergence treatments were applied in four replications (Table 1). Herbicides, applied as presowing treatments, were incorporated to 8 cm depth into the soil 5 days before planting whereafter the pots were covered with a plastic foil. Pre-emergence herbicides were sprayed on the soil surface immediately after tuber planting. Post-emergence treatments were applied at the 4-leaf stage of *C. esculentus*. A spray volume of 400 L ha<sup>-1</sup> water was used in each treatments. The effect of the herbicides were evaluated 21 days after the treatments as compared to the untreated control.

## Results and discussion

Excellent (100%) weed control was obtained with acetochlor, S-metholachlor, izoxaflutol and EPTC. *C. esculentus* tubers – except following the isoxaflutol treatments - failed to produce shoots. *C. esculentus* tubers produced shoots after treatment with isoxaflutol, but the leaves were whitened and the death of the plants occurred at the four-leaf stage. Among the pre-emergence treatments pendimethalin and triasulfuron gave 95 and 75% effect, respectively. Among post-emergence treatments bentazon and glyphosate-isopropylamin salt resulted in a 100% effect. Plants decayed after a few days, and regrowth was not observed after the

treatments. Fatty acid biosynthesis inhibitors (setoxydim and fluazifop-P-butyl) resulted in 95 and 90% effect of *C. esculentus*.

Table 1. Effects of herbicide treatments

Commercial name	Active ingredient	Dose	Application time	Percent effect
Untreated control				0
Acenit A 500 EC	500 g L <sup>-1</sup> acetochlor+50 g L <sup>-1</sup> AD-67 antidote	2.6 L ha <sup>-1</sup>	Pre-emergence	100
Dual Gold 960 EC	960 g L <sup>-1</sup> S-metolachlor	1.6 L ha <sup>-1</sup>	Pre-emergence	100
Logran 75 WG	75% triasulfuron	20 g ha <sup>-1</sup>	Pre-emergence	75
Merlin WG	75% isoxaflutol	140 g ha <sup>-1</sup>	Pre-emergence	100
Stomp 330	33% pendimethalin	6.0 L ha <sup>-1</sup>	Pre-emergence	95
Witox 72 EC	72% EPTC	5.5 L ha <sup>-1</sup>	Presowing	100
Basagran	480 g L <sup>-1</sup> bentazone	4.0 L ha <sup>-1</sup>	Post-emergence	100
Fusilade Forte	15% fluazifop-P-butyl	2.8 L ha <sup>-1</sup>	Post-emergence	90
Nabu-S	12.5% setoxydim	6.0 L ha <sup>-1</sup>	Post-emergence	95
Gialka 480 Plus	480 g L <sup>-1</sup> glyphosate-isopropylamine	5.0 L ha <sup>-1</sup>	Post-emergence	100

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# Characterization of the effects of various adjuvants on flumioxazin activity

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## Introduction

Flumioxazin is a new N-phenylphthalimide herbicide that has been registered for grapes and olives in Europe and in the USA as either pre-emergence application in peanut, soybean or post-directed in cotton. The manufacturer clearly states to use a non-ionic surfactant with flumioxazin. Most of the research on efficacy of flumioxazin has been conducted with non-ionic surfactants (Askew et. al., 2002). Once fully commercialized, users would likely use various adjuvant combinations with flumioxazin to optimize its efficacy. Adjuvants are often used to improve spray retention, wetting of the leaf surface, and to enhance uptake of the active ingredients (Green and Hazen, 1998). However, little research on the effects of various types of adjuvants on flumioxazin activity has been previously reported (Price et. al., 2004). In the current research, a study was conducted to characterize the contact angle, spread patterns, and wetting ability of six major types of adjuvants in tank mixture with flumioxazin.

## Materials and methods

Six adjuvants including a non-ionic surfactant (Trend), an ethylene phenol ether (Saldo), a parafinic oil (Sunoil 11<sup>®</sup>), a methylated seed oil (Headland Fortune), ouria ammonium nitrate (UAN), and an organosilicone (Silwet 806) were studied. Plants of *Solanum nigrum* were raised in a greenhouse and sprayed at two growth stages (4-6 leaves, 10cm, 0.43 g dry.weightplant<sup>-1</sup> or 7-9 leaves, 22cm, 0.74 g dry.weightplant<sup>-1</sup>). Percentage control was calculated on basis of the reduction of dry weight compared to the controls. Contact angles were measured by depositing 1 µl droplets on vertical positioned leaf sections mounted with a double side tape on rectangular glass slides under a stereoscope. The experiment was repeated twice and each mean represents at least 20 different droplets. Droplet spreading was recorded 1 min after droplet deposition. Spray retention was measured as described by Young and Hart (1998). Efficacy was assessed by treating plants with 2, 4, 6 or 12 droplets (0.5µl each; 1.6 µg a.i./droplet) of flumioxazin alone or in combination with Trend or Silwet 806.

## Results and discussion

The non-ionic surfactant and organosilicone adjuvant increased flumioxazin activity particularly at the 0.5X rate (200 g a.i. ha<sup>-1</sup>) when applied to medium sized plants (Table 1). In contrast, the MSO decreased flumioxazin activity at the 0.5X rate. Adding the organosilicone adjuvant resulted in complete wetting of the entire leaf even with 1 µl droplet solution, i.e. a zero contact angle. Fluorescent dye showed a rapid infiltration of the flumioxazin+organosilicone mixture into the leaf. The non-ionic surfactant Trend produced a contact angle of 51 degrees and the highest retention (71%). The organosilicone adjuvant required 2 droplets whereas the non-ionic surfactant required 4 droplets for maximum efficacy on *S. nigrum* plants. These results showed that the effects of adjuvants on flumioxazin activity could be characterized.

Table 1. Percentage control of medium and large plants, contact angle of 1µl droplet, spread of 1µl droplet, spray retention of foliar application, and percentage control of increasing numbers of droplets on *S. nigrum*.

Treatment	Percentage control		Contact angle (degrees)	Droplet spreading (mm <sup>2</sup> )	Spray retention (%)	Control based on number of droplets			
	Medium <sup>1</sup> Plants	Large <sup>2</sup> Plants				2	4	6	12
	%					%			
FL	88	55	55	0.49	32	12	30	72	100
FL+Trend	82	72	51	0.73	71	66	95	91	99
FL+Saldo	82	58	32	0.73	63	- <sup>3</sup>	-	-	-
FL+Sunoil	73	68	54	0.72	41	-	-	-	-
FL+Fortune	79	50	44	0.74	64	-	-	-	-
FL+UAN	85	52	63	0.55	49	-	-	-	-
FL+S 806	82	72	67	0.00	29	100	100	100	100
LSD (0.05)	20		7.2	0.06	17	26			

<sup>1</sup> 4-6 leaves, 10cm, 0.43g d.wt./plant.

<sup>2</sup> 7-9 leaves, 22cm, 0.74g d.wt./plant.

<sup>3</sup> Not measured.

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# Possibilities for reducing herbicide inputs in sugar beet

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## Introduction

Effective weed control is a critical component of profitable sugar beet production. Left uncontrolled, weeds may reduce yield, interfere with harvest, reduce the value of the crop, and increase future weed problems. One of the basic principles of annual weed control is that post-emergence herbicides are more effective on small weeds than large weeds. Norris (1991) estimated that for every day the application of phenmedipham plus desmedipham passed the optimum time of application, control was reduced by one to two percent. The most popular post-emergence herbicides are phenmedipham, desmedipham, ethofumesate, metamiltron and chloridazon. In sugar beet herbicides are applied from two to five times after crop emergence (Bauer, 1997; Bee et al., 1995, Dexter & Luecke, 1997; Fisher et al., 1995; Wilson, 1999). Herbicide efficacy also depends on the herbicide mixture used. Results have shown that on some weed species it is possible reduce herbicide doses in sugar beet (Lajos & Lajos, 2000). The objective of this study therefore was to evaluate the efficacy of reduced dose rates of these herbicides for the control annual broad-leaved weed.

## Materials and methods

The experiment was conducted for 3 years in Dotnuva, Lithuania on a light loamy soil. The experimental design was a randomised complete block with four replications. The treatments were phenmedipham+desmedipham+ethofumesate (Betanal Expert, 91+71+112 g a.i. l<sup>-1</sup>, Bayer Crop Science ), metamiltron (Goltix, 700 g kg<sup>-1</sup>, Maktheshim Agan Industries Ltd.), chloridazon (Pyramin Turbo, 520 g l<sup>-1</sup>, BASF A/S) and chloridazon + quimerac (Fiesta T, 360+60 g l<sup>-1</sup>, BASF A/S). Herbicides were applied at three doses (1/1 N, 3/4 N and 1/2 N of the recommended dose) as three applications. The first application was done at the early cotyledon stage of the weeds. Subsequent applications were applied when the next weeds flush emerged or 7-14 days after the first flush. Four weeks after treatments dry weights of weeds were recorded. A quadrat of 0.20 m x 1.25 m was randomly thrown in each plot. Weed dry weight were transformed to  $Y = \sqrt{X}$  and analyzed using STATISTIKA software.

Table 1. Treatments and doses

Treatments	Dose (g a.i. ha <sup>-1</sup> )		
	1/1 N	3/4 N	1/2 N
Phenmedipham+desmedipham+ethofumesate	91+71+112	68+53+84	46+36+56
Phenmedipham+desmedipham+ethofumesate+	91+71+112	68+53+84	46+36+56
Metamiltron	700	525	350
Phenmedipham+desmedipham+ethofumesate+	91+71+112	68+53+84	46+36+56
Chloridazon	650	488	325
Phenmedipham+desmedipham+ethofumesate+	91+71+112	68+53+84	46+36+56
Chloridazon+quimerac	540+90	405+68	270+45
Phenmedipham+desmedipham+ethofumesate+	91+71+112	68+53+84	46+36+56
Metamiltron+	700	525	350
Chloridazon	650	488	325
Phenmedipham+desmedipham+ethofumesate+	91+71+112	68+53+84	46+36+56
Metamiltron+	700	525	350
Chloridazon+quimerac	540+90	405+68	270+45

## Results and discussion

The weed spectrum differed between years. In 2002 *Chenopodium album* L. and *Tripleurospermum perforatum* (Merat) M.Lainz dominated the weed flora, in 2003 *C. album*, *T. perforatum*, *Fallopia convolvulus* (L.) Löve, *Polygonum aviculare* L., *Galium aparine* L. and *Lamium purpureum* L. were the most prevalent weed species and in 2004 *Sinapis arvensis* L., *Viola arvensis* Murray, *C. album* and *Thlaspi arvense* L were the most frequently found species. Results indicated that addition of met amitron, chloridazon and chloridazon + quimerac increased efficacy by 10 to 98% compared to phenmedipham+desmedipham+ethofumesate. Similar results have been reported previously ((Lajos & Lajos, 2000). The addition of met amitron significantly increased effectiveness, whereas addition of chloridazon and chloridazon + quimerac did not. The most effective treatments were mixtures consisting of 5 and 6 different active ingredients, e.g. phenmedipham + desmedipham + ethofumesate + met amitron + chloridazon and phenmedipham + desmedipham + ethofumesate + met amitron + chloridazon + quimerac. Differences between treatments were constant across dose rates (Table 2).

Table 2. The effect of herbicide mixtures on the weed dry weight ( $\text{g m}^{-2}$ )

Treatments	Dose ( $\text{g a.i. ha}^{-1}$ )		
	1/1 N	3/4 N	1/2 N
Phenmedipham+desmedipham+ethofumesate	114,8	175,8	218,0
Phenmedipham+desmedipham+ethofumesate+ Metamitron	13,0**	38,4*	109,2*
Phenmedipham+desmedipham+ethofumesate+ Chloridazon	65,0	95,3	195,2
Phenmedipham+desmedipham+ethofumesate+ Chloridazon+quimerac	58,0	92,3	160,0
Phenmedipham+desmedipham+ethofumesate+ Metamitron+	5,8**	19,21**	49,6**
Chloridazon			
Phenmedipham+desmedipham+ethofumesate+ Metamitron+	2,9**	12,5**	38,2**
Chloridazon+quimerac			

\*, \*\*: differences significant at the 5% and 1% level, respectively.

In conclusion, the potential for reducing herbicide doses depended on the herbicide and the weed spectrum. The larger the number of active ingredients included in the herbicide mixture was, the more effective it was. Applying phenmedipham + desmedipham + ethofumesate in mixture with met amitron and chloridazon or met amitron and chloridazon + quimerac at doses reduced by 25 to 50% produced consistent control of *C. album*, *T. perforatum*, *G. aparine*, *F. convolvulus* and *P. aviculare*.

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# Influence of shepherd s purse (*Capsella bursa pastoris* (L.) Medic.) and its control on seed productivity of lucerne (*Medicago sativa* L.)

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## Introduction

The efficient weed control is one of the main prerequisites for high-yielding and profitable production of lucerne seeds. Shepherd s purse (*Capsella bursa pastoris* (L.) Medic.) is a permanent and wide-spread companion of lucerne (Kolev, 1963; Dimitrova, 2001; Cuturilo et Nicolic, 1986). The weed is included in the approved list of the economically important pests of agricultural crops in the Republic of Bulgaria, as well as in the group of host weeds for viruses. Several studies indicated the need to control weeds in lucerne and reported efficient and selective herbicides (Boyles et al., 1987; Dimitrova, 1995; Livenskii & Cherenkov, 1990; Wilson, 1986). The objective of the study was to determine the influence of shepherd s purse (*Capsella bursa pastoris* (L.) Medic. and its control on seed productivity of lucerne (*Medicago sativa* L.).

## Material and Methods

The study was carried out in the period 2000-2002 under field conditions, on slightly leached chernozem soil, without irrigation. The trial was laid out by the block method with four replications in a seed production lucerne stand at an inter-row spacing of 25 cm with a severe natural weed infestation of *Capsella bursa pastoris* (L.) Medic. The following variants have been included in the trial:

Variants	Products	Rate g a.i./ha	Application Time
A1	Control	-	-
A2	Hand weeding	-	Season long control (weed free)
B1	Imazethapyr	40	Early post-emergence: Lucerne = 2-3 cm height
B2	Imazethapyr	80	Early post-emergence: Lucerne = 2-3 cm height
C1	Imazethapyr	40	Post-emergence: Lucerne = 8-10 cm height
C2	Imazethapyr	80	Post-emergence: Lucerne = 8-10 cm height

Imazethapyr was applied with 500 l water volume/ha

Seeds were harvested from the regrowth. The following characteristics were observed: degree of weed infestation (by the quantity weight method); herbicidal efficiency; yield and qualities of lucerne seeds (germinable energy, germinability, 1000-seed weight).

## Results and Discussion

The usual practice in our country is to harvest seeds from the lucerne regrowth. For that reason the seed producers often omit the treatment of the primary growth with herbicides against *Capsella bursa pastoris* (L.) Medic. relying on its mechanical removal by the cutting of the sward. The necessity of treatment is emphasized by the biological characteristics of the weed which is an annual wintering species with a great diversity of forms. In the species there are plants developing as ephemeral, spring, late spring, late and wintering weeds. Our observations show that the weed wintering and ephemeral forms which are of importance to lucerne are found at the greatest density. The herbicide imazethapyr shows a great selectivity to lucerne, irrespective of its stage of development. Its high herbicidal efficiency is due to its wide-spectrum action with regard to the weeds, stability within a wide range of temperature

conditions and its capability to be absorbed by the soil, as well as by vegetating plants (Table 1). The highest effect against the weeds (93-95 %) was achieved when treating at early vegetation at lucerne height of 2-3 cm. and early stages of weed development. At that time at the low rate of 40 g a.i./ha the effect achieved was almost equal to that at the twice as high rate which is of interest in an ecological and economic aspect. The treatment at a later lucerne stage (at lucerne height of 8-10 cm.) which often happens in wide practice led to a decrease of the effect and an increase of the herbicide rate. That was explained by the advanced stage of weed development and by the limited access of herbicidal solution to them because of the lucerne canopy. *Capsella bursa pastoris* is an aggressive weed with regard to the essential factors of production. The weed not only strongly decreases the yield and the quality of herbaceous forage (Dimitrova, 2001, Temme et al., 1983), but also exerts a negative influence on the reproductive organ formation, which leads to reduction of its biological potential. The grounds for that were given by the seed yield from the weeded check (A2) exceeding that of the zero check (A1) by 46% (Table 1). The elimination of the weed competitive effect by treatment with imazethapyr resulted in an increase of seed yield by 19 to 42% depending on the crop stage and the herbicide rate with good and very good statistical significance of differences. The treatment at early vegetation at lucerne height of 2-3 cm. had markedly better results also with regard to seed productivity. As far as the seed qualities are concerned they met the standard requirements, but there was no trend to difference between the separate variants. *Capsella bursa pastoris* (L.) Medic. is an aggressive weed of economic importance to lucerne exerting a negative influence on its reproductive capabilities; The lucerne treatment at early vegetation at lucerne height of 2-3 cm with imazethapyr at the rate of 40 g a.i./ha ensured 93% herbicidal efficiency and increased its seed productivity by 35%.

Table 1. Influence of *Capsella bursa pastoris* (L.) Medic and its control on the seed productivity of lucerne (*Medicago sativa* L.) and herbicidal efficiency

Variants	Seed yield, kg/ha					Herbicidal efficiency, %
	2000	2001	2002	average* (2000-2002)	%	
A1	174	203	190	189	100	-
A2	252	305	274	277	146	-
B1	239	287	261	255	135	93
B2	243	296	265	268	142	95
C1	201	253	223	225	119	68
C2	224	269	244	245	130	80

\* Note:  $LSD_{0.05} = 1,8080$ ,  $LSD_{0.01} = 2,5716$ ,  $LSD_{0.001} = 3,7224$

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## First trials with selective and non-selective weed control with Organic Interceptor™

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### Introduction

Weed control in organic farming is often troublesome, time consuming and expensive. Since herbicides are not allowed, weeds are managed by means of agronomical practices, such as crop rotation, cover crops, false seedbed preparation and mechanical control, namely harrowing, hoeing, mowing and clipping. However, chemical weed control is possible utilizing environmental-friendly herbicides, derived from natural substances by physical means and without any chemical and unnatural transformation. Organic Interceptor™ is the first certified organic herbicide and is the only herbicide that can be utilized in New Zealand by organic farmers, even if with some restrictions. Up to now, this herbicide is not authorized in Europe, neither in conventional nor in organic agriculture. James et al. (2002) described the characteristics of Organic Interceptor as follows: a) a non-selective contact herbicide disrupting membrane permeability causing rapid dehydration, desiccation and brown-off within a few days; b) no systematic action; c) derived from the liquid residues created during processing of pine trees for pulp and paper containing 680 g L<sup>-1</sup> of emulsifiable pine essence; d) low acute oral and dermal toxicity and inactivated on contact with soil. It has been claimed to be eco-friendly (Anon., 2001).

Organic Interceptor was tested by James et al. (2002) in different conditions, e.g. on newly emerged weeds on recently cultivated soil, on established weeds in orchards and vineyards and on previously mown, run-out pasture. Kempenaar et al. (2003) applied Organic Interceptor on hard surfaces and on fallow soils. Good results were obtained spraying Organic Interceptor at high concentrations, ranging from 10 to 20%, and at high spray volumes, up to 3200 L ha<sup>-1</sup>. Considering the available literature information is lacking on the use of Organic Interceptor for in-crop weed control. This research, funded by Intrachem Italia and still in progress, is aimed to: 1) evaluate Organic Interceptor activity on some noxious weeds; 2) test Organic Interceptor efficacy for weed control in sugar beet, onion and processing tomato crops. In this presentation, the results of the first year of investigation are reported.

### Materials and methods

The evaluation of Organic Interceptor herbicidal activity on noxious weeds was carried out in greenhouse trials performed on pot-grown weeds. A first experiment concerned prostrate knotweed (*Polygonum aviculare*) comparing the following treatments: 600 g a.i.ha<sup>-1</sup> glufosinate-ammonium (Basta, 200 g a.i.L<sup>-1</sup>, Bayer Cropscience); Organic Interceptor (10% v/v using a spray volume of 500 L ha<sup>-1</sup>); Organic Interceptor (10% v/v using a spray volume of 1000 L ha<sup>-1</sup>); Organic Interceptor (20% v/v using a spray volume of 500 L ha<sup>-1</sup>); Organic Interceptor (20% v/v using a spray volume of 1000 L ha<sup>-1</sup>); and untreated control. Experiment was arranged according to a completely randomized design with 3 replications. Plants were treated at flowering stage. Weed biomass and weed mortality were recorded 1 month after herbicide application.

Organic Interceptor efficacy was tested in field trials in sugar beet, onion and processing tomato. In each crop, experiments were arranged according to a complete randomized block design with 3 replications and 6 weed control techniques were compared: untreated control;

chemical weed control; mechanical weed control; Organic Interceptor 5% v/v; Organic Interceptor 10% v/v and Organic Interceptor 20% v/v. The volume rates of Organic Interceptor applications were 2000 l ha<sup>-1</sup> in sugar beet and in tomato, 700 l ha<sup>-1</sup> in onion. Herbicides for the chemical weed control treatment were chosen among the most widely used in Italy. Both in greenhouse and field experiments, herbicides and Organic Interceptor were applied using a backpack sprayer working at a pressure of 5 bars. In all field trials Organic Interceptor was applied post-emergence. In sugar beet and tomato Organic Interceptor was only applied between the rows. On onion, Organic Interceptor is claimed to be selective, hence it was applied as an overall spray. At harvest, the following data were registered: crop yield, crop plant density, average weight of crop products, weed biomass production, average biomass per weed, total weed density and density of some representative weed species. Data were submitted to a one-way ANOVA and mean separation was done by means of Fisher's Protected LSD test.

### Results and discussion

In the greenhouse trial, 20% v/v Organic Interceptor applied at 1000 L ha<sup>-1</sup> was most effective on *Polygonum aviculare* and this treatment was significantly more effective than glufosinate-ammonium.

The results of field trials are summarized in Table 1.

Table 1. Weed biomass and crop yield in sugar beet, onion and processing tomato trials

Weed control technique	Sugar beet		Onion		Tomato	
	Weed biomass (t ha <sup>-1</sup> )	Crop yield (t ha <sup>-1</sup> )	Weed biomass (t ha <sup>-1</sup> )	Crop yield (t ha <sup>-1</sup> )	Weed biomass (t ha <sup>-1</sup> )	Crop yield (t ha <sup>-1</sup> )
Untreated control	18.67	30.93	49.11	2.06	54.93	1.08
Mechanical control	1.78	42.75	1.79	59.32	8.00	77.84
Chemical control	2.80	47.56	4.35	62.93	18.22	43.87
Org. Int. 5%	8.30	37.72	47.40	2.64	43.31	1.59
Org. Int. 10%	5.63	44.81	39.33	3.79	38.80	14.47
Org. Int. 20%	3.76	44.62	33.64	5.46	39.22	18.74
LSD (P<0.05)	6.21	11.91	10.49	7.46	15.98	21.65

On basis of these preliminary results, it can be concluded, that Organic Interceptor at 5% v/v was ineffective. In sugar beet, Organic Interceptor at 10% and 20 % v/v provided satisfying weed control and high crop yields. Data from onion and tomato trials were less encouraging. In onion, good crop yield and effective weed control were only obtained with mechanical and chemical weed control, whereas Organic Interceptor treatments, even at 10 and 20% v/v failed. In tomato, weed control and crop yield obtained with Organic Interceptor at 10 and 20% v/v provided weed control intermediate between the untreated control and mechanical and chemical weed control. Further researches are in progress, in order to optimize Organic Interceptor application technique and application program.

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# **Control of *Anthemis arvensis*, *Chenopodium album* and *Galium aparine* control by different doses of growth regulator herbicides**

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## **Introduction**

In several European countries like Great Britain, France, Germany and the Scandinavian countries, a tendency to use pesticides more rationally has been observed in the recent years due to a political demand for a reduction in the use of the pesticides. This policy is caused by an increasing concern about the hazard to the environment and the levels of pesticide residues in the harvested plant products (Gauvrit, 1991; Proven et al., 1991; Schwarz, 2004; Thonke, 1991; Whiting et al., 1991).

In Poland a similar tendency as in others EU member countries can be observed and in recent years attempts have been made to optimise herbicide use, i.e. to minimise the amounts of biologically active substances applied to the fields. The major objective of these activities is to suppress the weeds to the level where they do not reduce crop yields rather than killing the weeds. This change in weed control practice is strictly connected with the change in the view on human interference with the environment. Regulating rather than controlling weeds becomes the overall aim of a herbicide treatment, i.e. weakening weeds and limiting their growth to the level where they do not interfere with the cultivated plant. (Domaradzki & Rola, 1999).

## **Materials and methods**

In the years 2000-2002 in total 16 field experiments were conducted (10 in spring wheat and 6 in spring barley). The experimental design used was randomized blocks with three replications and a plot size of 20 m<sup>2</sup>. All experiments were situated in productive fields in the Lower Silesia region (south-west part of Poland). In the field experiments 3 herbicides were evaluated: 2,4-D+dicamba (Aminopielik D 450 SL, 417,5+32,5 g l<sup>-1</sup>, Rokita-Agro, Poland), (1/1=3 l ha<sup>-1</sup>), mecoprop+MCPA+dicamba (Chwastox Trio 540 SL, 300+200+40 g l<sup>-1</sup>, Organika-Sarzyna, Poland) (1/1=2 l ha<sup>-1</sup>) and mecoprop-P+carfentrazone-ethyl (Aurora Super 61.5 SG, 600+15 g kg<sup>-1</sup>, FMC Corporation) (1/1=0,8 kg ha<sup>-1</sup>). Each herbicide was used at the maximum recommended dose (1/1) and in three reduced doses (3/4; 1/2 and 1/4 of the full dose).

Herbicides were applied with a knapsack sprayer ("Gloria"), at a pressure of 0.25 MPa and in a spray volume of 250 l ha<sup>-1</sup>. The application was performed from the 3-4-leaf growth stage and up to the beginning of tillering (BBCH=13-22) corresponding to the 2-6-leaf stage of the weeds (BBCH=12-16). Fresh weight of weeds was taken 5-6 weeks after the application. The aboveground part of the weeds was cut (each species individually) and the weight was recorded from 3 randomly chosen 0.25 m<sup>2</sup> areas in each plot. Herbicides efficacy was assessed on basis of the fresh weight reduction in the herbicide treated plots relative to the untreated control.

## Results and discussion

2,4-D+dicamba was only fully effective on *Anthemis arvensis* and *Galium aparine* at the full dose. In contrast *Chenopodium album* was controlled not only by the full dose but also by the 3/4 and 1/2 doses.

The application of the herbicide containing mecoprop, MCPA and dicamba controlled *Chenopodium album* at the full dose and the 3/4 and 1/2 doses. The full and 3/4 dose provided good control of *Anthemis arvensis* and *Galium aparine*.

The mecoprop-P and carfentrazone-ethyl mixture applied at the full dose and the 3/4 dose controlled *Chenopodium album* and *Galium aparine* very effectively. In the case of *Anthemis arvensis* not even the full dose of this herbicide provided full control.

In summary, the results have shown that it was possible to control *Chenopodium album* by reduced doses (50 to 75% of the full dose) of 2,4-D + dicamba, mecoprop + MCPA + dicamba and mecoprop-P + carfentrazone-ethyl. Also *Galium aparine* was controlled by mecoprop + MCPA + dicamba and mecoprop-P + carfentrazone-ethyl with doses reduced by 25%. Only against *Anthemis arvensis* the full dose was required to achieve a satisfactory level of control.

Table 1. Reduction in the fresh weight of *Anthemis arvensis* (ANTAR), *Chenopodium album* (CHEAL) and *Galium aparine* (GALAP) by different doses of growth regulator herbicides.

Active ingredients	Dose per ha	Reduction of fresh weight in %		
		ANTAR	CHEAL	GALAP
2,4-D + dicamba	1/4	49	81	56
	1/2	68	93	73
	3/4	76	99	78
	1/1	95	99	91
Mecoprop + MCPA + dicamba	1/4	53	78	61
	1/2	73	91	79
	3/4	87	99	89
	1/1	97	99	96
Mecoprop-P + carfentrazone-ethyl	1/4	45	60	67
	1/2	61	84	87
	3/4	79	92	97
	1/1	85	96	99

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# Chemical control of *Bryophyllum* species: the effect of surfactants on the foliar uptake of herbicides

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## Introduction

Introduced from Madagascar in the mid-late 1800s, mother of millions (*Bryophyllum* spp.) has spread throughout NE Australia and is now declared as a Class 2 environmental weed in Queensland. Currently there are three herbicides registered for controlling the weed in Queensland and New South Wales, but there has been little formal investigation into any benefits that may be afforded by including surfactants in tank mixes to aid the killing effect. Furthermore, approximately six species of mother-of-millions are naturalised in Australia, each with differing gross morphologies, and potentially micro-morphologies and chemistries of their respective leaf surfaces.

Thus the objectives of this study were to investigate:

- the leaf surface characteristics of two commonly occurring mother of millions, *B. delagoense* (Eckl. & J.Zeyh) Schinz and the hybrid *B. daigremontianum* (Raym.-Hamet & H.Perrier) A.Berger × *Bryophyllum delagoense* (Eckl. & J.Zeyh) Schinz to further our understanding of their similarities/ differences;
- the physical properties of various spray solutions, using the registered herbicides Grazon<sup>®</sup> DS Herbicide and Starane<sup>®</sup> 200 Herbicide in combination with various surfactants;
- any interaction between spray solutions and different *Bryophyllum* spp., including any difference in killing effect due to the addition of surfactants,

and thus to establish whether chemical control can be improved and tailored to a particular species.

## Materials and methods

The chemical and physical properties of leaf surfaces were investigated through extraction and gas chromatographic (GC) analysis of polar and non-polar components of the epicuticular waxes, and using scanning electron microscopy (SEM), respectively. The Wilhelmy plate method was used to measure equilibrium surface tension (EST) of various herbicide × surfactant solutions to select, from three candidates, one surfactant to be used in subsequent glasshouse trials. Finally, through conducting two extended pot trials, the effect of Pulse Penetrant<sup>®</sup> added to picloram present as the hexyloxypropylamine salt (Grazon<sup>®</sup> DS Herbicide, 100 g a.i. L<sup>-1</sup>, Dow AgroSciences) plus triclopyr present as butoxyethyl ester, Dow AgroSciences) and Starane<sup>®</sup> 200 (200 g a.i. L<sup>-1</sup> fluroxypyr present as the methylheptyl ester, Dow AgroSciences) was investigated. A symptom-adjusted epinasty scoring system was used to monitor plants throughout the pot trials.

## Results and discussion

Extraction of epicuticular waxes revealed similar results for both *Bryophyllum* species studied with a mean yield of 151 mg kg<sup>-1</sup> fresh leaves. Non-polar *n*-alkanes (C31-C35) constituted 46 % of extracts with C33 alkanes dominating (86 %). Polar components including alcohols and aldehydes constituted the remaining 54 % of total extracts. Scanning electron micrographs of leaf surfaces showed a rough epicuticular wax in control treatments of both

species (Figure 1A). Herbicide and surfactant treatments were solubilised and redeposited surface waxes (Figure 1B).

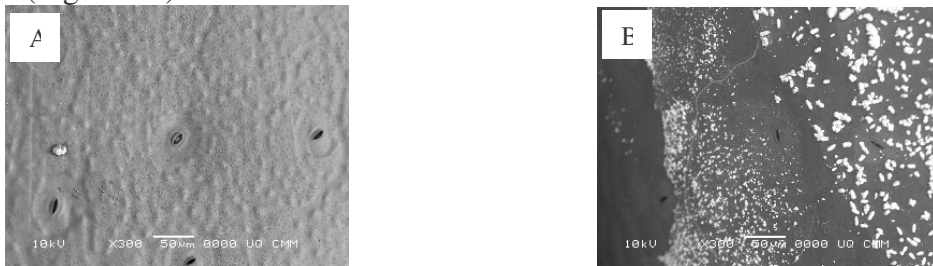


Figure 1. SEM images of *B. delagoense*. (A) control. (B) Starane<sup>®</sup> 200 and Pulse<sup>®</sup> treatment. [Scale bars = 50 µm]

Of the three surfactants tested, Pulse Penetrant<sup>®</sup> most significantly reduced the EST of both Grazon<sup>®</sup> DS and Starane<sup>®</sup> 200, thus it was chosen for use in subsequent pot trials and spray retention studies.

Glasshouse pot trials revealed that species differed significantly in their responses and that the addition of Pulse Penetrant did not necessarily improve the killing effect of Grazon<sup>®</sup> DS and Starane<sup>®</sup> 200. Indeed for *B. delagoense*, Grazon<sup>®</sup> DS in the absence of a surfactant killed more effectively than did Grazon<sup>®</sup> DS+ Pulse Penetrant<sup>®</sup>.

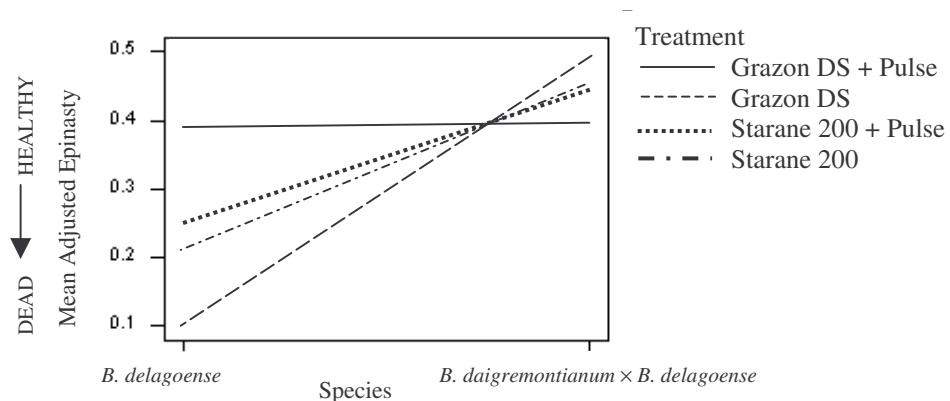


Figure 2. Comparison of symptom-adjusted epinasty means for herbicide × surfactant combinations for *B. delagoense* and *B. daigremontianum* × *B. delagoense*.

Epicuticular wax surface morphology and constituents were similar for both species. Surfactants effectively reduced spray solution surface tension. However, this did not correlate with an increased killing effect, possibly due to the dilution of active ingredients across the leaf surface area as has been reported in analogous studies (Leaper and Holloway 2000).

Despite having similar epicuticular wax composition and surface morphology, *B. delagoense* and the hybrid react differently to herbicide × surfactant treatments. Furthermore, surfactants do not necessarily improve herbicide efficacy in these species. Thus chemical control of *Bryophyllum* spp. is believed to be species-specific, and requires further investigation in the field.

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# LP-333 – a new adjuvant based on linseed oil - increased herbicide activity in fibre flax cultivation

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## Introduction

The history of adjuvant application in plant protection dates back to the beginning of 1970's. A main component of many adjuvants are linseed oil (Nalewaja et. al.1973; 1974). The progress in surfactant synthesis that has taken place since then (Green & Beestman. 2004) has allowed a more effective utilization of linseed oil as an adjuvant improving herbicide performance in agriculture. The aim of the study was to evaluate the use of a new linseed oil based adjuvant. LP-333 for increasing herbicide activity in fiber flax.

## Materials and methods

LP-333 is a new linseed oil based. tank-mix adjuvant formulated at the Institute of Natural Fibers (INF) in Poznań. LP-333 contains 80 % raw linseed oil, 10 % of a non-ion surfactant and 6 and 4 % of two cationic surfactants.

Field trials were carried out in 2003-2004 at the Experimental Farms in Białobrzezie (Dolnośląskie District ) and Sielec Stary (Wielkopolska District) belonging to the Institute of Natural Fibers.

The adjuvant LP-333 was applied at 1.5 l ha<sup>-1</sup> with the following herbicides: chlorsulfuron at 9.0 g ha<sup>-1</sup>. haloxyfop-P at 104 g ha<sup>-1</sup> and quizalofop-P-ethyl at 75 g ha<sup>-1</sup>. The graminicides were applied 6 days after application of chlorsulfuron. Treatments were applied at the 'herring-bone' stage of fiber flax. Weed control and fiber flax crop response to adjuvant LP-333 were assessed.

The predominant weed species in the field trials were: *Chenopodium album*, *Polygonum nodosum*, *Lamium spp.*, *Brassica napus*, *Viola arvensis*, *Amaranthus retroflexus*, *Elymus repens* and *Echinochloa crus-galli*.

## Results and discussion

The adjuvant LP-333 generally improved the performance of the herbicides. Chlorsulfuron applied in mixture with LP-333 controlled effectively the majority of the dicotyledoneous weeds except *Viola arvensis* and *Lamium spp.*. The graminicides haloxyfop-P and quizalofop-P-ethyl applied in mixture with LP-333 showed good effect on the grass species *Elymus repens* and *Echinochloa cruss-galli*. Including the adjuvant LP-333 allowed for a reduction of the doses of chlorsulfuron. haloxyfop-P. quizalofop-P-ethyl by 20-25 % with no loss of efficacy.

Applying the herbicides in mixture with LP-333 did not cause any crop injury. Chlorsulfuron applied in mixture with LP-333 resulted in a significant increase in flax straw and seed yield. Applying the graminicides in mixture with LP-333 had no significant effect on straw and seed yield.

Table 1. Herbicide efficacy in fiber flax (Average of 2 trials).

Treatment (g or L ha <sup>-1</sup> )	Broad- leaved weed biomass control [%]	Grass weed biomass control [%]	Yield of flax [dt ha <sup>-1</sup> ]		Fiber flax susceptibility to herbicides <sup>1</sup>
			Straw	Seed	
Control (weed biomass g m <sup>-2</sup> )	113.5	28.4	62.1	9.5	1
Chlorsulfuron (11.25)	94.7	23.5	63.6	9.9	1
Chlorsulfuron (9.0) + Olbras Super (1.5)	90.6	33.4	62.3	9.4	1
Chlorsulfuron (9.0) + LP-333 (1.5)	93.0	10.2	64.9	10.0	1
Chlorsulfuron (11.25) Haloxypop-P (130)	86.4	90.8	63.3	9.6	1
Chlorsulfuron (11.25) Haloxypop-P (104)+ Olbras Super (1.5)	86.4	79.5	60.4	9.1	1
Chlorsulfuron (11.25) Haloxypop-P (104 + LP-333 (1.5)	93.3	97.8	61.2	9.2	1
Chlorsulfuron (11.25) Quizalofop-P-ethyl (100)	95.3	94.7	63.0	9.4	1
Chlorsulfuron (11.25) Quizalofop-P-ethyl (75) + Olbras Super (1.5)	79.3	94.3	63.6	10.4	1
Chlorsulfuron (11.25) Quizalofop-P-ethyl (75)+ LP-333 (1.5)	92.4	91.4	61.1	9.1	1
LSD (0.05)			2.1	0.5	

<sup>1/</sup> According to a 1-9 scale. where: 1 – no injury. 9 – total damage

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## Challenges and solutions for annual grass weed management in small grain cereals in Europe

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### Introduction

Infestation of annual grass weeds in cereals is a major limitation to successful cereal production not only in Europe but around the globe. The elimination or effective growth reduction including prevention of seed set of the most important competitive grasses infesting cereal crops is essential for producing sustainable and economically viable grain yields and quality in cereals.

The most important cereal crops grown in Europe are wheat and barley. Cereals in many situations and locations remain the economically most sustainable crop for farmers.

Among the key grass species infesting cereals in Europe are *Alopecurus*, *Apera*, *Avena*, *Lolium* and *Phalaris*, the relative importance and occurrence being dependent mainly on geography and soil type.

In general, grass weeds are much more competitive to cereal crops than broadleaved weeds, why it is a very important objective of farmers to achieve near perfect control of grasses.

Several grass herbicides in Syngenta's portfolio contribute to managing the key grass weeds in European cereals. Syngenta's most recent development product is pinoxaden, a new, highly active grass herbicide for use in wheat and barley.

This paper presents the basic properties of pinoxaden and highlights its exciting features for managing grass weeds in cereals.

### Pinoxaden

Pinoxaden is a new selective post-emergence herbicide discovered by Syngenta Crop Protection AG, Basel, Switzerland, being developed for the control of annual grass weeds in various cereal crops, including wheat and barley.

Pinoxaden is a novel grass active compound from the chemical class of phenylpyrazolines. For maximal crop safety, it is formulated with the safener cloquintocet-mexyl. Pinoxaden is taken up primarily through leaves of grasses and then translocated basipetally and acropetally in treated plants. The primary mode-of-action is the inhibition of the Acetyl-coenzyme A carboxylase.

At use rates of 30 - 60 g ai/ha, one of pinoxaden's unique features is its high activity against a broad range of grasses including *Avena spp.*, *Apera sp.v.*, *Alopecurus sp.*, *Lolium spp.* and *Phalaris spp.* as well as several other annual grasses.

Another exciting feature of pinoxaden is its excellent crop safety in wheat and barley without any negative effect on grain yield.

Pinoxaden is recommended to be applied in autumn or spring from the 2-leaf stage up to the flag-leaf stage of weeds. It will be used with a tailored adjuvant providing optimized activity by maximizing the spread on the leaf surface and the uptake into leaves. Pinoxaden is rainfast in 30-60 minutes after application.

Results from replanting studies show that after the application of pinoxaden no limitations on rotational crop plantings apply. Pinoxaden has an excellent toxicological, ecotoxicological, and environmental profile.

Based on its broad grass weed control spectrum, its excellent crop tolerance to several cereal crops, and its wide application window, pinoxaden will set a new and exciting standard in grass weed control in cereals.

## **Outlook**

Pinoxaden offers an exciting new tool to manage grass weeds in cereal crops and as such complements Syngenta's existing cereal herbicide portfolio comprising of clodinafop, tralkoxydim, prosulfocarb, triasulfuron and dicamba.

Pinoxaden will be globally launched, and registrations and first sales are expected from 2006.

## **Acknowledgement**

The authors express their thanks to all those who have contributed to the successful development of pinoxaden.

# The influence of relative humidity on *Anthemis arvensis* and *Stellaria media* control by tribenuron-methyl used alone and with adjuvants

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## Introduction

The activity of foliar-applied herbicides is influenced by weather factors that can modify plant morphology and mechanism of their action: uptake, translocation and degradation in plants. Previous research showed that efficacy of herbicides may be reduced at low humidity conditions due to a decreased uptake and translocation rate (Anderson et al., 1993, Coetzer et al., 2001). Adjuvants may enhance herbicide activity under unfavourable climate condition by improving spray delivery, increasing spray retention on leaves and enhancing foliar penetration (Morton & Harvey, 1994).

The objective of this experiment was to determine the influence of relative humidity on the activity of the sulfonylurea herbicide tribenuron-methyl applied alone and in mixture with adjuvants.

## Material and methods

The activity of tribenuron-methyl applied alone and in mixture with two adjuvants was investigated. Seeds of *Stellaria media* and *Anthemis arvensis* were sown in pots filled with a mixture of peat and sand. After sowing the pots were placed in controlled climate chambers running at 50% and 90% RH. Temperature and light intensity were identical in the two climate chambers (20/10 °C and 250  $\mu\text{mol m}^{-2}\text{s}^{-1}$ ). The plants were sprayed at the 4-leaf stage. Tribenuron-methyl at 7.5 and 15 g a.i.  $\text{ha}^{-1}$  was applied alone or in combination with two adjuvants: 1.5 l  $\text{ha}^{-1}$  Olbras 88 EC (refined fatty acids) and 0.125 l  $\text{ha}^{-1}$  Trend 90 EC (nonionic surfactant). The herbicide was applied using a laboratory sprayer equipped with a mobile TeeJet XR 11003-VS nozzle operated at pressure of 200 kPa. The sprayer was calibrated to deliver a spray volume 250 l  $\text{ha}^{-1}$ .

Fresh weight was determined three weeks after spraying. The experiment was carried out using a completely randomized blocks with three replications.

## Results and discussion

The growth of both *Stellaria media* and *Anthemis arvensis* was greater at 50% RH compared to 90% RH. Comparing the two species, *Stellaria media* produced more biomass than *Anthemis arvensis* and the difference in biomass produced at 50% and 90% RH was also greater.

Tribenuron-methyl completely controlled *Stellaria media* at both RH regimes, irrespectively of tribenuron dose. Effectiveness of the herbicide was not affected by addition of adjuvants. At 50% RH the growth of *Anthemis arvensis* treated with tribenuron-methyl was markedly more reduced when the herbicide was applied in mixture with the adjuvants. Low humidity significantly decreased tribenuron-methyl effectiveness when it was used alone. At the high relative humidity a satisfactory control of *Anthemis arvensis* was achieved at both herbicide doses and addition of the adjuvants did not promote tribenuron-methyl activity.

The present study has shown, that the detrimental effect of low humidity on tribenuron-methyl effectiveness could be overcome by addition of adjuvants to the spray solution, and this finding is in agreement with previous results (Kudsk et. al. 1990). The experiment has shown a significant influence of adjuvants on tribenuron-methyl effect on *Anthemis arvensis*, whereas the activity on *Stellaria media* was not affected by adjuvants. Variations in the influence of adjuvants on different weed species have been reported previously. Mathiassen & Kudsk (1993) reported that adjuvants did not affect *Stellaria media* control by glufosinate, whereas an increase in efficacy was observed on other species following adjuvant addition.

Table 1. The influence of relative humidity on effectiveness of tribenuron-methyl applied alone and in mixture with adjuvants. Figures in parentheses indicate percent reduction in fresh weight compared to the corresponding untreated.

Treatments	Doses	Fresh weight of <i>Anthemis arvensis</i> (g)		Fresh weight of <i>Stellaria media</i> (g)	
		50% RH	90% RH	50% RH	90% RH
Untreated	-	3.69	2.18	9.19	5.33
Tribenuron-methyl	15 g·ha <sup>-1</sup>	1.87 (49)	0.19 (91)	0.09 (99)	0.08 (99)
Tribenuron-methyl	7.5 g·ha <sup>-1</sup>	1.62 (56)	0.19 (91)	0.11 (98)	0.08 (99)
Tribenuron-methyl	15 g·ha <sup>-1</sup> + 0.05 %	0.26 (93)	0.14 (93)	0.07 (99)	0.08 (99)
+ Trend 90 EC	7.5 g·ha <sup>-1</sup> + 0.05 %	0.35 (91)	0.18 (92)	0.07 (99)	0.08 (99)
Tribenuron-methyl	15 g·ha <sup>-1</sup> + 1.5 l·ha <sup>-1</sup>	0.26 (93)	0.16 (93)	0.08 (99)	0.09 (98)
+ Olbras 88 EC	7.5 g·ha <sup>-1</sup> + 1.5 l·ha <sup>-1</sup>	0.35 (90)	0.20 (91)	0.09 (99)	0.09 (98)
LSD (0.05)	adjuvant		0.33		0.08
	dose		0.28		0.07
	relative humidity		0.28		0.07

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# Propoxycarbazone-Na activity as affected by nozzle design and spray volume

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## Introduction

Propoxycarbazone-Na is a new systemic herbicide for post-emergence control of several grasses and some broad-leaved weed species in wheat, rye and triticale (Feucht et al. 1999). The active ingredient is absorbed through the leaves and roots, from where it is transported throughout the plant. The speed of action depends on environmental conditions, increasing with increasing temperature and soil moisture. When early post-emergence application is made in spring, temperature is usually low and soil moisture level mostly adequate, and propoxycarbazone-Na is mainly taken up via the roots. In dry conditions it is advisable to increase leaf absorption by adding appropriate adjuvants (Amann 2002, Fandrich *et. al.* 2001) or applying the herbicide using an appropriate application technique.

It is well known that changes in application technique can affect herbicide performance and spray deposition on plants, but the reasons for this are not always clear. Field studies were conducted to determine the effect of nozzle type, spray quality and spray volume on the activity of propoxycarbazone-Na against oat (*Avena sativa* L.).

## Material and methods

Field experiments in spring wheat were carried out during 2002-2004 at the Experimental Station of Plant Protection Institute in Winna Góra. Treatments were made at the 6- to 8-leaf stage of spring wheat on weeds with 4-6 leaves. Oat (as model weed) represented a grass weed with a difficult-to-wet foliage. Oat was sown at a rate of 50 kg ha<sup>-1</sup> between rows of spring wheat at a distance of 24 cm. A 70% water dispersible granule formulation of propoxycarbazone-Na was used throughout the experiments (Attribut 70 WG, 700 g kg<sup>-1</sup>, Bayer CropScience). The herbicide was applied at rates from 35 to 42 g a.i. ha<sup>-1</sup> in a spray volume of 125 and 250 l ha<sup>-1</sup> using a plot sprayer. In all experiments propoxycarbazone-Na was applied in tank-mixture with the adjuvant Torpedo II (consisting of four ingredients). The nozzles from Spraying Systems<sup>®</sup> included in the study were: TwinJet (TJ), Turbo TeeJet (TT), and AI TeeJet (AI). The AI nozzle is an air inclusion nozzle; TT is a low drift nozzle, whereas TJ is a twin nozzle with two flat spray tips. At a pressure of 300 kPa the nozzles produced a coarse, medium and very fine spray quality, respectively.

The trials were carried out using 16,5 m<sup>2</sup> plots using a completely randomised block design with 4 replications. Herbicidal efficacy was evaluated by recording the fresh weight of treated plants. Oat plants were harvested ca. 5-6 weeks after treatment and percent growth reduction relative to the untreated control was calculated.

## Results and discussion

The results from the field experiments showed that droplet size produced by different nozzle types influenced the performance of propoxycarbazone-Na. Generally, oat control was increased as droplet size decreased at constant spray volume. Droplet size obtained with TJ and TT nozzles enhanced oat control compared to droplet size with the air inclusion nozzle. Earlier research has indicated that weed control was improved for species with waxy leaf surfaces (difficult to wet), when using a nozzle producing smaller droplets (Jensen and Kirknel 1994; Kierzek 2002). The use of very coarse sprays can reduce biological efficacy of

some post-emergence herbicides (Enfalt et al. 1997; Jensen 1999) even if the active ingredient is taken up mainly via the roots, as is the case with propoxycarbazone-Na (Pontzen 2002).

Droplet density (number per unit area) has been suggested to be of major importance considering the effectiveness of foliar applied herbicides. In this study, increasing spray volume rate or reducing droplet size increased droplet density, and generally increased phytotoxicity. The results of this study showed that treatments with volume rate of 250 l ha<sup>-1</sup> were more effective than treatments applied in 125 l ha<sup>-1</sup>, regardless of nozzle type.

Despite the fact that many papers have reported that propoxycarbazone-Na is taken up mainly through the roots, the present results suggest that leaf uptake may also contribute to the overall herbicide activity. Therefore, under unfavourable conditions (e.g. low soil moisture before and after treatment) choosing a proper nozzle will provide a better distribution of the deposit on the leaves and result in a better performance of propoxycarbazone-Na.

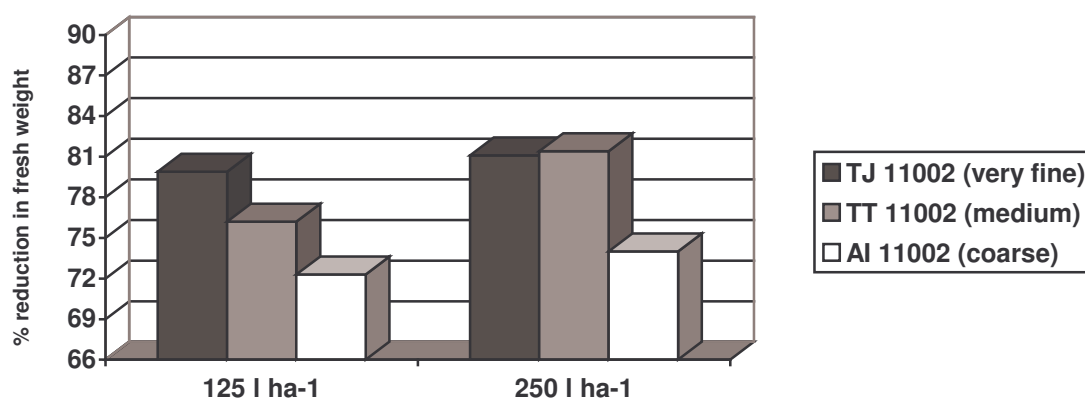


Figure 1. Three-year averages control of oat (*Avena sativa* L.) with propoxycarbazone-Na applied at rates from 35 to 42 g a.i. ha<sup>-1</sup> as affected by nozzle type (spray quality) and spray volume. Values are % reduction in fresh weight as compared with untreated plants.

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## Efficacy of flupyrsulfuron-methyl used for weed control in winter wheat

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### Introduction

The average cultivated area of winter wheat in Poland is 1.9 millions hectares representing above 11% of the total agricultural land. The crop technology is changing and adapting to the new market requirements. This includes growing high quality bread cultivars, which in general are less tolerant to frost (Research Centre for Cultivar Testing, 2003). That leads to plant losses and relatively poor crop survival in severe winter conditions. Such crops produce thinned and open canopies easily infested by weeds germinating in the spring. Hence, the weed spectrum often comprises of species associated with spring cereals, e.g., *Chenopodium album* (CHEAL). This requires the use of herbicides that can control both autumn and spring germinating weeds without negative effect on the crop safety or following crops.

An increase in the number of fields infested by *Alopecurus myosuroides* (ALOMY) is another challenge for weed control in winter wheat in certain regions in Poland. Therefore, the flexible grass herbicide should provide efficacious control not only of the key grass weed – *Apera spica-venti* (APESV), but also *Alopecurus myosuroides* (ALOMY). The key broad-leaved weeds in winter wheat, especially in more intense farming conditions, are *Viola arvensis* (VIOAR), *Galium aparine* (GALAP), *Brassica napus* (BRSNN), *Centaurea cyanus* (CENCY), *Matricaria spp.* (MATSS). Those species are the key target for weed control strategies in winter wheat.

Flupyrsulfuron-methyl is an effective herbicide with foliar and root activity against some grass weeds and a wide range of broadleaved weeds in cereals (Russell et al., 2002). Flupyrsulfuron-methyl is a sulfonylurea herbicide and inhibits the plant enzyme acetolactate synthase (ALS) (Teaney et al., 1995). Early post-emergence application often results in a better effect (Adamczewski & Krawczyk, 2002) and the objective of this investigation was to evaluate the efficacy of flupyrsulfuron-methyl in winter wheat as influenced by application timing in the autumn.

### Materials and methods

Field trials were conducted in the Institute of Plant Protection at the Agricultural Experimental Farm Winna Góra and at the Regional Experimental Station Toruń, during the 2002 – 2004 growing seasons. The experimental design was a completely randomised block with four replications. Herbicide treatments were applied with a CO<sub>2</sub> operated knapsack sprayer. Flupyrsulfuron-methyl (Lexus 50 WG, 500 g a.i. kg<sup>-1</sup>, Du Pont de Nemours), chlorotoluron (Dicuran 80 WP, 800 g a.i. kg<sup>-1</sup>, Syngenta Crop Protection AG or Lentipur Flo 500 SC, 500 g a.i. L<sup>-1</sup>, NUFARM GmbH Co KG) and metribuzin + flufenacet (Expert Met 56 WG, 140+420 g a.i. kg<sup>-1</sup>, Bayer AG) were applied at three timings in the autumn (Table 1). Broadleaved weed control was evaluated at the stem elongation stage of winter wheat and grass weeds control was evaluated at the heading stage of these weeds. Grain yields were taken each year from the whole plots in Winna Góra and from one square meter in Toruń. Yield data were subjected to analysis of variance, and the differences in the mean values were compared by a t-test at a significance level of P<0.05.

## Results and discussion

Weed control efficacy and winter wheat yield are presented in Table 1. The yield of the untreated plot was significantly lower than the herbicides treated plots. Flupyr-sulfuron-methyl provided good control of all broad-leaf weeds and *Apera spica-venti* regardless of the application timing except for *Viola arvensis*, which was well controlled only when the herbicide was applied early post-emergence. Flupyr-sulfuron-methyl was effective against *Alopecurus myosuroides* when applied early or late post-emergence. The spring germinated *Chenopodium album* was well controlled by flupyr-sulfuron-methyl applied in the autumn. The standard herbicides (chlorotoluron and metribuzin+flufenacet) were generally less effective than flupyr-sulfuron-methyl.

In conclusion, flupyr-sulfuron-methyl applied early post emergence provided the most effective weed control compared to pre-emergence treatment or late post emergence treatment. However, optimum herbicide application timing also depends on the composition of the weed flora.

Table 1. Percent weed control following herbicide treatments in the years 2003-2004.

Treatment	Dose g a.i. ha <sup>-1</sup>	Appl. Time							Yield* (t ha <sup>-1</sup> )	
			ALO MY	APES V	BRSN N	VIOA R	CHEA L	GAL AP	2003	2004
Untreated [plants.m <sup>-2</sup> ]		-	193,4	31,3	3,25	17,8	11,5	4,50	4,95	7,98
flupyr-sulfuron -methyl	10	Pre	68,8	100	97,3	63,5	100	-	5,91	-
chlorotoluron	1000	Pre	71,8	100	80,8	28,1	80,0	-	5,89	-
flupyr-sulfuron -methyl	10	E-Post	91,9	100	94,6	87,0	100	100	6,44	8,58
metribuzin+ flufenacet	49 + 147	E-Post	74,1	100	93,0	62,4	80,0	100	5,83	8,82
flupyr-sulfuron -methyl	10	L-Post	95,8	100	95,1	78,7	100	100	6,57	8,77
chlorotoluron	1000	L-Post	84,4	100	72,3	49,4	100	100	6,54	8,73
LSD (0.05%)									0,686	0,460

\*Yield originates only from field experiments at Winna Góra.

Pre: Pre-emergence treatments, after sowing; E-Post: Early Post-emergence until the 1-leaf stage of wheat; L-Post: Post-emergence from the 2-to 3-leaf stage of wheat.

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## Preliminary studies on the potential to reduce metsulfuron-methyl dose

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### Introduction

The competitive ability of wheat is influenced by many factors, e.g. nitrogen supply, weed species, and crop density (Iqbal & Wright, 1997; Melander et al., 2003). Some varieties exhibit earlier ground cover, denser canopy and increased tillering and crop biomass at higher nitrogen rates. Weed ground cover and dry matter following treatment with reduced rate herbicide tended to be lower with higher nitrogen rates (Richards, 1993). Adjuvants improved the effect of spring-applied isoproturon on some weeds (Mathiassen et al., 1993). Mixture of some herbicides with oil adjuvants allowed for herbicide dose reductions up to 50% without negative effect on the yield of winter wheat (Hristov, 1996).

### Material and methods

Field experiments were conducted in two consecutive years 2002/03 and 2003/04 at the Plant Protection Institute in Kostinbrod to investigate the effects of metsulfuron-methyl at 100, 75 and 50% of the recommended dose (6 g a.i. ha<sup>-1</sup>) in winter wheat (cv. Sadovo 1). The herbicide was applied alone and in tank mixture with the plant growth regulator Tritimil (20% phenylftalamino acid + 2% 4-chlorphenoxyacetic acid + 20% chlorcholinchlorid + 1% microelements) and a liquid fertilizer (Humus life universal, containing soil micro organisms, N, P, K, Ca, Mg and others microelements). The experimental design was a randomized complete block design with 3 replications. Individual plots were 2 x 5 m. A nitrogen fertilizer at 100 kg ha<sup>-1</sup> was applied at the end of March. The metsulfuron-methyl treatments were applied in the beginning of April, when broad-leaved weeds were at the 2-3 to 7-8 leaf stages. The dominant weeds are listed in the Table 1. The data were subjected to analysis of variance. An index of crop depression (ID) was calculated based on spike length, grain number per spike and grain weight per spike.  $ID = 1/n(\sum(Pv-Pc)/Pc)$ , where n=3, Pv is the value for the herbicide treatment, and Pc is the corresponding value of the control.

### Results and discussion

Metsulfuron-methyl at the recommended rate had a relatively good effect against annual and perennial broad-leaved weeds (Table 1). The total weed biomass was reduced by 58.8% and 81.6%, respectively. At the 75% dose of metsulfuron-methyl the reduction in fresh weight of annual and perennial weeds were similar to those of full dose. In tank mixture with Humus life and Tritimil the efficacy of metsulfuron-methyl at low rates decreased significantly, especially when applied with Tritimil.

In spite of this trend the yield of winter wheat was higher with metsulfuron-methyl+Tritimil (Table 2). The ID's for Tritimil and metsulfuron+Tritimil showed that Tritimil stimulated plant productivity and allowed for the use of lower doses of metsulfuron-methyl. The reduction of herbicide dose had no negative influence on the yield of winter wheat. On the contrary, the yield was increased compared to the treatments of metsulfuron-methyl alone by 14.1, 16.3 and 34.6% respectively. This could be due to the interspecific competition between winter wheat and weeds and a significant reduction in the density and biomass of *Cirsium arvense* (L.) Scop. The data showed that the reduction of metsulfuron-methyl dose had no significant influence on yield of winter wheat. Domaradzki & Rola (2002) also reported that lowering herbicide doses by 33 to 50% provided satisfactory weed control without significant decrease of winter wheat yield.

Table 1. Efficacy of metsulfuron-methyl on annual and perennial broad-leaved weeds in winter wheat in 2004, 30 days after treatment.

Weeds (number.m <sup>-2</sup> )	Metsulfuron-methyl			Metsulfuron-methyl + 1 L ha <sup>-1</sup> Humus life			Metsulfuron-methyl + 0.3 L ha <sup>-1</sup> Tritimil			
	C	A	B	D	A	B	D	A	B	D
Annual weeds										
<i>Veronica hederifolia</i> L.	72	14	53	110	21	32	84	141	128	126
<i>Viola arvensis</i> Murr.	114	45	163	209	60	113	126	128	120	50
<i>Galium aparine</i> L*	299	142	104	101	103	243	195	193	168	164
<i>Anthemis arvensis</i> L.	9	0	4	2	1	3	0	4	6	0
<i>Fallopia convolvulus</i> (L)	6	3	4	13	28	3	45	1	3	2
<i>Thlaspi arvense</i> L.	1	3	1	11	0	0	3	0	0	0
Others	0	4	0	8	4	3	5	7	0	6
Total weight (g)	557.3	229.3	299.3	477.3	129.3	404.0	396.0	400.0	420.0	456.0
Perennial weeds										
<i>Cirsium arvense</i> (L)	30	21	13	12	4	4	5	9	1	0
<i>Convolvulus arvensis</i> L	0	9	3	9	0	0	21	0	0	0
Total weight (g)	174.7	32.0	26.0	28.0	2.67	6.67	21.0	10.7	1.0	0

\* Number shoots; C =control; A=6 g a.i. ha<sup>-1</sup> metsulfuron-methyl; B=4.5 g a.i. ha<sup>-1</sup> metsulfuron-methyl; D=3 g a.i. ha<sup>-1</sup> metsulfuron-methyl

Table 2. Influence of reduction in metsulfuron-methyl dose on crop growth and yield.

Dose a.i. ha <sup>-1</sup>	Plant height cm	Spike length cm	Grain number per spike	Grain weight per spike	Yield t.ha <sup>-1</sup>
<u>Metsulfuron-methyl</u>					
Untreated	96.76 C	7.75 C	31.90 C	1.54 C	3.22 C
6.0	95.93 NS C	7.69 NS C	34.10 ++ C	1.62 NS C	4.33 +++ C
4.5	95.60 NS NS	7.83 NS NS	33.58 ++ NS	1.57 NS NS	4.23 +++
3.0	93.00 + NS	7.85 NS NS	32.78 + +	1.54 NS NS	3.55 +++ +++
F =	3.320	3.460	7.440	15.040	4.601
SD =	1.920	0.266	0.440	0.016	0.080
<u>Metsulfuron-methyl + 1 L ha<sup>-1</sup> Humus life</u>					
Untreated	101.2 C	7.61 C	30.80 C	1.45 C	3.25 C
6.0	99.2 NS C	7.58 NS C	31.70 + C	1.54 NS C	3.77 ++ C
4.5	98.2 ++ NS	7.73 NS NS	32.52 +++ NS	1.58 NS NS	3.82 +++ NS
3.0	98.3 + NS	7.68 NS NS	32.54 +++ NS	1.55 NS NS	3.72 ++ NS
Humus life alone	100.4 NS NS	7.63 NS NS	31.20 NS NS	1.47 NS NS	3.85 ++ NS
F =	11.360	5.310	18.75	5.660	3.16
SD =	0.773	0.162	0.417	0.048	0.16
<u>Metsulfuron+ 0.3 L ha<sup>-1</sup> Tritimil</u>					
Untreated	104.2 C	7.64 C	31.98 C	1.55 C	4.44 C
6.0	104.7 NS C	7.73 NS C	33.42 + C	1.58 NS C	4.94 +++ C
4.5	102.6 NS NS	7.65 NS NS	33.33 NS NS	1.57 NS NS	4.92 +++ NS
3.0	100.3 NS NS	7.99 NS NS	34.53 ++ NS	1.67 NS NS	4.78 ++ NS
Tritimil alone	100.5 NS NS	7.64 NS NS	33.20 NS NS	1.63 NS NS	4.19 + +++
F =	15.78	13.92	6.050	11.82	3.29
SD =	0.899	0.042	0.683	0.097	0.069

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# Influence of the growth regulator trinexapac-ethyl on winter rye under different nitrogen fertilization

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## Introduction

Winter rye is very susceptible to lodging especially under favorable growing conditions such as high nitrogen fertilization (Maciorowski et al. 2000. Ilumae 2002). Lodging mainly occurs when the foliage grows vigorously and the root system is not well developed. In high input agriculture application of plant growth regulators (PGR) has become common practice to prevent lodging. Trinexapac-ethyl is a growth regulator and its main area of action is reducing crop height and consequently preventing lodging and improving harvest index and yield of cereals (Amrein et al. 1989. Kerber et al. 1989). The plant growth regulator trinexapac-ethyl belongs to the cyclohexanedione group and is taken up by the foliage and translocated to the growing shoot, where it inhibits internode elongation (Kerber et al. 1989).

The aim of the experiments was to study the influence of trinexapac-ethyl on morphological traits of winter rye (stem, ear and internodes length, stem diameter chlorophyll content and yield under different nitrogen regimes).

## Material and methods

Biological activity of trinexapac-ethyl (Moddus 250 EC, 250 g L<sup>-1</sup> trinexapac-ethyl, Syngenta) was studied in the years 1999-2003 in field experiments at the Experimental Station in Winna Góra belonging to the Plant Protection Institute in Poznan. The experimental design was a randomized complete block with four replications. Plot size was 16.5 m<sup>2</sup>. Both diploid and hybrid varieties of winter rye was used. Trinexapac-ethyl was applied at the 2-node stage (BBCH 32) at two doses (75 and 125 g a.i.ha<sup>-1</sup>) and at 50 g a.i.ha<sup>-1</sup> in mixture with 675 g a.i. ha<sup>-1</sup> chlormequat-chloride (CCC) at three nitrogen fertilization levels (0, 80 kg ha<sup>-1</sup> applied at the beginning of vegetation and 160 kg ha<sup>-1</sup> applied as 80 kg ha<sup>-1</sup> at the beginning of vegetation and 80 kg ha<sup>-1</sup> during shooting). Trinexapac-ethyl was applied with a Gloria plot sprayer equipped with R 110-03 flat fan nozzles in a spray volume of 200 l/ha using a nozzle pressure of 200 kPa.

Two, three and four weeks after application the chlorophyll content of the flag leaf was measured using the SPAD method and plant height was measured. Lodging was assessed visually at growth stage BBCH 89 using a 1-9 scale where 1 is 100% lodging and 9 is no lodging. Before harvest 25 plants were collected at random from each plot for morphological measurements: stem, ear and internodes length, stem diameter between 3<sup>rd</sup> and 4<sup>th</sup> internodes and number of grains per ear. The yield was taken with a small plot combiner (Wintersteiger). Thousand-kernel weight (TKW) was determined in the laboratory (3 x 250 grains from each plot). Protein content was determined using Near Infrared Radiation (NIR) refraction at 1400-2500 nm (Analyser Informatic 8100 Parten Co).

## Results and discussion

In Poland about 1.5 mill hectares of winter rye are cultivated. Recently hybrid cultivars have become more popular. These cultivars are higher yielding at high nitrogen fertilizer levels, but also more susceptible to lodging compared to diploid cultivars. According to other authors trinexapac-ethyl can be used in winter rye to reduce lodging (Kerber et al. 1989. Hafner 2001. Ilumae 2002).

The results of four years of field experiments at our institute showed that trinexapac-ethyl is an effective plant growth regulator in winter rye. Some of the results obtained are presented in Table 1. Chlorophyll content measurement showed the significant influence of nitrogen fertilization and plant growth regulator treatments. In the no nitrogen treatment plant growth regulators increased chlorophyll content more than in plots with nitrogen application. Trinexapac-ethyl, as well as nitrogen fertilization and environmental factors, influenced crop growth. In the very dry 2003 growing season the canopy height was lower than in 2002, a year with average rainfall. The growth of the diploid cultivars was influenced by both nitrogen fertilization and the PGRs, while the growth of hybrid cultivars were only influenced by the PGRs.

Crop lodging was noted only in wet growing seasons and in nitrogen treated plots. Trinexapac-ethyl used alone and with CCC restrained lodging but did not prevent it completely. Internode measurement indicated no major differences between the PGR treatments and larger differences were only recorded for the 3<sup>rd</sup> and 4<sup>th</sup> internodes. Measurement of stem diameters revealed that no differences were found for the 3<sup>rd</sup> and 4<sup>th</sup> internodes. TKW and protein content was higher in plots with nitrogen fertilization and application of PGRs did not influence these parameters. Grain yield was higher after nitrogen fertilization and application of PGRs increased yield.

In conclusion, the results obtained from the field experiments showed a very strong effect of trinexapac-ethyl use alone or in mixture with CCC on winter rye especially on hybrid cultivars.

Table 1. Influence of trinexapac-ethyl use alone and with CCC on diploid winter rye.

N kg ha <sup>-1</sup>	Trinexapac-ethyl g ha <sup>-1</sup>	Chlorophyll content (SPAD)	Plant height cm	Lodging in scale 1-9	TGW (g)	Protein content (%)	Yield t ha <sup>-1</sup>
0	Check	532	128	9	34.4	12.5	6.26
	75	553	108	9	34.2	12.5	7.33
	125	579	115	9	35.5	12.3	7.34
	50+675 (CCC)	567	111	9	34.6	12.5	7.32
80	Check	560	131	6	36.6	13.2	8.28
	75	576	115	7	37.2	13.3	8.40
	125	604	112	8	36.6	13.4	8.53
	50+675 (CCC)	583	114	8	37.0	13.2	8.69
160	Check	605	130	3	36.8	14.0	8.25
	75	626	112	5	38.0	13.7	8.52
80+80)	125	648	108	6	37.4	13.6	8.53
	50+675 (CCC)	648	112	6	37.6	13.7	8.75
LSD	(0.05%)	n.d.	8.69	-	n.d.	n.d.	0601

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## **Aminopyralid: A new selective herbicide for broadleaf weeds control in cereals**

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### **Introduction**

Aminopyralid (code name DE-750) is a new pyridine carboxylic acid herbicide under development by Dow AgroSciences for use in cereals around the world. Aminopyralid provides post-emergence broadleaf weed control in cereals. Aminopyralid is a selective systemic herbicide, absorbed rapidly by the roots and leaves and translocated both acropetally and basipetally, accumulating in new growth. Symptoms are typical for auxinic mode of action: twisting, curvature and uneven cell growth, leading to plant death. Aminopyralid produces no significant soil or water metabolites except CO<sub>2</sub> and exhibits very low acute and chronic toxicity (practically nontoxic) to mammals, birds, fish, and aquatic invertebrates. Aminopyralid is only slightly toxic to algae and aquatic vascular plants and degrades rapidly in water by photolysis, with a water half-life of 0.6-days.

A key feature of aminopyralid for the Mediterranean region is the excellent post-emergence control of *Papaver rhoeas*, including ALS resistant and 2,4-D tolerant biotypes, at a rate as low as 10 g a.i. ha<sup>-1</sup>. Cereals tolerance to aminopyralid has been excellent and aminopyralid may be applied from BBCH 13 to BBCH 31. In Europe, aminopyralid will be marketed in combination with florasulam. When applied at 10 to 15 g total active substance per hectare, that combination provides excellent control of the most economically important broadleaf weeds of the European cereals market.

### **Materials and methods**

Field trials were carried out in the major South European cereals growing countries from 2001 to 2004 to evaluate the efficacy, crop safety, window of application and tank mix compatibility of aminopyralid applied alone and in combination with florasulam.

Field trials were designed as randomized small plot trials, with 4 replicates and plot size of 20 to 30 m<sup>2</sup>. Applications were made either with a typical knapsack sprayer or with compressed gas backpack. Spray volumes varied from 150 to 300 l ha<sup>-1</sup>. Trials were visually evaluated for crop tolerance and weed control using the standard 0-100 % scale. In addition, yields were measured in all crop safety studies. Aminopyralid and aminopyralid + florasulam were formulated as water dispersible granules (WG) for these trials.

### **Results and discussion**

This field trial program demonstrated that aminopyralid + florasulam applied at 10 + 5 g active substance per hectare is a wide spectrum broadleaf weeds herbicide providing control of most economically important broadleaf weeds of the European cereals market,

including *Galium aparine*, *Anthemis arvensis*, *Matricaria spp*, *Stellaria media*, *Papaver rhoeas* (including resistant biotypes), *Cirsium arvense*, *Polygonum convolvulus*, *Polygonum aviculare*, *Sylibum marianum*, *Chrysanthemum coronarium*, *Chrysanthemum segetum*, *Sinapis arvensis*, *Raphanus raphanistrum*, *Bifora radians* and *Vicia sativa*. See results presented in table 1.

Crop safety trials demonstrated also that cereals tolerance to aminopyralid + florasulam is excellent and that such combination may be applied from BBCH 13 to BBCH 31.

Table 1. Efficacy of aminopyralid + florasulam against broadleaf weeds in South Europe ( field data from 2001 to 2004).

Weed species	Number of locations	% control of aminopyralid + florasulam @ 10+5 g a.i. ha <sup>-1</sup>	% control of tribenuron @ 15 g a.i. ha <sup>-1</sup>
ANTAR	7	94.5	81.4
BIFRA	6	90.1	88.7
CHYCO	2	89.9	84.8
CHYSE	4	76.5	49.9
CIRAR	12	92.9	91.1
GALAP	14	94.5	27.5
MATSS	7	99.1	93.5
PAPRH	22	93.0	89.3
(susceptible)			
PAPRH (ALS resistant)	14	96.9	25.4
PAPRH (2,4-D resistant)	8	97.4	91.4
PAPRH (ALS & 2,4-D resistant)	7	97.2	28.3
POLAV	2	93.6	74.4
SINAR	3	95.6	97.0
STEME	2	99	98

## Preliminary results on the control of *Orobanche ramosa* L. with glyphosate in tomato

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### Introduction

*Orobanche ramosa* L. (broomrape) is distributed mainly in the Mediterranean region, North Africa, and Asia. It is a parasitic species that infests plants belonging to many botanical families (Linke *et al.*, 1989). Severe yield quality and quantity reductions are caused to the host plants. Particularly in tomato, yield losses can reach the 25% as found by Fracchiolla and Boari (2000) or the 75% of total yield according to Hodosy (1981). *O. ramosa* is able to produce a large amount of seeds that are very small and remain viable in the soil for many years even in the absence of the host (Holm *et al.*, 1997). The seeds germinate in response to specific host root exudates; they form *haustoria* that are able to penetrate the root up to the vascular cylinder. In this way, this chlorophyll-lacking holoparasite can interfere with water and mineral intake in the host plant (Parker and Riches, 1993). Due to this ability, the control of broomrape is very difficult and any method (chemical, physical, biological, mechanical) is limited in effectiveness (Quasem, 1998; Boari and Vurro, 2004). Chemical control has been investigated in several host crops such as favabean (Miccolis and Montemurro, 1999), potato (Goldwasser *et al.*, 2001) and tomato (Foy *et al.* 1989; Nandula 1998).

In glasshouse grown tomato, Quasem (l.c.) evaluated the activity of different doses of fifteen herbicides in controlling this species. Among the active ingredients tried, chlorsulfuron, pronamide and pendimethalin gave the best results. However, they were phytotoxic for tomato plants. Up to now also in tomato no selectivity could be observed. The aim of this work was to evaluate, under field conditions, the effectiveness of glyphosate and its selectivity for the processing tomato crop. Particularly, the combined effects of different doses and numbers of applications were investigated.

### Materials and methods

Experiments were conducted in a field naturally infested by *O. ramosa* and located in Altamura (Apulia - Southern Italy). Processing tomato (cv *Genius*) was transplanted, on 20<sup>th</sup> May 2003, in double rows (40 cm between rows within the double row, 120 cm between double-rows, and 35 cm for plants within a row). Crop was irrigated with drip system and grown according to the standard practices of the area.

Four doses of glyphosate (9 – 18 - 36 and 54 g of a.i. in 400 l ha<sup>-1</sup> of water) were used and each of them was applied 1 – 2 – 3 and 4 times. First treatment was made as soon as first shoots emerged (27<sup>th</sup> July) whereas the other treatments were planned with a weekly timing. Experimental plots (1 double rows/plot and 3 m in length) were arranged in a split-plot design with four replications, assuming herbicide rate as main factor. In each plot, the following data were collected: a) number of parasitized plants; b) average number and dry weight of shoots per plant; c) phytotoxicity symptoms on the plants, evaluated according to the EWRS scale ranging from 1(no symptoms) to 9 (dead plant) (Vercesi, 1983). All data were subjected to analysis of variance and Duncan's test was performed to determine significant differences between means.

### Results and Discussion

Effect of the number of treatments: As shown in table 1, the reduction of parasitized plants was statistically higher in the plots treated 4 and 3 times than in the others. In these plots, a percentage of 73.9 and 59.1 %, respectively, was calculated. The reduction of the number of

shoots per plant in plots treated 4 times was the highest (95.7%) but significantly different only from the values recorded in the plots sprayed 1 and 2 times. Shoot dry weight reduction was significantly higher in plots with 4 treatments (97.7%) than in the plots treated 1 time (85.9%). Effect of the herbicide dose: The reduction of parasitized plants ranged from 59.4% to 44.2%. The number of shoots/plant was reduced from the 91.3% to the 86% whereas the reduction of shoot dry weight per plant ranged between 96.3% and 90.2%. However, statistical analysis gave no significant difference within all data. Phytotoxicity effects: Phytotoxicity effects were rated equal to 3 (EWRS scale) in the plots sprayed 4 times at the highest dose, even though they disappeared within fifteen days. In the other plots no phytotoxicity symptom was recorded. Interaction between number of treatments and glyphosate dose: No significant interaction was found between the number of treatments and the herbicide doses used in the trial.

In conclusion, glyphosate showed good effectiveness and acceptable tomato crop selectivity in the control of *O. ramosa*. All treatments reduced both the number of parasitized plants and the amount of shoots per plant. More trials are needed to find optimal a.i. rates, timing and number of applications. These preliminary results suggest that the herbicide was particularly effective when it was applied 3 or 4 times, regardless of the application rate.

Table 1 – Effect of the number of glyphosate applications on *Orobanche ramosa* infestation

Number of treatments	Reduction of the infestation (%) (*)		
	Parasitized plants	Shoots/plant	
		number	dry weight
4	73.9 a	95.7 a	97.7 a
3	59.1 a	91.9 ab	95.6 ab
2	33.7 b	85.9 b	91.6 ab
1	30.8 b	85.1 b	85.9 b

(\*) Data are calculated with respect to the control. Means followed by the same letter, within each column, do not significantly differ at the 5% level (Duncan's test).

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# **DECID'Herb, a decision support system on the WEB, designed for sustainable weed management in cultivated fields**

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## **Introduction**

Most Decision Support Systems (DSS) dedicated to weed management developed in the late XX<sup>th</sup> century were based on a comparison of the short-term economic return of the various management options available (e.g. Kwon et al., 1995; Stigliani & Resina, 1993). They sometimes included a function related to the delayed costs due to the seed production from weeds that escape the control measures. Indeed, theoretical analysis of weed control strategies indicated that sound decision rules could not be based on the basic threshold concept because of the long term consequences of control options (Munier-Jolain et al., 2002). Few DSS were designed specifically for the reduction of environmental impacts so far. PC-Plant Protection, the Danish DSS designed for this purpose is based on adjustments of efficacy targets ascribed to herbicide applications, but do not take into account the ecotoxicological properties of the various herbicides available (Rhydahl, 1993).

In contrast to previous DSSs, DECID'Herb does not offer to optimise a given variable, but it accounts for the multicriteria nature of the decision process. Indeed, if a given field is infested by only a very low abundance of weakly competitive weed species, the 'best' decision would be the 'no treatment' option, to save the cost of the treatment, to save the time of application and to avoid any possible environmental impact. But if any available technical option would be very efficient, free and easy to use, and without any risk for the environment, then the 'best' option would become this one, that would reduce the risk that the few weeds produce seeds that may give rise to later management problems within a few years. This simple example shows us that the decision problem is not an optimization issue, but the point is to weight different criteria that are not easy to compare, among which the efficiency against the infesting weeds, the financial cost, the potential environmental impact, and others...

## **Presentation of the Decision Support System**

DECID'Herb is a WEB software that may be used by a farmer or any advisor that has to make sound decisions at the field scale. It is designed to help decision-making in the framework of Integrated Weed Management with reduced impact on the environment. The DSS suggests a list of 'good' management programmes for a given field from the harvesting of the previous crop to the harvesting of the current crop (Figure 1). The main inputs necessary to make the decision are:

- the field physical properties that might affect the risks of runoff and leaching, and therefore the contamination of surface and ground water after the application of an herbicide. The potential environmental impact of each available herbicide option is then assessed using the indicator developed by van der Werf & Zimmer (1998).
- the crop species, its phenological stage and canopy development at the date of decision;
- the crops grown and herbicides used during the four previous years; this information is used for the assessment of the risk for selecting populations resistant to any herbicides or group of herbicides.
- the cropping system (crop sequence and soil tillage options) scheduled for the following years;
- the weed species observed at the date of decision (post-emergence decision) or expected to emerge (pre-emergence decision) and an assessment of their abundance.

- the availability of the field and the farmer for weed control operation during the cropping season, as a function of the climate, the soil type and the labour scheduling at the whole farm level.

The software includes two sub-models. The first sub-model provides an assessment of the risk associated with each weed species listed. This risk depends on both the risk for yield loss in the current crop, and the risk for an increase in infestation levels in the future crops. Those risks are estimated following expert knowledge about weed biology and weed populations' behaviour as a function of the cropping system. The expert knowledge is simulated using a fuzzy logic system. The second sub-model uses a multicriteria method to select five 'good' options among a list of control programmes. The criteria considered are the following: (i) the expected efficiency of each option on the listed weed species, with higher weights given for the species with higher risk values according to the first sub-model, (ii) the cost of each option, (iv) its potential environmental impacts, (iv) the risk for selecting herbicide resistant biotypes, and (v) the timetable of the operations in the labour scheduling at the farm level. The multicriteria method used is an interactive one: the user defines an expected cost for the whole programme, and determines the weight he ascribes to the environmental issue. If no 'good' option is found on a multicriteria basis, the user is asked to change his choices, either by accepting a higher cost, by reducing the importance of the environment, or by changing the scheduled cropping system.

The screenshot shows the 'Classement des programmes' (Ranking of programs) page in the DECID'Herb software. It features a table of suggested programs and two interactive panels for adjusting parameters and criteria.

N°	Programme herbicide	Matière(s) active(s)	Efficacité (entre 0 et 10)	I <sub>phy</sub> (entre 0 et 10)	Nombre de passage	Applicabilité (entre 0 et 10)	Risque de Résistance (entre 0 et 10)	Coût (€/ha)
1	Caméa 38g/ha + Energy Puma (Brouac, Dulse) 1l/ha	tribénuron-méthyl(B) + fénoxaprop-P-éthyl(A), méfenpyr-diéthyl(C)	9,25	9,88	1	10,00	0,00	53,56
2	Allié 38g/ha + Energy Puma (Brouac, Dulse) 1l/ha	metasulfuron-méthyl(B) + fénoxaprop-P-éthyl(A), méfenpyr-diéthyl(C)	9,38	7,63	1	10,00	0,00	54,18
3	Harmonie extra 50g/ha + Energy Puma (Brouac, Dulse) 1l/ha	thifensulfuron-méthyl(B), tribénuron-méthyl(B) + fénoxaprop-P-éthyl(A), méfenpyr-diéthyl(C)	9,26	9,78	1	10,00	0,00	54,42
4	Caméa 38g/ha + Lesau Villenon 109g/ha	tribénuron-méthyl(B) + flupyr-sulfuron-méthyl(B), thifensulfuron-méthyl(B)	9,07	7,45	1	10,00	0,40	53,14
5	Lesau Villenon 109g/ha	flupyr-sulfuron-méthyl(B), thifensulfuron-méthyl(B)	9,03	7,45	1	10,00	0,40	34,27

Below the table, there are two panels for adjusting parameters and criteria:

- Modifier les paramètres:**
  - Priorité à l'environnement: importante
  - Coût souhaité du désherbage: 50 €/ha
  - Nombre maximal d'interventions souhaité: 1, 2, 2-3, 3 ou plus
  - Type de système de culture souhaité: Protection intégrée, Protection raisonnée
- Modifier les critères:**
  - coût (uniquement du ou des herbicide(s))
  - efficacité
  - applicabilité (contraintes organisationnelles)
  - risque écotoxicologique (I<sub>phy</sub>)
  - risque de sélection de résistance

Figure 1. Output page of DECID'Herb: list of suggested weed control programs. The user may change the decision criteria and run again if the suggested programs are not satisfying.

## Discussion and conclusion

A first version of the software is currently being tested. It still should be improved by aggregating some more expert knowledge to extend its validity to a larger range of situations. It may already be used as a tool for the experimental testing of cropping systems designed to reduce environmental impacts following the principles of Integrated Weed Management.

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# Evaluation of the weed module in the Danish decision support system “Crop Protection Online” adapted to Norwegian conditions.

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## Introduction

The Norwegian decision support system (DSS) named “VIPS”, financed through the national action plan to reduce pesticide use, has no weed module. Herbicides in cereals constitute by far the largest volume of the pesticides used in Norway; hence it was decided to develop a DSS for weed. We were aware of the web-based weed module in the Danish DSS named “Crop Protection Online” (CPO) and we contracted a three-year project with the Danish Institute of Agricultural Sciences with the aim to adapt the system to Norwegian conditions. Field data from the official efficacy testing of herbicides in cereals in Norway supplemented by relevant Danish data are used in the system. The DSS is also customized according to the legislation and agricultural practice in Norway. The aim of this study was to evaluate three prototypes of the system in field trials in spring cereals.

## Materials and methods

Rydahl (2003) and Kudsk (1999) have described the weed module in CPO. The Norwegian prototypes have three efficacy target levels: medium, low (medium - 15 %) and high (medium + 15 %), but never below 50 and never above 97 % effect. Medium level was the efficacy level assumed to be required in spring cereals. Weed density, recorded before spraying by counting weeds in 8 squares (0,25 m<sup>2</sup>each) placed randomly, was used as input data in the system. Additional inputs were crop species, expected yield level, temperature, drought stress and growth stage of weeds and crop. In the field trials, the two first ranked recommendations at each target effect level, selected on basis of the costs of treatment, were included in the experiment. The trial design was a randomised block with three replicates. Seven trials were carried out in spring barley in 2003. In 2004 one spring oat, five spring wheat and three spring barley trials were conducted. The effect of the treatments were recorded by counting the number of weeds, assessing weed coverage 4-5 weeks after application and assessing the coverage again just before harvest. Yield was also recorded. The treatments chosen by the prototypes were expressed in percent of the labelled dose of the herbicide(s).

## Results and discussion

The dose rates of the treatments recommended by the medium efficacy prototype varied from 30 to 100% of the labelled dose (Figure 1). On average the dose was 38% lower than the labelled dose, however there was no significant difference in weed control compared to high efficacy prototype (Table 1). One of the 100% dose incidents was due to drought stress and the other to the occurrence of *Galium aparine* and large plants of volunteer oil seed rape. Use of the high efficacy prototype resulted in no reduction of the herbicide doses. The treatments selected by the low efficacy prototype showed an average herbicide reduction of 75% compared to the labelled doses resulting in a significant reduced weed control compared to the other prototypes. Weed coverage at harvest, however did not vary significantly between prototypes and did not exceed 6% compared to 20% in the untreated plots. There were no significant yield differences between the prototypes in spring wheat and barley. The results from the validation trials have shown that there is no risk associated with the use of the medium efficacy prototype in spring cereals. The low efficacy prototype seems to be safe

regarding the yield, but the weed effect is probably not satisfying and some seed dispersal can be expected. In cereal mono cropping systems, however, limited seed dispersal could be acceptable.

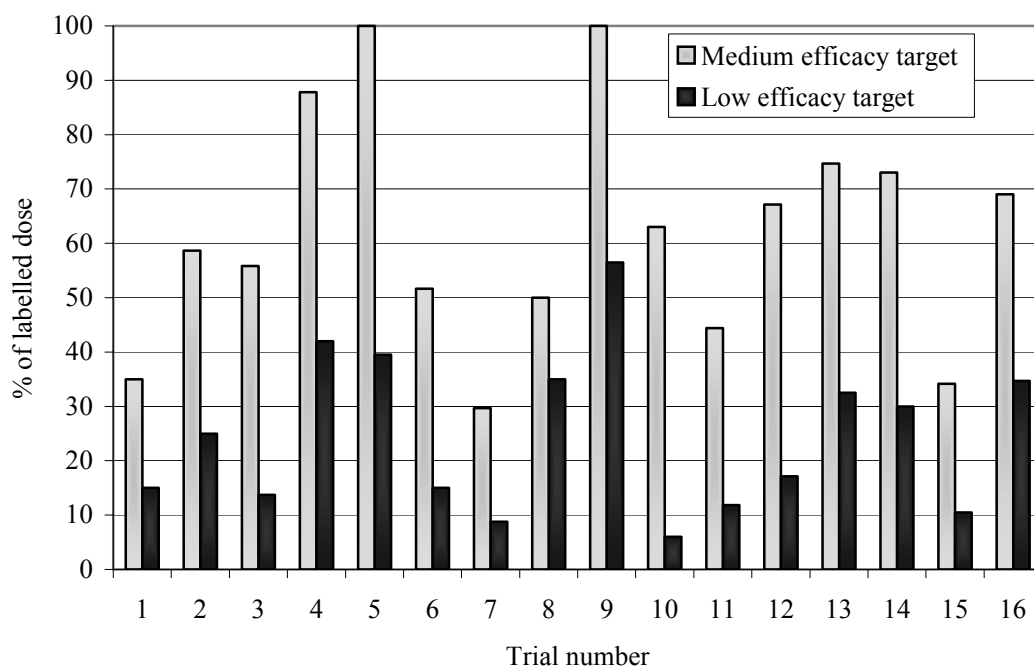


Figure 1. The treatment options from the medium and low efficacy prototypes calculated in percent of the standard labelled dose (average of two choices).

Table 1. Effect on weeds and yield of the herbicide mixtures and doses recommended by the prototypes. Numbers in brackets are number of trials.

Weed species	Efficacy target							LSD 5%	
	Untreated	High		Medium		Low			
	Number of weeds per m <sup>2</sup>	First choice	Second choice	First choice	Second choice	First choice	Second choice		
		<u>% weed control, 3-4 WAT</u>							
STEME (8)	91	89	92	89	81	77	74	18	
GAESS (5)	87	94	93	96	92	73	69	19	
SPRAR (5)	44	91	98	90	97	62	89	21	
Total weed number(16)	137	88	86	84	84	62	65	14	
		<u>% weed coverage</u>							
3-4 WAT (9)	21	4	3	5	6	9	8	3	
At harvest (7)	20	2	2	2	3	6	6	5	
		<u>Yield (t ha<sup>-1</sup>)</u>							
Spring barley (10)	4.76	5.03	4.94	4.96	4.91	4.93	5.05	0.21	
Spring wheat (5)	5.41	6.11	6.15	6.22	5.9	5.92	5.96	0.43	

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# THE RESPONSE OF *ECHINOCHLOA CRUS-GALLI* AND DRILLED ONION TO TANK-MIXTURE OF GRAMINICIDES WITH OXYFLUORFEN

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## Introduction

The broadleaved and grass weeds have commonly infested the drilled onion (*Allium cepa* L.) crops under Polish climate conditions. Among the monocotyledons species an important role plays *Echinochloa crus-galli* (L.) P. BEAUV, which usually appears in large amounts from the end of May and beginning of June and could determine a threat for onion plantations. The drilled onion is recognized as a poor competitor, and weeds emerging throughout the vegetation period will cause the yield losses. The weeds competition is highly risky especially early after emergence period of onion. Pendimethalin and propachlor are considered as the basic herbicides for post-sowing and postemergence application in onion (Boulton et al, 2000; Palczynski et al, 2001). Oxyfluorfen is rated also as an important herbicide for postemergence application. This herbicide could be used at low rates as a split application method from early onion growth stages, as well as some graminicides (Dobrzanski et al, 1995). Under practical conditions, the optimum timing of oxyfluorfen application often coincides with graminicides use. Therefore, the possibility of using these two different substances for controlling both, broadleaved and grass weeds at one treatment would be advisable. It is also well known that some of graminicides applied together with broadleaved weed herbicides can change their biological activity. Wanamarta et al (1993) among others described the antagonism between sethoxydim and bentazon. Matysiak and Nalewaja (1999) confirmed that bentazon restrains the absorption of sethoxydim, which causes antagonistic effects between these herbicides. Holshouser and Coble (1990) concluded that the most tank mixtures of graminicides and broadleaved herbicides reduced the grass control. The purpose of the presented experiment was to assess the behavior of the grass weed (*Echinochloa crus-galli*) and drilled onion on the tank mixture of some graminicides with the broadleaved herbicide oxyfluorfen.

## Materials and methods

The studies were undertaken in 2002-2004, at the Research Institute of Vegetable Crops in Skierniewice, on pseudopodsolic soil over loamy sand [1.5% OM; pH (H<sub>2</sub>O) – 6.5]. The trial was designed in a randomized block in four replications and plot size of 12.2 m<sup>2</sup>. The onion seeds (*Blonska* cv.) were sown 15-16<sup>th</sup> of April each year. Two days after sowing, the whole area was treated with propachlor (4800 g·ha<sup>-1</sup>), and 2-3 days before onion emergence glyphosate (540 g·ha<sup>-1</sup>). After onion emergence (1.5-2 leaves stage) oxyfluorfen 24 g·ha<sup>-1</sup> (two times of split application) was applied. Then, at 2-3 onion leaves stage and *Echinochloa crus-galli* 2-4-6 leaves, the tank-mixtures of oxyfluorfen (Goal 240 EC) at rate 60 g·ha<sup>-1</sup> with the following graminicides: haloxyfop-R (Perenal 104 EC), quizalofop-P-ethyl (Leopard 05EC, sethoxydim (Nabu 45 EC) and quizalofop-P-tefuryl (Pantera 040 EC) were applied. The standard was a separate treatment of quizalofop-P-ethyl and oxyfluorfen. The boom plot sprayer with fan nozzles Tee-Jet: DG 11002 VS, at the pressure 400 kPa and 200 L·ha<sup>-1</sup> water volume was used. *Echinochloa crus-galli* control was estimated in percent 14, 21 and 29 days after application (DAT) and the crop phytotoxicity level were evaluated after 14 and 30 DAT. The onion yield data was subjected to an analysis of variance. Means were compared using Newman-Keul's test at significance level P= 0.05.



## Results and discussion

The presented results clearly indicate the very successful *Echinochloa crus-galli* (ECHCG) control using the tank mixtures of oxyfluorfen with almost all graminicides in comparison to the standard treatment - quizalofop-ethyl applied alone (Table 1). The first estimation (14 DAT) and the following ones (21 and 29 DAT) showed the very close biological activity and comparable dynamics decay of *Echinochloa crus-galli* process at all treatments, with the exception of tank-mixture of quizalofop-P-ethyl (50 g $\text{ha}^{-1}$ ) + oxyfluorfen (60 g $\text{ha}^{-1}$ ). The ECHCG control level were presented relatively: 2002 – 97% (14 DAT) and 100% (21 and 29 DAT); 2003 – 51, 52 and 48%; 2004 - 87, 91 and 88%. It means that in years 2003 and 2004 the antagonism between quizalofop-P-ethyl and oxyfluorfen was clearly noticed. The reason for this antagonism was probably the long period of drought during vegetation season of 2003 and 2004. These factors changed morphological appearance of ECHCG leaves and caused a layer of wax on them. The greenhouse experiments conducted in 2004 confirmed such antagonism between these chemical substances. In 2003 only the slightly decrease of ECHCG control was also observed at tank-mixture of haloxyfop-R + oxyfluorfen.

Table 1. The *Echinochloa crus-galli* response to tank-mixture of graminicides with oxyfluorfen in drilled onion

Herbicides	Rate g $\text{ha}^{-1}$	ECHCG control in %			Phytotoxicity to onion in %		Marketable yield in t $\text{ha}^{-1}$
		(from - to)			14 DAT	30 DAT	
		14 DAT	21 DAT	29 DAT			
haloxyfop-R+oxyf.	52+60	85-98	80-100	66-100	5.6	0.8	34.8
quizalofop-P-ethyl+ oxyf.	50+60	51-87	53-100	48-100	6.2	0.7	34.9
sethoxydim+oxyfluorfen.	450+60	95-98	96-100	96-100	5.6	0.7	35.9
quizalofop-P-tefuryl+oxyfl.	60+60	96-99	97-100	96-100	7.4	0.8	36.5
quizalofop-P-ethyl	50	93-98	95-100	90-100	0.2	0	36.7
oxyfluorfen	60	21-45	8-18	x	6.8	0.8	32.3
Check	-	0	0	0	0	0	29.6

oxyf. = oxyfluorfen; x-plots were weeded after 2<sup>nd</sup> evaluation; DAT-days after treatment; ECHCG coverage at check plots during first evaluation: 2002=33%; 2003=26%; 2004=48%;

The phytotoxicity to onions clearly did not increase due to graminicides addition to oxyfluorfen. The phytotoxic effects observed on onion were well known symptoms caused by oxyfluorfen, appearing usually after this herbicide application.

The marketable yield of onion was diverse depending on variable conditions in years. Onion yielding at all the treatments was relatively at the same level, and slightly differed in relationship to the weed control efficacy, performed by the treatments.

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# Influence of soil applied herbicides on soyabean yield structure under different agrotechniques

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## Introduction

Soyabean growth and yield structure depends on different factors: the variety, soil and air temperature and humidity, light intensity, fertilization, agrotechniques (Perestova, 1984; Chalyi and Perestova, 1984; Terenteva et al., 1984; Myakushko et al., 1984) as well as on the applied herbicides (Peneva, 1994, 2002, 2003), according to their mode of action (Schmidt, 1999). The aim of this study was to determine the effect of several soil herbicides applied alone or in mixtures on soyabean yield structure, under narrow-row and wide-row sowing.

## Materials and methods

Field experiments were conducted in 1995 and 1996 on a chernozem soil in soyabean crop. The experimental design was a split plot, with wide-row (70 cm) and narrow-row (24 cm) sowing as the main plots followed by ten treatments with three replications at a plot size 10 m<sup>2</sup>. The herbicide rates were in kg a.i. ha<sup>-1</sup>: 1, untreated control; 2, trifluralin 0.96; 3, metolachlor 2.00; 4, alachlor 1.92; 5, linuron 1.00; 6, metobromuron 2.00; 7, trifluralin 0.96+linuron 1.00; 8, trifluralin 0.96+metobromuron 2.00; 9, metolachlor 2.00+linuron 1.00; 10, alachlor 1.92+linuron 1.00. The yield structure was determined at soyabean harvesting, according to the plants mean height, the number of seeds per plant, the number of pods per plant and the seed weight per plant. The data were statistically analyzed through ANOVA and *t*-test for independent samples.

## Results and discussion

The graphs ([Figure 1, A and B](#)) are based on the mean values, obtained in the analyses. The SEM (standard error of the means) for the yield structure elements in the wide-row sowing was: seed weight per plant (a) = 1.78, pods per plant (b) = 3.51, mean height per plant (c) = 4.45, seeds per plant (d) = 8.43 and for these in the narrow-row sowing – a = 1.07, b = 2.42, c = 3.94, d = 5.32, respectively.

The differences in the elements of the yield structure between the wide- and the narrow-row sowing were analyzed and proved by ANOVA (df = 9, 20 and  $F_{\text{theoretical}} = 2.40$ ) at a level of significance  $p < 0.05$ . All the four elements of the yield structure were better expressed in the wide-row sowing. Although the crop density was about 500 000 plants ha<sup>-1</sup> in both the wide- and narrow-row sowing (70 cm/4 cm and 24 cm/10 cm, respectively), the relative percentage of the mean plant height in the treatments with herbicides was bigger in the narrow-row sowing compared to the corresponding untreated controls (114 to 137 % in the narrow-row sowing and 104 to 118 %, respectively, in the wide-row sowing). The relative percentages of the other yield structure elements in all the treatments were lower in comparison to the control in the narrow-row sowing.

The differences between all the treatments were analyzed by ANOVA (df = 1, 4 and  $F_{\text{theoretical}} = 7.71$ ) and confirmed by *t*-test at  $p < 0.05$ . All the differences between each one of the treatments and the corresponding controls in relation to the yield structure elements were significant at  $p < 0.05$  and were proved in both the wide- and the narrow-row sowing. The yield structure was worse in the untreated controls and in the treatment with metolachlor (in both the wide and narrow-row sowing) and metolachlor+linuron in the narrow-row sowing. The best yield structure was in the treatments with trifluralin alone and with metobromuron or linuron.

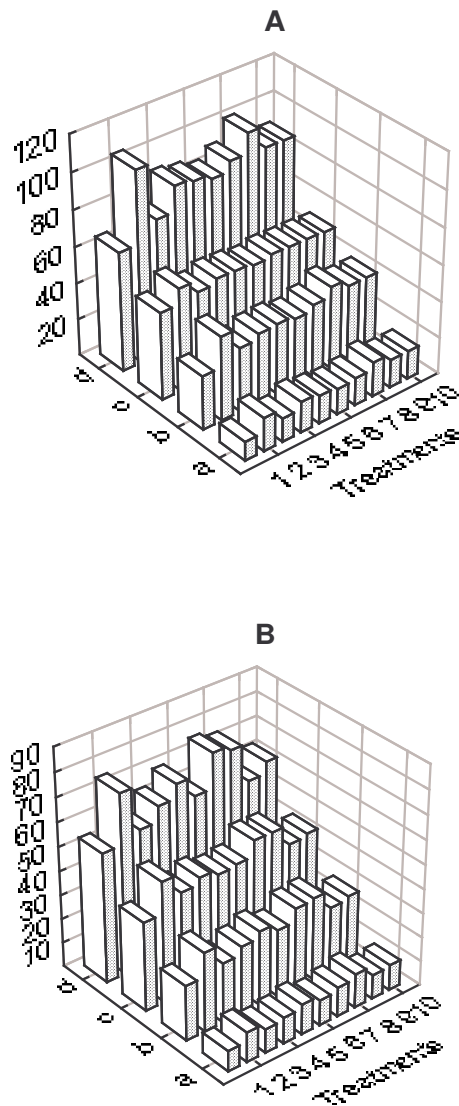


Figure 1. Yield structure in the wide-row sowing (A) and the narrow-row sowing (B). a – seed weight / 1 plant (g), b – pods / 1 plant, c – mean height / 1 plant (cm), d – seeds / 1 plant

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# Optimisation of some systemic herbicide doses on the control of *Elymus repens* (L.) in Romanian orchards

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## Introduction

The weeds have been always hazardous to all agricultural crops including fruit orchards as well. The investigations on the weed distribution carried out in Romania during 1960 – 2004 have emphasised that the highest frequency was recorded with *Elymus repens* representing 30% in the plots investigated. Between the weeds, soil type, climate, phreatic water, the technology applied, certain relationships are established which by their cumulated influence will determine the amount and fruit yield quality. *Elymus repens* frequency related to major soil types found in Romania is: Ca – saturated humus soils (16.5%); heavy – clay soils (36.1%); luvisols (60.4%); alluvial or undeveloped soils (38.3%); with an average of 32.6%. In Romania the orchards acreage covers 215, 000 ha, namely 1.43% of the total agricultural area. Weed mapping during 1993 – 2003 emphasised that out of 51 "key" weed species growing in the fruit orchards, 39.6% - 48.2% were monocotyledonous and 51.8% - 60.4% were dicotyledonous; the most spread and hazardous monocotyledonous perennial weeds was *Elymus repens* (13.2-24.9%). For this weed control, various herbicide rates and optimum timing of application were tried and evaluated at different weed development stages in the apple orchards.

## Material and methods

The investigations were performed in bear apple orchards. The weed mapping was carried out during the growing season starting with March-April and the weed groundcover was determined between August-September. Soil types, their texture and pH vary even in the same orchard, therefore investigations regarding weed variety and number were necessary in each apple orchard. The experiment was designed in randomised patch in 4 replicates along the tree rows (1.2 m width and 200 m length). A biological and ecological description of *Elymus repens* in the orchards is presented. Three herbicides were studied: a.i. *glyphosate* 400 g l<sup>-1</sup> in commercial product doses of 1.0; 1.5; 2.0; 2.5; 3.0; 3.5 and 4.0 l ha<sup>-1</sup>; a.i. *PMG glyphosate* 360 g l<sup>-1</sup> + *helping ion + surfactant*, in commercial product doses of 1.0; 1.5; 2.0; 2.5; 3.0; 3.5 and 4.0 l ha<sup>-1</sup>; a.i. *haloxifop – R – metil* 100 g l<sup>-1</sup> in commercial product doses of 0.5; 0.75; 1.0; 1.25 and 1.5 l ha<sup>-1</sup>. The herbicides were applied at three *Elymus repens* groundcover density degrees: small (15%) patch; medium (35%) patch and big (over 50%) patch, at two heights of the common couch-grass weed, 10 cm early postemergent application and 20 cm regular postemergent application. Phenological observations and biological measurements were done. The herbicide efficacy and selectivity was scored on EWRS scale.

## Results and discussion

Table 1 shows the optimum herbicides doses at two application timings, three density levels of *Elymus repens* and two heights of the weed, in apple orchards. This optimum-dosing model, used together with the patch spraying practice, is an alternative to the economic thresholds and allows achieving maximum profits.

Table 1. Optimum herbicides doses at two application timings, three density levels of *Elymus repens* and two heights of the weed, apple orchards.

<i>Elymus repens</i> density	Commercial product doses 1 ha <sup>-1</sup>					
	a.i. <i>glyphosate</i> 400 g.l <sup>-1</sup>		a.i. <i>PMG 360 g.l<sup>-1</sup> + helping ion + surfactant</i>		a.i. <i>haloxifop –R-metil</i> 100 g.l <sup>-1</sup>	
	Height 10 cm	Height 20 cm	Height 10 cm	Height 20 cm	Height 10 cm	Height 20 cm
Small (15%) patch	1.0	2.0	1.0	2.0	0.5	0.75
Medium (35%) patch	2.0	3.0	2.0	2.5	0.5	1.0
Big (over 50%) patch	3.0	4.0	3.0	3.5	0.75	1.5

The best control of *Elymus repens* resulted to be done during the early spring, when the weeds have 6-8 leaves (10cm height) by applying the smallest tested rates of the upper mentioned herbicides. If is not applied in spring, summer treatments (over 20cm height) should use maximum tested rates of herbicides (table 1), because of a large amount of growth rhizomes. Otherwise, low dosed herbicide application will permit the redevelopment of the weed, asking for new other treatments (figure 1).



Fig. 1. Redevelopment of *Elymus repens* from rhizomes after a low dosed herbicide application (Gallant super)

Application of herbicides as part of IWM emphasised the best efficacy for: Roundup 2000 (*glyphosate* 400g.l<sup>-1</sup>) at rates of 1-4 lha<sup>-1</sup>, Touchdown system 4 (*PMG 360g.l<sup>-1</sup> + helping ion + surfactant*) at rates of 2-3.5 lha<sup>-1</sup> in 150 l of water ha<sup>-1</sup> and Gallant super (*haloxifop –R-metil*100g.l<sup>-1</sup>) at rates of 0,5 – 1,5 lha<sup>-1</sup> in 200 l of water ha<sup>-1</sup> (note 1 EWRS).

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## Relating pre-spraying weather conditions to herbicide uptake and efficacy

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### Introduction

The Minimum Lethal Herbicide Dose (MLHD) provides a weed species and plant size specific estimate of the lowest dose of a photosynthesis inhibiting herbicide required to control a weed population. The methodology does not take into account the weather conditions experienced by the weed plants. The decision support system Gewis (<http://www.opticrop.nl>) has been used in the Netherlands for a couple of years. This system utilizes information about weather conditions around the time of spraying to indicate to farmers the optimal time within the next few days to apply a specific herbicide. Gewis takes into account parameters related to weed physiology (e.g. thickness and hydration of the cuticle), information related to the herbicide (e.g. mode of action, physical and chemical properties) as well as weather related information (e.g. radiation, temperature, precipitation.) Gewis is partly based on site-specific recorded weather data for the last three days and on forecasts for the next three days.

Farmers often combine the advice on the optimal spraying time with a reduced dose application. The Sugar Beet Research Institute (IRS) demonstrated in 2000 that both the MLHD methodology and the Gewis decision support system led to an even greater dose reduction than the traditional low dosage system used in sugar beet in the Netherlands. It was hypothesized that an extension of the MLHD, in which the advised dose would also be based on weather conditions, would result in a further reduction in herbicide use. An important element of the MLHD-concept is the minimization of the risk of treatment failure. Hence, recommendations based on recorded weather data seem to be more in line with this concept than recommendations based on forecasted weather data. Pre-spraying weather conditions are assumed to mainly have an effect on herbicide uptake through an effect on wax formation and the permeability of the cuticle. The current research was undertaken to determine the influence of pre-spraying weather conditions on herbicide uptake and efficacy for a combination of four weed species and three herbicides, in order to verify the general validity of the formulated hypothesis.

### Materials and methods

In 2004, outside pot experiments were conducted in Wageningen (The Netherlands) in order to study the influence of pre-spraying weather conditions on herbicide uptake and herbicide efficacy. Four species with contrasting cuticle development were used: *Brassica napus*, *Chenopodium album*, *Senecio vulgaris*, and *Solanum nigrum*. The cuticle of *B. napus* easily develops into a thick waxy (apolar) layer under hot and dry conditions whereas the cuticle of *S. nigrum* is affected very little by weather conditions. A polar herbicide, bentazone (Basagran, 480 g L<sup>-1</sup>, BASF) and an apolar herbicide, phenmedipham (Herbasan, 160 g L<sup>-1</sup>, Bayer Cropscience) were used as their uptake is supposed to be differentially influenced by the development and hydration status of the cuticle. The third herbicide treatment was bentazone + 1% v/v of an oilseed rape oil. To assess whether pre-spraying weather conditions had an influence on uptake and herbicide efficacy, 8 batches of plants were sprayed from July 15<sup>th</sup> till August 25<sup>th</sup> with one-week intervals. Herbicides were applied at 5 doses. After spraying, plants were placed in a greenhouse to standardise as much as possible the post-spraying weather conditions. Fifteen days after treatment plants were harvested and a log-



logistic curve was used to fit dry weight to herbicide dose rate. Additionally, herbicide uptake was measured 1 day after application using radiolabelled herbicides (samples provided by BASF and Bayer Cropscience). Weather data were recorded throughout the experiment.

## Results and discussion

In Figure 1 the type of information that is retrieved from these experiments is illustrated for the combination of phenmedipham and *S. nigrum* applied at spraying time 3, 6 and 8. Figure 1 (A) shows the percentage of phenmedipham uptake in *S. nigrum* leaves. The uptake rate was highest at spraying time 3 and lowest at spraying time 6. Differences in uptake at the three spraying times were statistically significant. In Figure 1 (B) dose response curves for the same application times are presented. *S. nigrum* plant dry weight (DW) was used as response variable and the highest phenmedipham dose was equal to half of the recommended field rate. The nonlinear regression analysis revealed that the only parameter differing between the dose response curves was the ED<sub>50</sub> value. Increasing ED<sub>50</sub> doses were observed for spraying times 3, 8 and 6, indicating that herbicide efficacy was highest for spraying time 3 and lowest for spraying time 6. Hence, in this case there is a clear correlation between uptake rate and herbicide efficacy. Despite both the uptake of phenmedipham and development of the leaf surface of *S. nigrum* were expected to be relatively insensitive to weather conditions, clear differences were found in both uptake rate and efficacy between the spraying times. Although not fully analysed, weather data suggest that spraying time 6 was preceded by a hot and relatively dry period whereas rain (2 mm) was recorded before spraying time 3 and mean temperatures were about 2 degrees lower in the 12 hours preceding application.

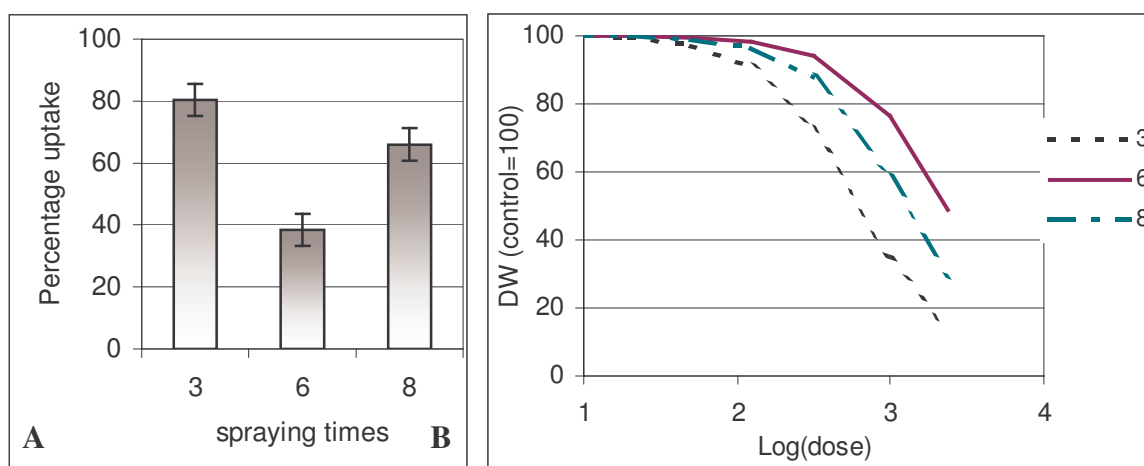


Figure 1. (A) Percentage leaf uptake of phenmedipham in *Solanum nigrum* at spraying times 3, 6 and 8. (B) Relationship between the DW (control=100) and dose of product for *Solanum nigrum* sprayed with phenmedipham at spraying times 3, 6 and 8.

Further statistical analysis will first address the relationships between herbicide efficacy and uptake rate for the 4 × 3 combinations of weed species and herbicides tested and then the relationship between uptake rate and pre-spraying weather conditions will be investigated. These results should reveal whether accounting for pre-spraying weather conditions are an option that could improve the MLHD-technology.

## References

<http://www.opticrop.nl> for information on MLHD and Gewis

## Analysis of bioassays

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### Introduction

Bioassays are experiments with biologically active compounds. In herbicide selectivity studies it is common to run suites of bioassays with dose-response curves for different plant species and/or different herbicides. The same principles apply to the study of compounds in toxicology and pharmacology. In herbicide research and development, potencies of compounds usually are compared at some a priori response levels; say 50% reduction (ED50) in biomass. For example, ED10 denotes 10% effect, which is 90% of the untreated control. To describe herbicide selectivity we compare tolerance of crops, ED10, and sensitivity of weeds, ED90. These ways of comparing compound potencies and/or species sensitivity also are widely used in ecotoxicology of xenobiotics. We have developed software which is capable of carrying out simultaneous nonlinear regression analyzes on several dose-response curves. The software package is called *drc*. The functions in *drc* provide a convenient means for specifying models, controlling the minimization and retrieving relevant results, including comparisons of parameters of interest and graphic presentations. The package is an add-on package for the language and environment R (R Development Core Team, 2004) which is open source and freely available (see <http://www.R-project.org>). We present some of the functionality of the package *drc*, applied to specific examples.

### Materials and methods

Available built-in models in *drc* include

- 1) three- and four-parameter Gompertz curves,
- 2) three- and four-parameter logistic curves,
- 3) models allowing for hormesis.

It is possible to define new functions (user-defined functions) and then fit them using the package *drc*.

For all built-in models, *drc* provides a self-starter facility, so the experimenter does not need to give initial estimates of the parameters.

The package provides a convenient way to

- 1) test for similar parameters among some or all simultaneously fitted curves,
- 2) reduce numbers of parameters using F-tests or likelihood ratio tests,
- 3) test for lack of fit (versus ANOVA),
- 4) calculate relative potencies (comparison of parameters),
- 5) compare arbitrary ED<sub>x</sub> values across experiments,
- 6) create plots of data and the fitted model.

More details on the package *drc* can be found in Ritz & Streibig (2005) and at the web page <http://www.bioassay.dk>.

## Results and discussion

Data by Streibig et al. (1999) was analysed using *drc*. To fit data and construct a plot only requires a few commands, enabling swift analysis of standard dose response curves. The resulting plot of the original data and the fitted model is given in Figure 1.

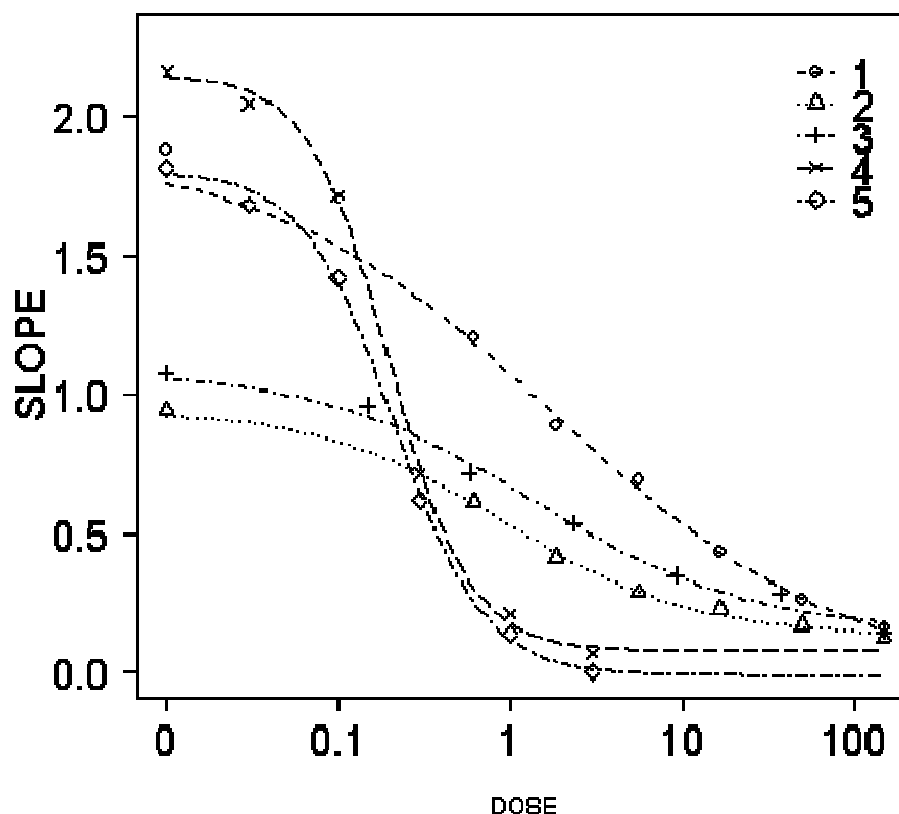


Figure 1. Plot of relationship between response and dose for data (Streibig et al. 1999).

A step-by-step analysis of the data with extraction and comparison of relevant parameters, is presented by Ritz & Streibig (2005).

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# Herbicide Research and Development – challenges and opportunities

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## Introduction

The agrochemical industry has been very successful in developing new herbicides. New chemistry with improved properties, especially providing significantly reduced use rates, and often new modes-of-action have been discovered, developed and launched in the market place for many different crops. This success positively influenced agriculture as a whole. However, these days the output of new active ingredients belonging to entirely new chemical classes is relatively low, and especially so for active ingredients with a new mode of action technical progress in the area of herbicides is relatively moderate (Schulte, 2004). Though this may not be evident today: if this trend continued, it could impact the success of farming and agricultural production long-term. To analyse parameters for this slow technical progress and to sketch potential opportunities for the future is the aim of the paper presented.

## Discussion

The main driver for technical progress - also in the herbicide area - is business attractiveness. However, such opportunities clearly are influenced by the excellent technical quality of existing ('old') herbicides, setting a very high technical, regulatory, and economic standard to match (or even to beat). In maize, for example, active ingredients discovered between 1950 and 1980 (e.g. triazines and acetanilides) still represent a significant portion of the market value. Due to their high market volume globally, the major food crops, such as maize, cereals, and rice, have always been attractive targets for new active ingredients. This might explain why, for instance in maize, there is a broad range of mode of actions available. The introduction of the herbicide-tolerance technology basically added another chemistry to those crops, although the compound glyphosate as such is old. Other crops like sugarcane, worth the development of a specific new active ingredient in the past (e.g. ametryne), are today considered more of a minor crop, where new active ingredients will need to be spin-offs from projects in a major crop like, such as cereals or maize, which *per se* offer a bigger market potential (Quadranti & Nevill, 2004).

The introduction of herbicide-tolerant crops, beginning in the early '90ies, has basically shifted value from chemicals to the seed business, while leaving the total expenditure for weed control unaffected. This shift was facilitated by charging 'technology fees', while at the same time offering a different weed control concept. Consequently, *conventional* crop protection research competes with traits research for R&D resources. But herbicide-tolerant crops (and GMOs in general) are considered as opportunity in the agribusiness.

Further, the consolidation process in the agrochemical industry (Copping, 2003) has reduced overall R&D resources. This, together with the ever increasing cost for herbicide development, represent a significant challenge to project prioritisation and, therefore, require more focus (or 'risk taking') at a very early stage, since it is not possible to maintain a broad diversification of research projects. From published patents one might conclude, that worldwide the numbers of chemistry labs has declined sharply, and therefore the diversity of chemical classes (measured by

numbers and patent diversity) declined. In such a business environment, generic companies may seem to have a better (cost) position, which in turn indirectly delimits overall technical progress.

However, the potential success of such innovations also depends on cost. Operating a farm successfully is a complex task (compare Zoschke & Quadranti, 2002). Farmers are under pressure, commodity prices for most agricultural produce are low, subsidies under scrutiny. In order to save cost, farmers might think of shaving product rates, applying less than recommended on the label. However, this not only puts the success of a weed control program at risk, but potentially favors the built-up of resistance of herbicides to weeds. Likewise, the intensive use of one and the same herbicide is likely to promote herbicide resistance development, but at an accelerated pace, if used in various crops grown in the same rotation; as we are currently experiencing with glyphosate in herbicide-tolerant crops in the USA.

In sharp contrast to limited R&D resources, regulatory requirements are increasing (Anonymus, 2005). Regulatory support work absorbs significant R&D resources, either directly by competition for test capacity in glasshouse and field, or indirectly through resource allocation at external labs and contractors. Again, generic companies are much less affected by such increasing regulatory demands, benefiting from the regulatory work and significant product support of R&D based companies.

Are there still opportunities, and are there still search targets? Is there still an incentive to invest into herbicide research?

Weed control will be necessary to secure crop yields, and for the foreseeable future chemical solutions will continue to be the preferred choice. The driver for profitability is and will be crop yield and quality. The high impact of weed competition on yields makes weed control mandatory. The decrease of (good) arable land (urbanisation, climatic changes) and the increase of the population already led to a intensification of crop production (output/surface) and this will continue.

To cope with the regulatory requirements today is a major task. Old compounds are under threat for various reasons. Requirements are unlikely to decrease, even though it is costly to maintain compounds in the market. In many parts of the world farmers are confronted with the fact their chosen weed control measures begin to lose effectiveness due to herbicide resistance, even face different levels of cross resistance.

Further questions to be addressed include the following: what are the expectations regarding weed shifts and/or the occurrence of (new) weed problems, e.g. due to development of herbicide resistance and/or resulting from climatic changes (global warming)? What are the needs arising from the intensification of agriculture in different parts of the world, e.g. in Eastern Europe or China? And what the consequences of various agro political changes, e.g. subsidy systems, bio fuel production?

Irrespective of the anticipated changes, farms become larger, and, therefore, farm practice changed and will continue to change. Farms in the Americas and Eastern Europe can reach thousands of hectares. These farms expect reliable weed control, but seek non-complex weed control tools as well. Speed, flexibility, and efficiency are of key importance. Low rate, one-shot products delivering broad spectrum weed control are wanted (but still not on the market for every crop). Mainly for reasons of convenience, the market favors products combining a number of herbicides in a stable

ready-made product; containing several active ingredients, even if their physico-chemical properties are very different. Such products are biologically near complete and handy for the end user.

### **Conclusion**

Compounds with a new mode of action are very welcome and are needed to manage herbicide resistance. However, all other parameters like crop tolerance (multi-crop fit ?), broad weed spectrum, timing flexibility, cost, favorable regulatory profile, and intellectual property must be lined up as well. This is a challenging task. However, in order to contribute to effective weed management and farming, we - the agrochemical industry – aim at meeting the challenge delivering successful herbicidal solutions for the future.

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# **Influence of the nitrogen fertilization and dose of herbicide on weed control in winter wheat**

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## **Introduction**

Herbicides applied in reduced doses, used in optimal conditions can limit weed infestation very well, and yield obtaining from this field is similar in comparison with objects, where full, recommended rate of herbicides were applied (Rola et al., 1998., Domaradzki & Rola, 2000). In research carried out in the years 1997 - 2000, a correlation between mineral fertilization, herbicide application time and efficiency of weed control was noticed (Rola & Banach, 2001).

The aim of investigations was to estimate the influence of top-dressing with nitrogen, different doses of herbicide and the term of application on the yield of winter wheat.

## **Materials and methods**

The experiments were carried out on a field in South-West Poland. Plots (20 m<sup>2</sup>) were established, randomized blocks method in three replications, on brown soil, in winter wheat crop. Top-dressing with nitrogen, ammonium nitrate and in two doses urea were applied in the plots. The dose of nitrogen fertilization (120 kg·ha<sup>-1</sup>) was applied in three different terms: T-1 – beginning of vegetation season – (50% of recommended dose), T-4 – the end of tillering – (25% of recommended dose), T-6 – beginning of ear formation – (25% of recommended dose). In the experiment herbicide Huzar 05 WG (iodosulfuron methyl = 5% and mafenpyr-diethyl = 15%) was applied in two different doses (maximum recommended, and 25% decreased) in three terms: full of tillering, the end of tillering (7 days before ammonium nitrate application) and the end of tillering (7 days after ammonium nitrate application together with dose of urea).

## **Results and discussion**

In favorable conditions (optimum application time, good condition of plant and small number) doses of herbicide can be decreased by 25% without any losses in yielding. The results of researches showed that the influence of the following factors: mineral fertilization, herbicide dose and term of application on the yield of winter wheat. The highest herbicidal efficiency together with the high yield was obtained when herbicide was applied in the maximum (recommended) dose in the end of tillering plants stage, but 7 days before ammonium nitrate application (T-3). The results can be used to estimate the optimum application time for nitrogen fertilizers and herbicides to obtain the expected herbicidal result and high winter wheat yield. The results are shown in table 1.

Table 1. Effect of the nitrogen fertilization terms and various doses of herbicide Huzar 05 WG on weeds control and yield of winter wheat

Treatment	Term of application	Dose per 1 ha	Yield (dt/ha)	Weigh of 1000 grains (g)	Weed control (%)	
					1	2
Untreated	-	-	31,5	41,99	132*	40*
Huzar 05 WG	T-2	200 g	48,4	45,68	76	82
Huzar 05 WG	T-2	150 g	39,2	42,10	63	76
Ammonium nitrate 34%	T-1, T-4, T-6	176,5+88,25+88,25 kg	39,0	44,99	0	0
Huzar 05 WG	T-2	200 g	65,7	46,99	78	83
Ammonium nitrate 34%	T-1, T-4, T-6	176,5+88,25+88,25 kg				
Huzar 05 WG	T-2	150 g	56,8	43,00	73	75
Ammonium nitrate 34%	T-1, T-4, T-6	176,5+88,25+88,25 kg				
Huzar 05 WG	T-3	200 g	73,1	49,77	91	92
Ammonium nitrate 34%	T-1, T-4, T-6	176,5+88,25+88,25 kg				
Huzar 05 WG	T-3	150 g	62,4	48,72	75	90
Ammonium nitrate 34%	T-1, T-4, T-6	176,5+88,25+88,25 kg				
Huzar 05 WG	T-5	200 g	63,2	49,82	78	91
Ammonium nitrate 34%	T-1 T-4, T-6	176,5+88,25+88,25 kg				
Huzar 05 WG	T-5	150 g	54,7	45,23	70	87
Ammonium nitrate 34%	T-1 T-4, T-6	176,5+88,25+88,25 kg				
Huzar 05 WG	T-3	200 g	60,1	50,03	84	92
Ammonium nitrate 34%	T-1	176,5 kg				
Coated urea 46%	T-3, T-6	65,25+65,25 kg				
Huzar 05 WG	T-3	150 g	52,1	47,76	72	90
Ammonium nitrate 34%	T-1	176,5 kg				
Coated urea 46%	T-3, T-6	65,25+65,25 kg				

\* - for untreated plots - number of weeds per sq. m

1 - monocotyledonous

2 - dicotyledonous

Term of application:

T-1 - beginning of vegetation season; T-2 - full of tillering; T-3 - the end of tillering (7 days before ammonium nitrate application, and together with dose of urea; T-4 - the end of tillering; T-5 - the end of tillering (7 days after ammonium nitrate application); T-6 - beginning of ear formation.

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# Influence of soil cultivation on degradation rate of sulfonylurea herbicides in winter wheat

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## Introduction

Sulfonylurea herbicides have been used in Poland for more than 20 years, mainly to control weeds in cereals and maize (Adamczewski 1988, Rola & Rola 2001). Sulfonylurea herbicides are degraded in the soil as a result of chemical reactions and soil microbes. The sulfonylurea herbicides are used at small rates ( $5\text{-}60\text{ g ha}^{-1}$ ) and its residues determination is very difficult. Biological tests are the accepted method to evaluate the phytotoxicity, degradation rate and residue level in soil.

The aim of the investigation, carried out in the years 2001-2004, was to estimate chlorsulfuron and amidosulfuron degradation rate depending on the kind of tillage system.

## Materials and methods

The experiments were carried out on a field in South-West Poland. Randomized plots ( $20\text{m}^2$ ) were used in 4 replicates in monoculture of winter wheat for two tillage systems: conventional and reduced. On those plots herbicides containing chlorsulfuron (Glean 75 WG) and amidosulfuron (Grotyl 75 WG) were applied in the stage of 23-25 BBCH of winter wheat, with rates of 25 and  $40\text{ g ha}^{-1}$ .

Five samples of soil were taken from each plot (layer 0-10 cm) at 2, 4, 6 and 8 weeks after herbicide application and additionally at harvest time. Soil samples were sieved, mixed and packed in plastic pots. The plastic pots were used for the biotest in white mustard. The seeds were planted at a depth of 0.3 cm in the pots (5 seeds per pots). Seedlings were grown in a greenhouse at  $25/15 \pm 3\text{ }^\circ\text{C}$  day/night temperature. Natural sunlight was supplemented with light from sodium lamps that provided a photosynthetic photon flux density of  $500\text{ }\mu\text{mol m}^{-2}\text{ s}^{-1}$  at plant height during a 16-h photoperiod. 14 days after sowing, dry weight of plants were determined and calculated (in comparison to the check plants – plant sowed in soil from untreated plots).

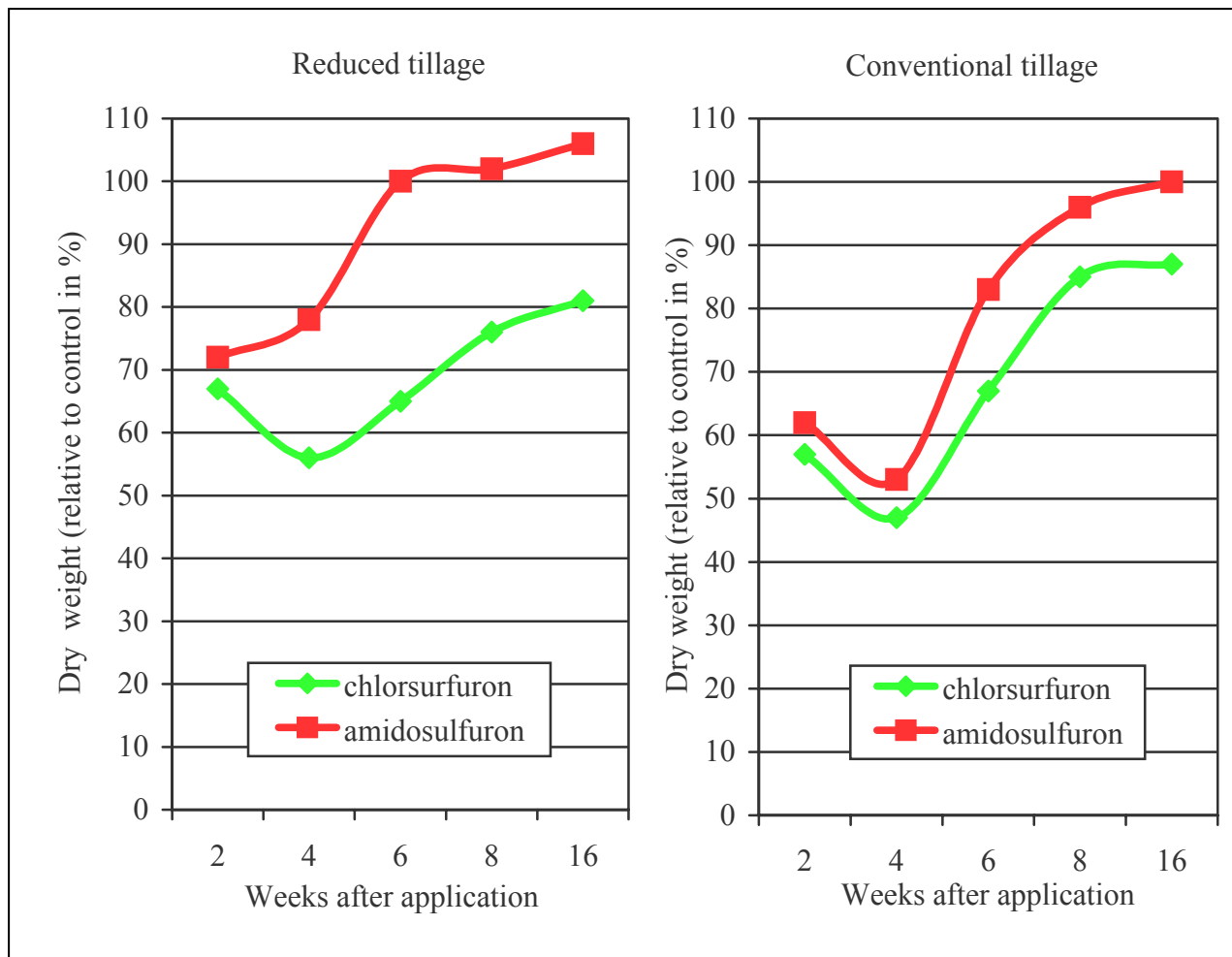
## Results and discussion

The results of the experiment showed that phytotoxic effects of chlorsulfuron and amidosulfuron on white mustard occurred in samples taken 2-8 weeks after application where the conventional tillage system was used (decrease of dry weight in comparison to control plants). In soil samples taken at harvest time (16 weeks after treatment) the phytotoxic effects were observed in treatments, where chlorsulfuron was applied. For amidosulfuron treatments, where the reduced tillage system was used phytotoxic effects were observed only in samples taken at 2-6 weeks after herbicide application. The dry weight of plants sowed in soil taken at harvest time was similar to the control. Residues of chlorsulfuron lead to a reduction of white mustard dry weight all sample times (sowed in soil taken at 2, 4, 6, 8 and 16 weeks after herbicide application). The results are showed in figure 1.

## Conclusion

- same tendency to both compounds
- influence of soil cultivation

Figure 1. Degradation rate of chlorsulfuron and amidosulfuron in soil (layer of 0-10 cm)



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# Isoproturon persistence in field soils under North-Western Himalayan conditions

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## Introduction

Isoproturon [3-(4-isopropyl phenyl)-1, 1 dimethyl urea] belonging to family of substituted ureas is a recommended herbicide in wheat. The broad spectrum activity of isoproturon has made it to be a popular herbicide for the effective control of annual grasses and broad leaved weeds (Hance and Holly, 1990). The reliance on herbicides may pose environmental hazards. Depending upon crop canopy and application method, up to 50% of a foliar applied pesticide can reach the soil. So a high percentage of the applied herbicide may also find its way into soil, irrespective of the method of application or target of spray. In the soil the herbicides can modify or alter many physiological and biological transformations. Therefore, it is imperative to understand how long isoproturon persists in soil. The increasing use of isoproturon in wheat in the hilly state of North-Western Himalaya has raised concerns about its fate in the environment. The pattern of dissipation and its persistence in the soil are important aspects to be investigated. Few studies on these aspects have been conducted in India (Kulshreshtha, 1982; Randhawa *et al.*, 1989) and also abroad (Almvik *et al.*, 2003; Perrin *et al.*, 1996) but especially for hilly regions there appears to be a gap in this field of study. With this objective in view, the present study was designed to evaluate persistence of isoproturon in wet and highly humid agro climatic zone of Himachal Pradesh.

## Materials and methods

A field experiment was conducted in wheat (var. HPW-89) in *Rabi* 2003-04 (Nov.-May) using a randomized block design. The soil consisted of 28% sand, 42% silt, 30% clay and 1.27% organic matter and had a pH of 5.16. Isoproturon (Masslon, 75 WP) was sprayed as post emergence treatment at three different doses. 1.5 kg ha<sup>-1</sup>, 2.0 kg ha<sup>-1</sup> and 2.5 kg ha<sup>-1</sup>. These chemical treatments were compared with untreated control. All the treatments were replicated thrice. Soil samples (0-15 cm depth) were collected immediately following application and then at 1, 3, 5, 7, 10, 15, 30, 45, 60, 90 and 120 days after herbicide application for each of the three replications. Soil samples collected were mixed thoroughly, air dried, then processed and extracted with dichloromethane. Isoproturon in the extracts were analyzed by using a spectrophotometric method (Katz, 1966 and Kulshreshtha, 1982) at 555 nm detecting isoproturon residues remaining in the soil. Soil samples from control plots were also extracted and analyzed using similar conditions.

## Results and discussion

The isoproturon residues ( $\mu\text{g g}^{-1}$ ) that were obtained in the samples of the different time intervals and doses applied are presented in Table 1. The initial deposits of isoproturon in soil were found to be 0.45, 0.61 and 0.80  $\mu\text{g g}^{-1}$  for isoproturon applied at 1.5 kg ha<sup>-1</sup>, 2.0 kg ha<sup>-1</sup> and 2.5 kg ha<sup>-1</sup>. In samples that were taken after 60 days of herbicide application the Isoproturon residues were less than the detection limit, 0.19 and 0.10  $\mu\text{g g}^{-1}$  respectively. Thus the corresponding losses of isoproturon were 100 per cent. 96.88 per cent and 87.30 per cent respectively. At harvest, the residue levels were also non detectable at all the three levels of

herbicide application. For the 3 application rates tested it can be concluded that the isoproturon persisted in the soil up to 45, 60 and 60 days respectively.

Table 1 Residues of isoproturon in soil (0-15 cm depth) treated with different doses

Days after herbicide application	Rates of isoproturon application		
	1.5 kg ha <sup>-1</sup>	2.0 kg ha <sup>-1</sup>	2.5 kg ha <sup>-1</sup>
0	0.450 (0.00)	0.610 (0.00)	0.796 (0.00)
1	0.436 (3.10)	0.596 (2.29)	0.783 (1.63)
3	0.381 (15.15)	0.545 (10.58)	0.727 (8.63)
5	0.327 (27.20)	0.463 (23.90)	0.645 (18.90)
7	0.254 (43.44)	0.407 (33.20)	0.610 (23.20)
10	0.232 (48.20)	0.370 (39.10)	0.538 (32.39)
15	0.112 (74.10)	0.305 (49.90)	0.480 (39.60)
30	0.076 (83.10)	0.167 (72.50)	0.250 (68.40)
45	0.034 (92.44)	0.101 (83.40)	0.200 (74.80)
60	ND (-)	0.019 (96.88)	0.101 (87.30)
90	ND (-)	ND (-)	ND (-)
120	ND (-)	ND (-)	ND (-)

Values in parentheses indicate per cent dissipation

The logarithmic plots of residual concentration against time are depicted in Fig. 1. The straight line relationships obtained for each of the three doses indicated an acceptable fit to first-order reaction kinetics. The correlation coefficient (r) for all the three doses (significantly) ranged from 0.95 to 0.98. The regression equations worked out for isoproturon (1.5 kg ha<sup>-1</sup>, 2.0 kg ha<sup>-1</sup> and 2.5 kg ha<sup>-1</sup>) were found to be  $Y = -0.0251x + 1.6097$ ,  $Y = -0.0222x + 1.8002$  and  $Y = -0.145x + 1.8889$  respectively. Their corresponding first-order half lives were derived from the slope of the lines and were found to be 11.9, 13.5 and 20.8 days respectively.

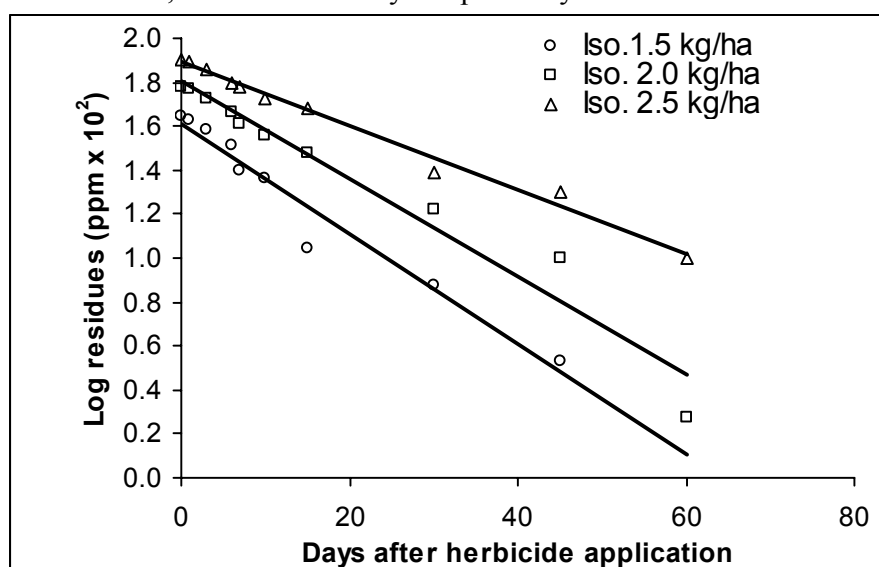


Figure1. First order dissipation curve of isoproturon in soil.

In general, rapid loss of herbicide applied at different rates was noticed in initial 15 days.



This might be due to the combined effect of various physicochemical parameters like favourable temperature, high rainfall, heavy texture of the soil (42% silt and 30% clay), high organic matter (1.27%) content etc. As the period of 20 days after herbicide application experienced total rainfall of 139.6 mm with the event of two heavy continuous showers i.e. on day 9, 10, 11 (26.5 mm, 24.4 mm, 66.0 mm) and on day 18, 19, 20 (22.8 mm, 72.5 mm, 12.0 mm), it is quite likely that the compound (aqueous solubility 70 mg/l) might have leached down to lower depths. The alike environmental conditions, heavy rainfall, high average humidity (>64%) and conducive temperature prevailed during the tenure of 30 days after herbicide spray must have resulted almost complete disappearance of the herbicide from field.

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# Effect of irrigation and herbicide application on the weed association in maize

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## Introduction

Soil properties, especially a soil water system, can determine the composition and structure of weed association. The distribution of certain weed species, as well as, their effects on the crop under rainfed conditions differs from irrigation conditions (Stanojević et al., 2000). The weed association of maize as a spring, broadcast crop, has a pronounced seasonal dynamics in the form of three aspects: spring, summer and autumn. The dynamics of weed association is affected not only by environmental conditions, but also by the application of maize cultivation measures, such as irrigation, application of herbicides, etc.

## Material and methods

Maize was grown during four years (1996-1999) on sandy loam soil at Zemun Polje, under the effects of two factors: a) non-irrigation and irrigation conditions and b) herbicide and no-herbicide application. The herbicide combination of atrazine+metolachlore was applied preemergence in the amount of 1.0 and 2.88 l a.i. ha<sup>-1</sup>. The control plot was untreated. Sprinkler irrigation was performed when soil moisture was below the lower level of easy available water, which amounts to approximately 75% of field water capacity for slightly calcareous chernozem. Hence the following irrigation rates were applied: 160 mm, 40 mm, 340 mm. Due to excessive precipitation in 1999 the irrigation was not performed. Fresh weight of each weed species was measured in the spring, summer and autumn period. Fresh weight of seven prevailing weeds and total fresh biomass of all weeds were analysed. Average values of total fresh weed biomass for the four-year period are processed by ANOVA test.

## Results and discussion

Total fresh weed biomass, averagely for four years, was lower in the treated than in the control variant under both, non- and irrigation conditions, and in all aspects of the weed association. Furthermore, total fresh weed biomass in spring and summer aspects, was statistically significantly higher under irrigation conditions in treated and untreated variants. On the other hand, total fresh weed biomass in the autumn aspect was greater under non-irrigation conditions,

Table 1. Distribution of certain weed species (%) in the total weediness - spring aspect

Weed species	Control		Treatment		Average
	Non- irrigation	Irrigation	Non-irrigation	Irrigation	
AMARE	25.71	26.65	17.56	12.89	20.70
SORHA	10.81	21.10	11.49	17.09	15.12
DATST	6.12	4.63	21.95	16.99	12.42
CIRAR	3.93	2.08	15.23	9.81	7.76
CONAR	7.04	10.77	3.10	8.52	7.36
SOLNI	12.41	9.54	3.03	4.42	7.35
CHEHY	5.32	3.21	2.23	2.50	3.32
Total fresh weight (g m <sup>-2</sup> )	1147.32 <sup>b</sup>	1887.76 <sup>a</sup>	292.54 <sup>d</sup>	541.91 <sup>c</sup>	967.38

LSD<sub>0.01</sub> = 139.2

probably due to faster drying of leaves in the lower part of the maize plants, better light passing and additional emergence of certain weed species in the second half of the maize growing period. Irrigation affected the increase of total fresh biomass of the species *Sorghum*

*halepense Convolvulus arvensis* and *Amaranthus retroflexus* on the control plot in the spring aspect (table 1). Perennial species *Convolvulus arvensis* and *Sorghum halepense* had greater total fresh biomass in the treated area. The summer aspect in the irrigated area was characterised by a greater total fresh biomass of the species *Solanum nigrum*, *Chenopodium hybridum*, *Datura stramonium* and *Sorghum halepense* in the control variant, while *Sorghum halepense* had greater total fresh biomass in the treated variant (table 2).

Table 2. Distribution of certain weed species (%) in the total weediness - summer aspect

Weed species	Control		Treatment		Average
	Non- irrigation	Irrigation	Non-irrigation	Irrigation	
AMARE	35.27	31.94	14.20	14.85	24.07
SOLNI	12.95	19.42	8.67	9.07	12.53
SORHA	4.94	6.92	11.77	20.67	11.75
CHEHY	8.52	9.24	10.61	10.13	9.63
CONAR	3.71	2.37	13.0	11.02	7.53
DATST	6.64	8.46	5.09	5.34	6.38
CIRAR	4.07	3.14	7.45	5.52	5.05
Total fresh weight (g m <sup>-2</sup> )	1820.54 <sup>b</sup>	2573.35 <sup>a</sup>	546.01 <sup>c</sup>	741.09 <sup>c</sup>	1420.25
LSD <sub>0.01</sub> = 249.6					

Irrigation in the autumn aspect affected the increase of total fresh biomass mainly of perennial species, *Sorghum halepense* and *Convolvulus arvensis* in control and treated variants (table 3).

Table 3. Distribution of certain weed species (%) in the total weediness - autumn aspect

Weed species	Control		Treatment		Average
	Non- irrigation	Irrigation	Non-irrigation	Irrigation	
SORHA	11.94	14.64	19.96	24.65	17.80
AMARE	24.84	23.75	16.46	5.73	17.70
SOLNI	20.17	20.48	12.77	12.69	16.53
CONAR	5.84	7.59	10.43	13.39	9.31
CIRAR	5.22	5.40	7.52	8.39	6.63
CHEHY	4.44	3.86	7.13	6.00	5.36
DATST	3.75	3.33	1.50	3.56	3.04
Total fresh weight (g m <sup>-2</sup> )	1132.12 <sup>a</sup>	787.32 <sup>b</sup>	284.30 <sup>c</sup>	240.10 <sup>c</sup>	610.96
LSD <sub>0.01</sub> = 115.0					

Means followed by the same letters are not statistically different for P<0.05 and P<0.01 (LSD-test)

Irrigation very strongly affects the composition and structure of the maize weed association and can be considered the key factor affecting weediness. The application of herbicides that significantly reduce the weed distribution, is not sufficiently efficient in weed control under irrigation conditions, especially in control of the species *Datura stramonium*, *Solanum nigrum* and *Amaranthus retroflexus*. Obtained results are in agreement with the results attained by Momirović et al, 1997 in the conditions of Zemun Polje.

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# Assessment of the time of application of foramsulfuron plus iodosulfuron efficacy for weed control in maize

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## Introduction

Herbicides are still the primary tools used for weed control in maize (*Zea mays* L.). However, the potential contamination of ground and surface water, the increased regulations for pesticide application, and economic considerations have caused a new wave of interest in post-emergence application of low-dosed herbicide mixed with different additives (Mekki & Leroux, 1994; Nalewaja et al., 1991; Skrzypczak & Pudelko, 1993). History has shown that new technology simply creates the opportunity for change – whether the change actually takes place and gains acceptance depends on the social, economic and political climate prevalent at the time (Townson et al., 1995).

Post-emergent treatments allow application to the whole field or to weed field patches because weeds are visible at application. However consistent control under various climatic conditions would be required if post-emergent treatments are to be used as a primary means of weed control in maize.

The objective of this research was to determine the effect of the grass and broadleaves weed control using different application times of foramsulfuron plus iodosulfuron in maize.

## Materials and methods

Field trials were conducted in maize (var. Veritis) grown at the Brody Research and Education Station of Agricultural University of Poznań, during the 2002-2004 growing season. Herbicide plots were established before maize planting. Individual plots consisted of four treated rows of 10 m length with 0.7 m between rows. Every year each experiment was arranged in randomised complete block design with four replications. All experiments were applied with bicycle plot sprayer fan nozzles. Treatments were applied at 2, 4, 6 and 8 leaves (BBCH 12-18) of the maize growth. Herbicide treatments are given in detail in Table 1. Foramsulfuron (45 g·ha<sup>-1</sup>) + iodosulfuron (1,5 g·ha<sup>-1</sup>) (Maister 310 WG 150 g·ha<sup>-1</sup>) and oil additive (1263 g·ha<sup>-1</sup>) (Actirob 842 EC 1,5 l·ha<sup>-1</sup> - methyl ester of rapeseed oil) were used in field research.

Assessments of weed control, especially grasses like *Echinochloa crus-galli* (L.) Beauv. and broadleaf weeds and selectivity were done 2 weeks after each treatment using visual estimations. Also, every year at the beginning of July (8-10 weeks after planting) the analysis of fresh mass of weeds per m<sup>2</sup> was done and weed control efficacy was calculated. Green matter as well as cob yield was assessed each year from the centre two rows of plots. Data of weed control and green matter yield of maize were subjected to an analysis of variance and treatment means were calculated with a least significant difference test at 5% of probability.

## Results and discussion

Phytotoxicity was observed on maize after herbicide treatment at 6 and 8 leaf stage of the plants. At 6 leaves of maize low shortening of the plants was observed, but at 8 leaves herbicide treatment gave more visible injury like shortening of the plants and twisting of the leaves.

The visual assessment of weed control indicated very good control of broadleaved weeds between 4 and 6 leaves of maize (Table 1). *Echinochloa crus-galli* was more effectively controlled when foramsulfuron plus iodosulfuron were applied at the 4-6 leaves of maize and

mostly all seedlings of ECHCG emerged. The analysis of fresh mass of weeds per square unit and weed control efficacy indicated also very good control when foramsulfuron + iodosulfuron with oil additives were used post-emergence at 4-6 leaves of maize. The late application at 8 leaves was less effective and weeds competed with maize for a long time. At this time of application less control of species was observed such as: *Chenopodium album* L., *Polygonum lapatifolium* L., *Polygonum convolvulus* L. and panicum weed like *Echinochloa crus-galli* (L.) Beauv.

Table 1. Weed control efficacy from applied treatments and their influence on green matter and cob yield of maize (2002-2004)

Treatment	Maize stage	Visual assessment			Fresh mass of weeds g ha <sup>-1</sup>	Weed control efficacy (%)	Yield (t ha <sup>-1</sup> )	
		Selectivity Scale 1-9**	Weed control				Green matter	Cobs
			ECHCG	DICOT				
Foramsulfuron + iodosulfuron and oil additive*	2 leaves	1	72	95	894	62	31,6	14,5
Foramsulfuron + iodosulfuron and oil additive*	4 leaves	1	88	97	222	91	32,1	15,6
Foramsulfuron + iodosulfuron and oil additive*	6 leaves	2	90	92	246	90	29,1	14,4
Foramsulfuron + iodosulfuron and oil additive*	8 leaves	4	63	68	1412	40	16,5	8,7
<b>Check – untreated</b>	-	<b>1</b>	<b>0</b>	<b>0</b>	<b>2380</b>	<b>0</b>	<b>10,8</b>	<b>5,4</b>
LSD <sub>(0,05)</sub>	-	1.76	12.0	16.6	535.4	-	4.1	2.0

\* foramsulfuron 45 g ha<sup>-1</sup> + iodosulfuron 1,5 g ha<sup>-1</sup> = Maister 310 WG 150 g ha<sup>-1</sup>

oil additive 1263 g ha<sup>-1</sup> = Actirob 842 EC 1,5 l ha<sup>-1</sup> (methyl ester of rapeseed oil)

\*\* 1 - no injury, 9 – complete damage of the crop

Herbicide treatments applied in maize by effective weed control influenced on the green matter and cobs yield. However, the late treatment gave some injury on maize plants and was less effective against weeds and significant lower yield was obtained. There were no differences in green matter and cobs yield when herbicide treatments were applied at 2-6 leaves of maize. Moreover yield of maize was no high because maize plants during two-vegetation season grown at low rainfall and low air temperature at the beginning of vegetation period.

### Acknowledgements

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Townson J. K. et al. 1995. 9<sup>th</sup> EWRS Symposium, Budapest, 11-20.

# Effect of different weed control measures (chemical, mechanical and integrated) on corn yield (*Zea mays* L.)

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## Introduction

Competition between the undesired plants and the crop has to be avoided if reasonable yields were to be achieved. Weeds were removed first by hand and mechanically when new farming tools were developed (Ghersa et al., 2000). Weed control approaches consider the efficacy of different mechanical and chemical tools to reduce weed stands (Radosevich et al., 1997). Breeding competitive crops to increase yield and reduce weed stands is a major objective in several parts of the world (Callaway and Forcella, 1999), and there are good examples of farmers successfully using continuation of weed control measures, including precise herbicide application (Moss, 1995). The aim of this study was to determine which methods of weed control achieved higher yield at corn cropping.

## Materials and methods

The experiment was carried out at Mahdasht agriculture research farm of Karaj Islamic Azad University (35°43' N latitude, 50° 56'E longitude, 1160m altitude) in 2003. This region is classified as semiarid. A random block design with four replicates and 10 treatments was used. -Treatments were alachlor; alachlor + cultivation; metribuzin; metribuzin + cultivation; trifluralin; trifluralin + cultivation; 2,4-D; 2,4-D + cultivation; weeds completely control and without any control. Alachlor, metribuzin, and trifluralin were applied preplanting and 2,4-D was applied at 2-5 leaf stage of corn (variety: Single cross 704). -Cultivation was done 15 days after the herbicide application. The crop rotation was winter wheat (*Triticum aestivum* L.) - Fallow?. Maize was sown at 8 seeds m<sup>-2</sup>, 4cm deep as 0.5m x 0.25m spacing. Each plot had 6 rows and 4m length.

## Results and discussion

Rows of kernels per corn cob:

This trait had significant difference between treatments. A maximum row number was detected in the hand weeded control, alachlor + cultivation, metribuzin + cultivation, trifluralin + cultivation respectively. In contrast a minimum row number was detected in the check and 2,4-D-treatment respectively. The experiments showed that an application of only one method (chemical or mechanical) was not completely useful. An integration of the methods can control weeds better and increase the row number (Table 1).

No. of kernels per row:

This trait had significant differences between treatments, too. Hand weeded control and alachlor + cultivation had maximum and no control treatment had minimum kernel numbers (Table 1). Herbicides and integrated treatments had better effects on this trait. Pre planting herbicides can control weed densities shortly after crop emergence therefore competitive influence of weeds is less than that of weed control with post emergent herbicides. Hall et al. (1992) defined the 3-leaf and 4-leaf stages of plant development as the critical period for weed control in maize. Rajcan and Swanton (2001) also determined a similar critical crop weed competition period.



Kernels weight:

Kernels weight was significantly different between treatments. Alachlor + cultivation, metribuzin + cultivation, Trifluralin + cultivation and weeds completely control had highly and no control treatment had lower kernels weight. Variation of kernels weight was related to others components yield (Table 1).

Kernels yield:

This trait was significantly different between treatments. Hand weeded control and alachlor + cultivation had higher yield and no weed control and 2,4-D treatments had lower yield respectively. Results showed that an integration of two weed control methods (chemical and mechanical) was better than application of only one of them and provided higher yield (Table 1).

Table 1. Comparison of yield components at 5% probability (Duncan's test)

Treatments	No. Row/Cob	No. Kernels/Row	1000Kernels W. (g)	Kernels yield (kg/ha)
Alachlor	9.75c	34.73e	205bc	6483e
Alachlor+cultivation	15.30a	46.35a	262a	12890b
Metribuzin	9.12cd	33.58f	204bc	5914f
Metribuzin+cultivation	15.20a	45.06b	260a	12130c
Trifluralin	8.75cd	32.53g	203bc	5886f
Trifluralin+cultivation	15.00a	44.13c	258a	11930c
2,4-D	8.25de	31.36h	200bc	4930g
2,4-D+cultivation	13.38b	40.31d	231b	11480d
Completely control	15.50a	47.00a	239a	13420a
Without control	7.50e	29.75i	196c	4269h

Means followed by different letters in each column, have significantly different at 5% level of probability (Duncan's test).

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# Extremely reduced herbicide rates applied with an adjuvant for weed control in sugar beets

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## Introduction

Inadequate weed control in sugar beet has a devastating effect on yield and harvest and processing efficiency. Sugar beet is sensitive to weed competition and requires multiple, timely applications of several herbicides in combination. Optimum weed control with standard herbicide rates is often achieved at the expense of sugar beet injury. Also recommended herbicide combinations are relatively expensive (Dale et al., 1999). In the 1990s, a sugar beet weed control program with mixtures of extremely low herbicide rates with adjuvant or adjuvant combinations was developed at North Dakota State University and was widely accepted by sugar beet growers in the USA (Warner & Dexter, 1995; Dexter et al., 1996). This program has been called the “micro-rate” program and is a cost-effective way to achieve acceptable weed control. However, application timing and herbicide active ingredient and adjuvant selection according to weed species and biotypes present are key to achieve control (Dexter & Luecke, 2001).

## Materials and methods

Field experiments were initiated in 2003 (Woznica et al. 2004) and continued in 2004, near Poznan, Poland, to determine weed control efficacy, sugar beet tolerance and root and extractable sugar yield following herbicide treatments applied postemergence at much lower rates than what is commercially recommended. All reduced herbicide rate combinations included phenmedipham + desmedipham + ethofumesate at 30 + 30 + 30 g ai/ha (Betanal Progress AM 180 EC), triflusaluron at 4.5 g ai/ha (Safari 50WG), clopyralid at 33 g ai/ha (Lontrel 300 SL) + methylated seed oil (MSO) adjuvant at 1.5 L/ha (Atpolan BIO 80 EC). Soil residual herbicides, either lenacil at 160 g ai/ha (Venzar 80 WP) or metamiltron at 280 g ai/ha (Gladiator 70 WG), were included in selected reduced-rate treatments. The MSO adjuvant provided a spray mixture pH between 7.3-7.8. Conventional treatment included phenmedipham + desmedipham + ethofumesate at 90 + 90 + 90 g ai/ha (Betanal Progress AM 180 EC) and triflusaluron at 15 g ai/ha (Safari 50WG) without adjuvant, which followed the label recommendations. All herbicide combinations were applied broadcast and re-applied whenever the newly emerged weeds were at the cotyledon stage. Treatments were applied with a CO<sub>2</sub>-pressurized backpack sprayer equipped with four TeeJet-11002 flat-fan nozzles and calibrated to deliver 170 L/ha at 250 kPa. Plots were four row wide (4 x 0.45 m) and 9 m long, and treatments were in a randomized complete block design with four repetitions.

## Results and discussion

Sugar beet was infested mostly with *Chenopodium album*, *Solanum nigrum*, *Cirsium arvense*, and volunteer *Brassica campestris*. To achieve adequate weed control, weed control programs with reduced herbicide rates required five to four applications when soil residual herbicides lenacil and metamiltron were included, compared to four applications of recommended conventional herbicide combinations without adjuvant.

Sugar beet injury from low rates of herbicide combinations applied with adjuvant was less than or equal to the standard herbicide rates, and weed control varied between 72-95%, depending on herbicide mixture composition (Table 1). Inclusion of a soil residual herbicide, either lenacil or metamiltron, with the reduced rates of phenmedipham, desmedipham,

ethofumesate, triflusaluron, and clopyralid generally provided the best weed control and reduced the number of applications from five to four. Sugar beet root and extractable sugar yield following the low-rate herbicide combinations applied with MSO adjuvant were similar to the standard rate treatments applied without adjuvant. Relative cost of chemicals used for weed control program with reduced herbicide-rate combinations applied with MSO adjuvant was 35 to 53% lower than the cost of the standard-rate program applied without adjuvant.

These data confirm high effectiveness of the micro-rate program that has been registered and widely accepted by sugar beet growers in the USA. However, further experiments in different locations and environments need to be conducted to optimize this economically attractive and environmental-friendly weed control program for Polish conditions.

Table 1. Weed control, sugar beet injury and root yield, and relative cost of chemicals applied

Treatment (number of applications)	Rates g ai/ha or ml/ha (per single application)	Weed control %	Sugar beet injury %	Root yield t/ha	Relative cost of chemicals %*
Untreated weedy check	-	0	0	18	-
Untreated hand-weeded check	-	100	0	66	-
Phenmedipham+desmedipham+ethofumesate + triflusaluron (4x)	90+90+90 +15	99	15	64	100
Phenmedipham+desmedipham+ethofumesate + triflusaluron + MSO adjuvant (5x)	30+30+30 +4.5 +1500	72	8	59	47
Phenmedipham+desmedipham+ethofumesate + triflusaluron + clopyralid + MSO adjuvant (5x)	30+30+30 +4.5 +33 +1500	92	6	64	58
Phenmedipham+desmedipham+ethofumesate + triflusaluron + clopyralid + lenacil + MSO adjuvant (4x)	30+30+30 +4.5 +33 +160 +1500	95	10	62	53
Phenmedipham+desmedipham+ethofumesate + triflusaluron + clopyralid + metatritron + MSO adjuvant (4x)	30+30+30 +4.5 +33 +280 +1500	92	5	64	65
LSD (0.05)		9	4	8	-

\* cost of conventional treatment (phenmedipham+desmedipham+ethofumesate+triflusaluron at 90+90+90+15 g/ha) applied four times=100% (calculations were based on herbicide and adjuvant prices in Poland in 2004)

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## Recent advances in adjuvant formulation technology

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### Introduction

Many environmental, biological and application factors influence efficacy of postemergence herbicides (Nalewaja et al. 1989) Activator adjuvants added to postemergence herbicide spray mixtures often increase spray retention and/or foliar absorption, which results in enhanced herbicide efficacy (McWhorter 1982, Wanamarta & Penner 1989). However, a single-component adjuvant rarely affects all important factors responsible for herbicide retention, deposit formation, and absorption, and therefore does not optimise herbicide efficacy under a wide range of environmental, application, and biological conditions (Woznica et al. 2003). The aim of this work was to search for additive and synergistic interactions of adjuvants applied in mixtures, incorporate the most effective compounds into one stable adjuvant formulation, and evaluate such formulations with various herbicides for efficacy.

### Materials and methods

Laboratory, greenhouse and field experiments were conducted at North Dakota State University, Fargo USA, and Agricultural University of Poznan, Poland, in 1998-2004. A wide range of nonionic, cationic, and amphoteric surfactants, petroleum, vegetable, and modified vegetable oils, inorganic and organic fertilizers, pH modifiers, and humectants were tested with various herbicides for efficacy. Several thermostable, homogenous, and convenient-to-use multi-component liquid adjuvant formulations were invented for tank-mix application. These adjuvant formulations were tested in the greenhouse and field with various herbicides including several salts of glyphosate, nicosulfuron, rimsulfuron, foramsulfuron, clopyralid, flumetsulam, 2,4-D dimethylamine, dicamba sodium, bentazon sodium, clethodim, quizalofop-P, and atrazine. All experiments were conducted using a randomised complete block design with four repetitions.

### Results and discussion

AG-1 500 SL formulation was developed mainly for herbicides that contain glyphosate (Woznica & Woznica 2004). This homogenous, liquid adjuvant formulation includes ammonium sulphate, cationic surfactant, and pH buffer. Glyphosate (Roundup Ultra 360 SL and other glyphosate formulations) efficacy against *Elymus repens*, both in soft and hard water, in the field with AG-1 500 SL adjuvant at 1.5 L/ha was similar to or higher than the solid ammonium sulphate fertilizer. This adjuvant was also significantly more efficacious than the commercial fatty amine ethoxylate adjuvant, especially with the hard water spray carrier (Table 1). In field and greenhouse experiments with a similar adjuvant invention containing ammonium sulphate, amphoteric surfactant, and pH buffer (Woznica et al. 2003b), this convenient-to-use adjuvant blend was effective with various glyphosate salts, and also may be beneficial for 2,4-D, dicamba, bentazon and other herbicides (data not shown).

L-64 (Woznica et al. 2003a), L-132 (Woznica et al. 2004), and AG-2 (Szewczyk & Woznica 2004) were developed for herbicides that require an oil-based adjuvant for adequate efficacy. L-64 is a blend of methylated seed oil, urea/ammonium nitrate fertilizer, various surfactants, and a basic-pH adjuster, whereas L-132 is a concentrated surfactant and oil blend with a basic-pH buffer that is important for solubility of herbicides, mainly from the sulfonylurea group. AG-2 is a modified oil blended with a surfactant mixture that has superior oil-emulsifying and herbicide-solubilizing properties. Nicosulfuron + rimsulfuron +

clopyralid + flumetsulam with L-64 at 1% v/v and L-132 at 0.5% v/v in maize were more effective than with the reference commercial adjuvant Scoil at 1% v/v, without any visible maize injury (Figure 1). These adjuvants and AG-2 adjuvant also provided high efficacy of foramsulfuron, quizalofop-P, and atrazine (data not shown).

Table 1. Effect of adjuvants on *Elymus repens* (L.) control by glyphosate (Roundup Ultra 360 SL at 2 L/ha) (Agricultural Experimental Station, Brody, Poland, 2003 - average from two experiments)

Adjuvant	Soft water Ca <sup>2+</sup> < 20 mg/L	Hard water Ca <sup>2+</sup> = 350 mg/L
	<i>Elymus repens</i> control 6 weeks after treatment (%)	
None	68	33
Ammonium sulphate 5 kg/ha	84	80
Adbios 85 SL 1 L/ha*	85	41
AG-1 500 SL 1.5 L/ha**	95	90
LSD (0.05)	11	

\*fatty amine ethoxylate; \*\*liquid formulation of ammonium sulphate, cationic surfactant, and pH buffer

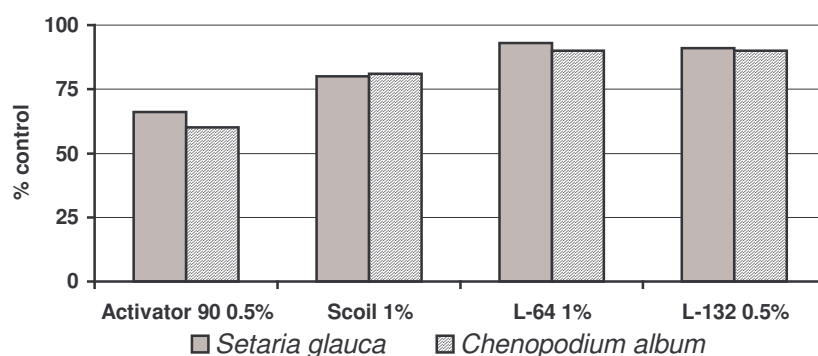


Figure 1. Weed control from nicosulfuron + rimsulfuron + clopyralid + flumetsulam (Accent Gold) as influenced by adjuvants (Activator 90 is a nonionic surfactant; Scoil is methylated seed oil; L-64 is a blend of methylated seed oil, urea/ammonium nitrate fertilizer, surfactants, and basic-pH buffers; L-132 is a basic-pH buffer, concentrated surfactant, and methylated seed oil blend).

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# Managing weeds in organically grown eggplant (*Solanum melongena* L.)

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## Introduction

High awareness of consumers about the negative side-effects of chemical pesticides, fertilizers, synthetic hormones, and genetically modified organisms shifted their preference to products that are organically cultivated (Neesen, 1998). It is projected that the area of organic farming will increase drastically in the near future, especially in Europe, North America and Australia (FAO, 2000). Weeds result in serious yield losses compared to other constraints in crop production (Abu-Irmaileh & El Kady, 1997). Various non-chemical weed control practices are favoured by organic farmers. Manual weed removal is slow and costly, rendering organic farming expensive and cumbersome. Soil solarization is a cost-effective method for the control of weeds and other pests (Abu-Irmaileh & Saghir, 1994). However, such practice is limited to the hottest months of the year. Fermenting organic matters, including animal manure placed in the furrows prior to planting effectively reduced weed populations in vegetables (Abu-Irmaileh & Abu-Rayyan, 2004). The objective of this study was to determine the effect of preplant fermentation of manure on weed populations in organically grown eggplant.

## Materials and methods

Field trials were conducted to evaluate the effect of preplant fermentation of fresh air-dry poultry manure placed in furrows for weed control in eggplant plots. Chemical fertilizers, pesticides or any other agricultural chemicals were not applied. In-furrow fermentation was conducted by thoroughly mixing organic materials with the soil to a depth of 20 cm in the planting furrows. The soil surface was either covered by black polyethylene sheets or left uncovered during the assigned period. The treatments included preplant fermentation of poultry manure for different periods (0, 2, 4, or 6 weeks prior to planting) at different rates (0, 5, and 10 kg m<sup>-2</sup>, with or without black plastic mulch removed after fermentation or kept onto soil throughout the crop cycle). Drip lines were 50 cm apart, with emitters 40 cm apart in-line. Soil thermometers were placed at a 15 cm depth in each plot. Weed species and dry weights of eggplants and weeds were recorded from the middle 1 m<sup>2</sup> of each plot during their flowering stages.

## Results and discussion

Fermenting poultry manure at 10 kg m<sup>-2</sup> under cover for six weeks prior to planting controlled weeds effectively and improved eggplant growth (Table 1). Better results were obtained when fermenting was followed by keeping the polyethylene sheets mulch films for the rest of the season when fermentation, at rates of 5 or 10 kg m<sup>-2</sup>, was done for 4 or 6 weeks. Weed control was significantly better at 10 kg m<sup>-2</sup>. This effect is thought to be due to the high soil temperature which was raised to about 47.7 °C at 15 cm depth and the resulting products of fermentation, such as organic acids and ammonia which can be toxic to many living organisms present within the manured top soil layer. Manure fermentation followed by retaining black polyethylene sheets inhibited further weed emergence and improved eggplant growth. Effectively controlled weeds included: *Amaranthus retroflexus* L., *Chenopodium album* L., *Chenopodium murale* L., *Polygonum aviculare* L., *Sonchus oleraceus* L., *Sisymbrium irio* L., *Centaurea pallescens* Del., *Lolium multiflorum* Lam., and *Malva parviflora* L. The following perennial weeds resisted to manure fermentation: *Cyperus esculentus* L., *Cynodon dactylon* (L.) Pers., *Melilotus indicus* (L.) All., *Prosopis farcta* (Banks



et Sol) Macbride, *Sorghum halepense* (L.) Pers., and *Vicia narbonensis* L. The underground vegetative propagules could have resisted the adverse effects of manure fermentation. Aerobic manure fermentation can elevate temperatures to about 63 °C and can effectively kill weed seeds within one week of composting (Bahman & Lesoing, 1999). It can be concluded that preplant fermentation of poultry manure can effectively control weeds, improve yield and serve the purpose of organic farming. This is an environmentally safe method as it drastically reduces the ill effects of spreading manure without proper management.

Table 1. The effect of different fermenting treatments at 10 kg m<sup>-2</sup> on dry weights of eggplant and weeds.

Treatment type	Fermenting period ( weeks)	Dry weight	
		Eggplant (g plant <sup>-1</sup> )	Weeds (g m <sup>-2</sup> )
Fermenting manure without cover	0	1.1 f	251.6 ab
	2	5.1 f	323.0 a
	4	10.2 f	362.9 a
	6	15.4 f	402.4 a
Fermenting manure under cover, then cover was removed	0	2.4 f	414.6 a
	2	7.9 f	460.3 a
	4	23.4 f	287.2 ab
	6	53.3 e	104.5 bc
Fermenting manure under cover, the cover was retained on surface for the rest of the season	0	102.4 d	11.3 c
	2	141.3 c	5.2 c
	4	228.8 b	0.1 c
	6	293.6 a	0.1 c

In the same column, means with same letters are not significantly different at P<0.05 (LSD test).

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## Weed management in organic sugar beet in rotation with other crops

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### Introduction

Agricultural foodstuffs obtained using techniques established by EU Reg. 2092 (enacted in 1991) have become widely accepted in Italy and such crops are grown over much larger areas than in other European countries. However, organic sugar beet is not grown in Italy in spite of the fact that organic sugar beet has been introduced elsewhere in Europe, particularly in Sweden, the UK, Denmark, Germany, etc. (Konig *et al.*, 2004). The use of conventional sugar in organically certified foodstuffs was permitted only during an initial period. Subsequently, it was necessary to use alternative sweeteners such as honey or to import organic sugar from outside Europe (Tugnoli, 2002). Organic cropping certainly requires greater professionalism on the part of the grower. In particular, for weed control it is necessary to adapt integrated strategies based on well planned crop rotation and properly timed weeding operations, etc. Unlike with sustainable management (Campagna, 2004), in the absence of herbicides, weed control must be based on reducing the number of weed seeds and vegetative organs of perennials in the soil, preparing the seedbed effectively, and conducting well-planned mechanical operations. In order to investigate the feasibility of organic cropping during a three-year period 2001-2003, a trial was conducted with sugar beet in a four-year rotation with wheat, sorghum and a bioinsecticidal crop on set-aside land. Manual weeding was not done, only operations with machinery available in the area, and already used by conventional growers, were used.

### Materials and methods

The trial was undertaken in the Po valley in the years 2001-2002-2003 on a total area of 4 ha characterised by clay soils (42% clay; 39% silt; 19% sand; 1.9% organic matter; pH 7.9). For each crop, strip tests were performed to compare conventional cultivation techniques (2500 m<sup>2</sup>) and organic techniques (7500 m<sup>2</sup>). The four-year rotation involved a sequence with ploughed-in bioinsecticidal crops as set-aside (*Raphanus sativus* var. Pegletta), sugar beet, wheat and sorghum. Assessments were made regularly on crop growth and weed emergence and development both where organic and conventional cropping were practised. The weeds were counted over 10 m<sup>2</sup> plots and repeated 10 times within the strip-test area. At the time of harvest, production tests were made over the entire area of both the organic and conventional trial.

### Results and discussion

Weather conditions differed during the period of experimentation with normal conditions prevailing in 2001 followed by 2002 and 2003 having abundant precipitation in late winter and early spring and very high temperatures in the beginning of the summer. In particular, 2003 was characterised by an absence of rain for the entire summer, whereas in 2002 there were abundant and frequent rainfall events during the summer.

Weed control - floristic surveys conducted on a periodic basis revealed (Table 1) larger numbers of weeds in early-sown sugar beet. In particular this was true for *Polygonum aviculare* and *Fallopia convolvulus*, which, however, were subsequently controlled by *Gastroidea poligoni*, present in large numbers due to the absence of insecticide application. With late-sown, crops numbers of more competitive weeds, such as *Amaranthus retroflexus* and *Echinochloa crus-galli*, were higher, probably because of the elimination of early

emerging weeds by mechanical methods. Harrowing conducted at the early growth stages of sugar beet eliminated weeds between the rows, whereas weeding along the row resulted solely in a reduction in weed numbers. Weed infestation was therefore concentrated prevalently along and in the rows. The higher sowing density resulted in a significant reduction in weed numbers and weed growth, but also had an adverse impact on beet root size. In conventionally grown crops, weed control was almost complete, including that of *Cuscuta campestris*, which is an insidious problem in organic cultivation. Weeds were effectively eliminated in both conventional and organic wheat. This was due initially to the good results obtained with the chain harrow and subsequently because of the competitiveness of the crop against the weeds, apart from problematic grass weeds such as *Alopecurus*, *Avena* and *Lolium*. A higher sowing density of wheat reduced weed emergence and subsequent growth. With sorghum, the same kind of difficulties occurred as with sugar beet in terms of lower weed control along the row, although sorghum appeared to be more competitive than beet, and chain harrowing was more effective against dicotyledons. By contrast, the grass weed *Echinochloa crus-galli* proved to be very competitive and difficult to control even under conventional cropping.

Harvesting tests – yield assessments over the entire plot area (Table 1) revealed a severe reduction in sugar beet root yield (on average more than 45%) compared with conventional sugar beets. This was observed with both early and late sowing, although the greatest losses occurred with the latest sown crops due to weed competition and the shortening of the growing period. This has a direct impact on the production of sucrose, in spite of the fact that good polarisation was also observed in organic sugar beets. Polarisation losses were more dependent on late sowing than cropping system, given that early harvesting allowed leaf spot damage to be reduced. The purity of the beet root pulp (PSD) was substantially higher in the organic crops, mainly due to the lower content of alpha-amino nitrogen in the beet roots as a result of the greater uptake of nitrogen by weeds. In wheat, an average three-year loss in production of nearly 25% was found along with a significant reduction in the specific gravity of the grain. This was attributable not only to the presence of weeds but also to the mild attacks of white mould and in particular rust. In sorghum, organic crop yields were similar to conventional yields in 2001, but were lower in the other 2 years because of more rain in the spring, which led to compaction of the soil making mechanical weed control less effective.

Table 1 – Weed surveys and harvesting tests conducted in the years 2001-02-03

Crops		Weeds number/10 m <sup>2</sup>							Harvesting index		
Sugar Beet		ECHCG	AMARE	CHEAL	FALCO	POLAV	POLLA	Others	Root	Pol.	Sugar
	Org1	0	0	61	33	42	52	43	61.5	100.6	61.8
	Org2	1	1	31	8	5	77	25	55.5	98.3	54.6
	Org3	7	15	16	4	2	45	23	45.5	95.5	43.4
	Conv	0	0	1	1	1	1	3	100	100	100
Wheat		ALOMY	AVELU	LOLMU	SINAR	PAPRH	MATCH	Others	Grain	Sp. grav.	
	Org	5	7	21	7	23	4	32	73.7	96.5	-
	Conv	0	0	3	0	0	0	1	100	100	-
Sorghum		ECHCG	AMARE	CHEAL	FALCO	POLAV	POLLA	Others	Grain	Sp. grav.	
	Org	9	7	4	1	0	37	8	68.1	94.7	-
	Conv	2	1	0	0	0	0	1	100	100	-

Organic 1-2-3: sowing sugarbeet sown 28 February – 15 March – 31 March; Others: *Compositae*, *Cruciferae*

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## Annual grass weed control in cotton by cereal cover crop mulches

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### Introduction

*Echinochloa crus-galli* and *Setaria verticillata* belong to the 10 most common and troublesome weeds of Greece (Damanakis, 1983). Cover crops have been used to manage weeds in several crops like cotton (Varco *et al.* 1999; White & Worsham, 1990). Rye (*Secale cereale*) is a commonly used cover crop that can reduce density and biomass of several weed species (Teasdale *et al.*, 1991). However, the use of cover crops from a farmers perspective must be justified economically by reduced herbicide input and/or by increased yield (Reddy, 2001). The objective of this research was to study the effect of 11 winter cereals as cover crop mulch on *E. crus-galli*, *S. verticillata*, *Digitaria sanguinalis* and cotton (*Gossypium hirsutum*) emergence and growth.

### Materials and methods

A field experiment was conducted during the 2001-2002 growing season at the University Farm of Thessaloniki, in northern Greece. Six triticale [Thisvi (Thi), Niovi (Nio), Vronti (Vro), Catria (Cat), Vrito (Vri), Artemis (Art)], three rye [Korytsa (Kor), Germany (Ger), Florina (Flo)], and two barley cultivars [Athenaida (Ath) and Thessaloniki (The)] were planted in fall and incorporated into the soil during spring 15 days before cotton planting. A split-plot design was deployed in a randomized complete block layout with four replicates. The experimental area was artificially infested by broadcasting 12 g m<sup>2</sup> of *E. crus-galli*, 6 g m<sup>2</sup> of *S. verticillata*, and 4 g m<sup>2</sup> of *D. sanguinalis* seeds, which were incorporated into the soil (5 cm deep) with a rotovator after cover crop destruction and before crop sowing. Cotton cultivar cv. Vered was sown in 90-cm row spacing. Two weeks after cotton emergence, the main plots were divided into subplots. Half of these subplots were treated with postemergence-applied quizalofop (Targa, Rhone Poulenc) 4 weeks after planting while the other half subplots were untreated. Also, crop and weed densities were determined 4 weeks after planting. Furthermore, *E. crus-galli*, *S. verticillata*, and *D. sanguinalis* plants were harvested in a 1 m<sup>2</sup> area in the two central rows of each subplot 12 and 16 weeks after crop planting. Weed stem number and fresh weight was measured at each sampling date.

### Results and discussion

Four weeks after crop planting, emergence of *E. crus-galli*, *S. verticillata*, and *D. sanguinalis* was reduced by 20 to 66%, 0 to 55%, and 25 to 87% respectively in plots where winter cereals had been incorporated, as compared with those of winter cereal-free plots (control) (data not show). Twelve weeks after cotton planting, total biomass of *E. crus-galli*, *S. verticillata*, and *D. sanguinalis* was reduced by 19 to 84%, 20 to 63% and 16 to 75%, respectively, in comparison with winter cereal-free treatment (Fig. 1). Similar results were reported by Barnes and Putnam (1983) who found that a living cover of spring planted rye reduced biomass of *D. sanguinalis* and *Chenopodium album* by 42 and 98% respectively, compared to no-rye treatment. Quizalofop application in half of the subplots gave excellent control of the three grasses. None of the cereal cover crop mulches had any detrimental effect on cotton emergence (Fig. 2). Cotton yield increased by 60 to 120% in subplots where winter cereals had been incorporated and not treated with graminicide, as compared with cotton grown in subplots free of cereals. In subplots treated with post-graminicide, cotton yield increased by 20 to 130%, as compared with the untreated ones (Fig. 2). The results of this study suggest that some winter cereals such as barley cultivar Athinaida could be used as

cover crop mulch for annual grass weed suppression in cotton and consequently to minimise herbicide application.

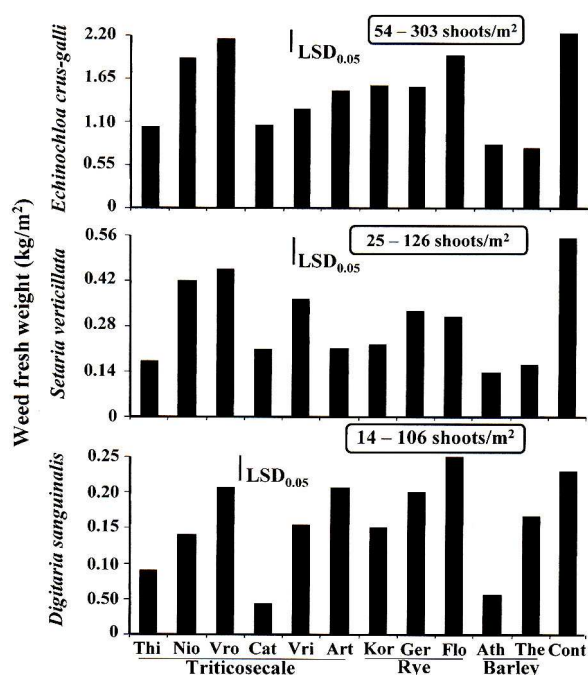


Fig. 1. Effect of 11 winter cereals used as cover crops on the fresh weight of three grass weeds in cotton (12 weeks after planting, WAP).

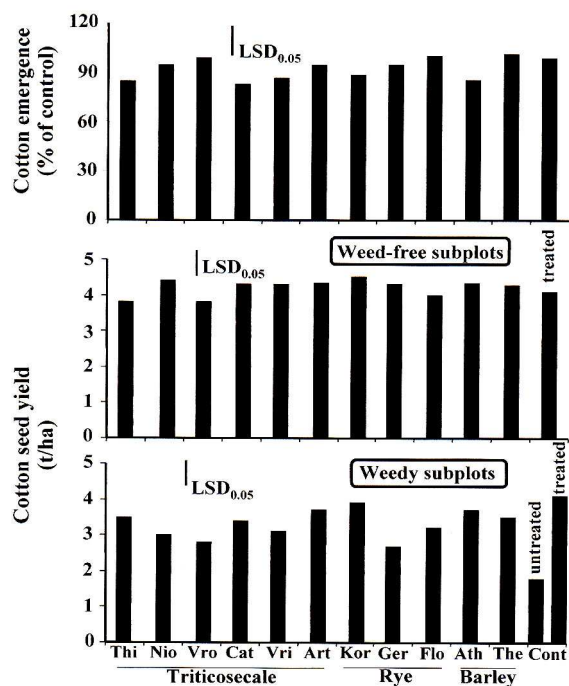


Fig. 2. Effect of 11 winter cereals used as cover crops on cotton emergence (4 WAP) and seed yield.

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# Organically grown durum wheat varieties under different intensity and time of mechanical weed control

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## Introduction

Mechanical weed control such as tine harrowing is an important option in organically grown winter cereals. As only partially weed suppressive, harrowing could be likely more effective, if integrated with other means, e.g. more competitive varieties. Such varieties have already been found in durum wheat (*Triticum durum* Desf.) (Paolini *et al.*, 2002) but their response to harrowing is still unknown. This study investigated the effects of variety, intensity and timing of tine harrowing on the efficacy of weed control in organically grown durum wheat.

## Materials and methods

A two-year study was conducted at Viterbo in 2001/2002 and 2002/2003. Both years, a factorial design with three replicates in randomized blocks was used, factors being: a) four durum wheat varieties: Colosseo and Nefer (more competitive), and Duilio and Creso (less competitive) (Paolini *et al.*, 2002 & unpublished); b) six weed control treatments: absent, chemical (tribenuron-methyl + diclofop-methyl at recommended dosages), intensive (tine angle lying 15° before the normal to the soil, i.e. + 15° C, Bàrberi *et al.*, 2000) early (11 February: 55 days after crop emergence, with broad-leaved weeds at a 3-5 true leaves stage) harrowing, intensive standard time (28 February) harrowing, moderate (tine angle lying 15° behind the normal to the soil, i.e. -15° C) early harrowing, and moderate standard time harrowing.

Wheat was drilled on a sandy-loam soil in plots of 18 m<sup>2</sup> on 28 November 2001 and 2 December 2002, placing 450 viable seeds m<sup>-2</sup> in rows 0.15 m apart. It was included in a wheat/cover crop/tomato/faba bean organic rotation, and given 100 and 70 kg ha<sup>-1</sup> P and N (phosphorite + organic fertilisers), respectively, partly at ploughing and partly top dressed. Crop plant mortality after harrowing and above-ground crop biomass and yield at harvest were assessed. Above-ground weed biomass, total and per species, at crop harvest was also obtained.

## Results and discussion

In the absence of control, on average 89 % of the proportion of total weed biomass at harvest (averaging 2.49 t ha<sup>-1</sup> DM over varieties) was broad-leaved species (mostly *Fumaria officinalis*, *Sinapis arvensis*, *Papaver rhoeas*, *Anchusa arvensis* and *Fallopia convolvulus* in decreasing order of abundance), and the remaining 11 % was *Lolium multiflorum*.

On average, intensive and moderate harrowing gave same weed biomass reduction (with a mean of 71 %). Harrowing was more effective in the more competitive varieties (Colosseo and Nefer), mainly when applied early (Tab. 1). Irrespective of variety, intensity and timing, harrowing increased the proportion of *L. multiflorum* in surviving weed biomass. Chemical control was 100% effective in all varieties. At both harrowing times, intensive harrowing gave higher crop plant mortality in less competitive varieties (Duilio and Creso, owing to smaller plants) (Tab. 1).

Averaged over years, in the control absence DM yield as percent of that under chemical control (weed-free crop) was higher in Colosseo (59 %) than in Duilio and Creso (averaging 42 %) ( $P < 0.05$ ), while Nefer gave 53 %. Chemical control gave similar DM yield across



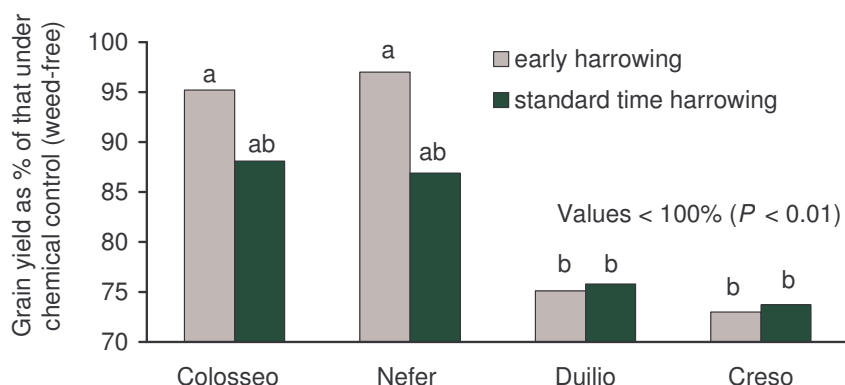
varieties in 2001/2002 (averaging 5.35 t ha<sup>-1</sup>), but DM yield was lower in Nefer and Creso than in the earlier Colosseo and Duilio (averaging 3.68 vs 4.32 t ha<sup>-1</sup>) in the drier 2002/2003.

Averaged over harrowing intensities and years, the yield of Colosseo and Nefer was similar to that achieved with chemical control under both early and standard time harrowing (Fig. 1). Instead, under the same conditions, Duilio and Creso gave significant higher yield loss (26% on average) than early harrowed Colosseo and Nefer.

**Table 1** Weed suppression and plant mortality in 4 durum wheat varieties harrowed at different intensities and timings averaging over 2 years. Means with different letters are significantly different ( $P < 0.05$ )

Variety <sup>(1)</sup>	Weed biomass suppression (%) <sup>(2)</sup> (average of 2 h. intensities)		<i>L. multiflorum</i> biomass (%) of total weed biomass at harvest) <sup>(3)</sup>	Crop plant mortality(%) <sup>(4)</sup> (average of 2 h. times)	
	Early harrowing	Standard time harrowing		Intensive harrowing	Moderate harrowing
	Colosseo (MC, <i>me</i> )	91.1 a	74.2 c	37	9.3 bc
Nefer (MC, <i>ml</i> )	87.9 ab	76.3 bc	43	9.1 c	6.2 cd
Duilio (LC, <i>me</i> )	59.9 d	60.5 d	38	14.9 a	5.4 d
Creso (LC, <i>ml</i> )	63.7 cd	54.6 d	44	12.8 ab	5.3 d

<sup>(1)</sup> MC, LC = more or less competitive (Paolini et al., 2002 & unpublished); *me*, *ml* = medium-early or late; <sup>(2)</sup> complement to 1 of the ratio *biomass under control absent/biomass after harrowing* at harvest; <sup>(3)</sup> all values higher ( $P < 0.05$ ) than that under control absent (9 -15 % depending on variety); <sup>(4)</sup> complement to 1 of the ratio *plants m<sup>-2</sup> 15 d after harrowing/plants m<sup>-2</sup> 4 weeks after emergence*.



**Figure 1** The effect of time of harrowing on the yield of 4 durum wheat varieties, averaged over 2 intensities (severe or moderate) and years (arcsin back-transformed means). Values without letters in common are different at  $P < 0.05$

Results suggest that time of harrowing is more important than its intensity in ensuring good weed control in durum wheat. Intensive harrowing does not seem to be more weed suppressive under light soil conditions, but is likely more expensive and may increase the risks of crop plant failure. However, effective harrowing seems to be related to early application in highly competitive varieties, likely more tolerant to tine injuries and more suppressive against surviving weed plants.

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# Effect of soil tillage on weed infestation in sown and transplanted horticultural crops

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## Introduction

On sandy-loam soils of the Tirrenic coastal areas where irrigated, conventionally grown horticultural crops follow winter wheat, minimum instead of traditional tillage is an attracting way to lower costs, maintain soil fertility (Holland, 2004) and ensure timely planting with no yield decrease (Paolini *et al.*, 2004). Minimum tillage, however, could increase weed control problems, if reliable, fully selective herbicides are not available (Paolini & Faustini, 2005) or allowed (organic farming). In these cases, control efficacy also likely depends on crop competitiveness, i.e. on its planting method and early growth. This study tested the effect of ploughing and disk harrowing on weed infestation in sown or transplanted horticultural crops.

## Materials and methods

The study was carried out at Viterbo in 2002 and 2003 on a sandy-loam soil hosting a two-year rotation winter wheat/summer horticultural crops/spring pepper, managed with either traditional (ploughing) or minimum tillage (disk harrowing). Each season, a split-plot design with three replicates in randomized blocks was used, factors being: a) two tillage techniques (main plots): ploughing at 35 cm depth and disk harrowing at 15 cm; b) six horticultural crops following wheat (sub-plots): the single row transplanted carrot, cauliflower, fennel and lettuce, and the single row sown (30 and 15 cm apart) French bean and radish; c) two levels of natural weed infestation (sub-sub-plots): present or absent (weed-free crop manually weeded).

After mineral fertilisation and seedbed preparation, the crops were planted in sub-plots of 18 m<sup>2</sup> on 7 September 2002 and 9 September 2003 and grown with irrigation. Crop and weed biomass, total and per species, were measured 4 and 7 weeks after planting.

## Results and discussion

Both years, in weedy plots main species were *Setaria viridis*, *Solanum nigrum*, *Amaranthus hybridus*, *Stellaria media* and *Convolvulus arvensis* in decreasing order of abundance.

As average over years, at both assessment times ploughing resulted in lower infestation in the weedy plots in all crops except for lettuce (Tab. 1). Four weeks after planting, the low-density transplanted cauliflower and the sown French bean and radish gave the highest weed

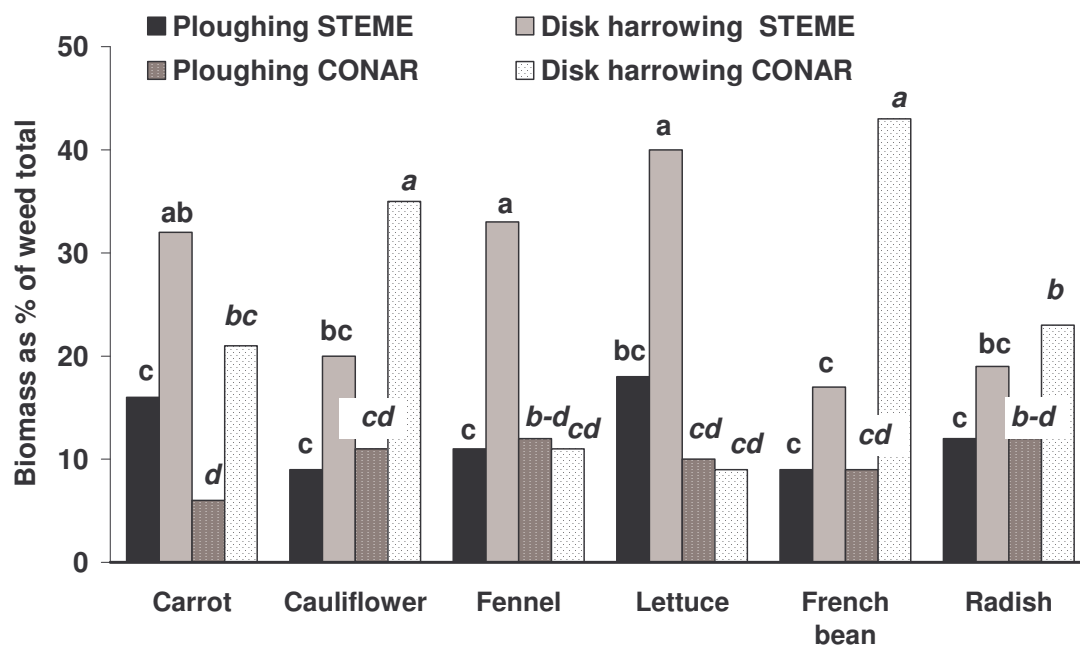
**Table 1** Weed biomass (t ha<sup>-1</sup>, DM) 4 and 7 weeks after planting (WAP) and crop biomass decrease (in italics) 4 WAP as % of weed-free ( $\sqrt{\quad}$  back-transformed means) of six weedy horticultural crops after two soil tillage techniques, averaged over two years. Values with no common letters are different at  $P < 0.05$  (LSD test for either 4 or 7 WAP). \*:  $> 0$ ,  $P < 0.05$

Crop/variety/final density (plants m <sup>-2</sup> )	4 WAP				7 WAP	
	Ploughing		Disk harrowing		Ploughing	Disk harrowing
Carrot/Nantese/18	0.49 g	6.3	0.78 b-d	<i>9.1</i>	0.99 fg	1.46 cd
Cauliflower/Gitano/2	0.71 d-f	8.7	0.86 a-c	<i>14.1*</i>	1.41 cd	2.21 a
Fennel/Vittorio/8	0.52 g	6.4	0.92 ab	<i>11.9*</i>	1.06 e-g	1.85 b
Lettuce/Integral/7.15	0.55 g	4.9	0.61 e-g	<i>3.6</i>	0.84 g	0.95 g
French bean/Cleo/40	0.73 c-e	9.5	0.99 a	<i>15.8*</i>	1.24 d-f	1.62 bc
Radish/Saxa/360	0.77 cd	<i>10.6*</i>	0.96 a	<i>16.0*</i>	1.27 de	1.74 b

biomass after ploughing, with moderate but significant increase after harrowing. The other relatively high-density transplanted crops gave least weed biomass after ploughing, with clear (carrot and fennel) or negligible (lettuce) increase by harrowing. Seven weeks after planting, lettuce also gave least and similar weed biomass with both tillage techniques, while all others gave higher weed biomass after harrowing, with greater increase for cauliflower and fennel.

After both ploughing and harrowing, weeds had significantly affected the biomass of radish 4 weeks after planting (Tab. 1). The same had occurred for French bean, cauliflower and fennel after harrowing but not after ploughing.

Harrowing gave higher abundance of *S. media* 4 weeks after planting in carrot, fennel and lettuce (Fig. 1). The same occurred for *C. arvensis* in carrot, cauliflower and French bean.



**Figure 1** Biomass of *S. media* (STEME) and *C. arvensis* (CONAR) as percent of total weed biomass in six weedy horticultural crops 4 weeks after crop planting, averaged over two years. Figures are arcsin back-transformed means. Values with no letters in common are different at  $P < 0.05$  [LSD test separately for STEME and CONAR (letters in italics)]

Compared to disk harrowing, in various cases ploughing was more effective in containing weed biomass at early crop stages, likely giving the cultivated plant a competitive advantage which could favour the efficacy of partially suppressive means. Harrowing also seemed to increase the relative abundance of some species, evidently favoured by lower soil disturbance.

However, results also suggest that ploughing sometimes gives little or even negligible benefit, which does not justify omitting the advantages of minimum tillage prevailing under certain conditions. This can occur when the crop, for pre-determined conditions, exerts high or, alternatively, poor competition. In our case lettuce gave quick growth and soil cover, and did not allow likely higher weed emergence after harrowing to result in higher weed biomass. On another hand, weed biomass in cauliflower differed little across tillage as low crop density constrained it poorly anyway: a partially suppressive mean alternative to ploughing (early hoeing and/or stale seed bed) could be likely more appropriate.

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# Non-chemical weed management in carrot

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## Introduction

Plants, like carrot with slow initial development, with a long growing period and poor weed tolerance, are very sensitive for weediness. And moreover, carrot is an important product of organic farming. Because of environmental aspects and because of the increasing demand for organically produced vegetables, more farmers convert their conventional farming systems into organic ones, partly because they want to leave out herbicides from cropping.

## Materials and methods

The objective of a 4-year field experimentation was to find the best combination of methods that are allowed for weed control in organic farming. Physical methods were compared with herbicide treatments as well. Fifteen treatments with 4 replicates were examined. Herbicide (as control) was applied pre-emergence with a mixture of 20mL 100 m<sup>-2</sup> S-metolachlor (Dual Gold 960 g a.i. L<sup>-1</sup>) and 20 g 100 m<sup>-2</sup> chlorbromuron (Maloran a.i. 50%). Used carrot variety was *Nanti* grown at 75 cm row spacing. Sowing depth was 3 cm.

Different treatments were used in crop rows and in the inter-rows. Treatments in the rows were: hand weeding and flaming, herbicide treatment and an untreated. Treatments in the inter-rows were: cultivator, hoeing, flaming and brush weeding. The same treatments were used as control as for weeds in the rows. The soil type was a chernozem-like sandy soil. Soil forming rock is calcareous sand. Precipitation differed between years.

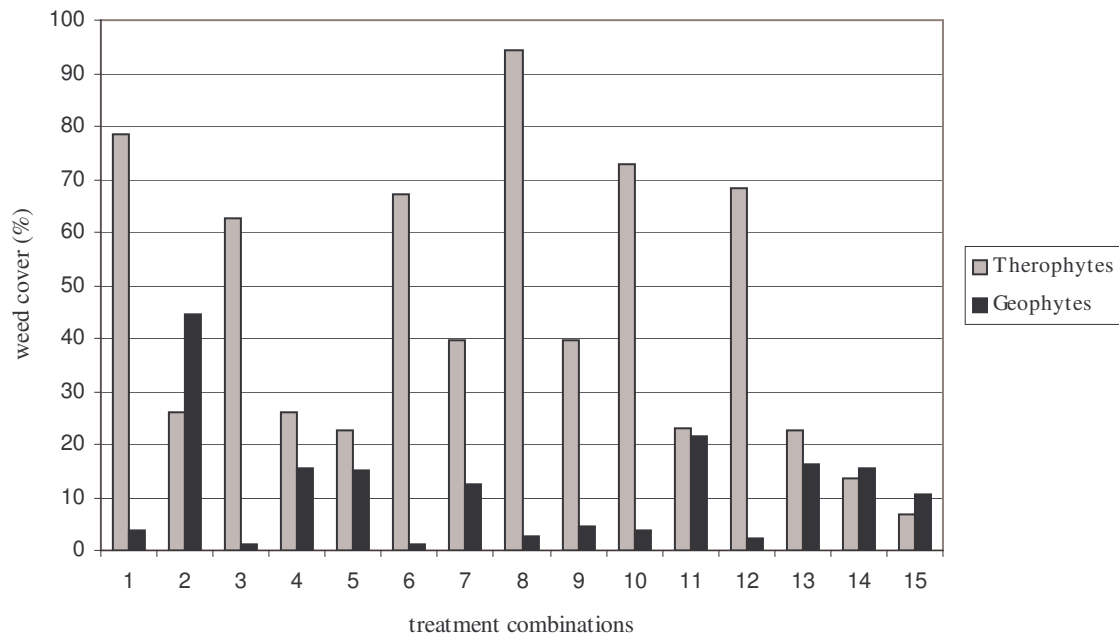
Weeds were assessed right before and two weeks after treatments by obtaining dry matter of weeds and of root and canopy of carrot. SPSS 9.0 program was used to analyse data and Tukey's test to compare means.

## Results and discussion

Two hand weedings during the growing season caused the lowest weed cover in the rows. It seems that interrow treatments had more significant effect on the yield than in-row treatments, because treatment-combinations with the same in-row treatment and with different inter-row treatments showed very different results. Cultivator and hoeing had better effects on carrot growing than brush weeding, so inter-row tillage serves not just weed control purposes but is also good for crop growth. Apart from the fact that mechanical weed control removes competitive weeds from the inter-rows, the tillage it causes is beneficial to the crop, especially the tillage caused by the cultivator and the hoe.

Only in one year out of the four years, brush weeding was more effective against inter-row weeds than the cultivator. That particular year was the driest one. In the other 3 years, the cultivator was most effective and effects persisted longer.

Mechanical weed control twice in the inter-rows increased coverage of geophyte-perennial weeds compared with their pair: the same in-row treatment but only one mechanical inter-row treatment (Fig. 1.). These pairs e.g. treatments 6 and 7, or treatments 10 and 11, etc. It is explainable with that stolons and rhizomes were disintegrated and thereby these weeds were propagated. The same treatment had the opposite effect on therophyte annual weeds that propagate with seeds: efficiently reduced their presence in all cases.



- Legend:*
- |  |   |
|--|---|
| <ol style="list-style-type: none"> <li>1. Control</li> <li>2. Herbicide</li> <li>3. Herbicide in rows + cultivator in interrows 1x</li> <li>4. Herbicide in rows + brush hoe in interrows 1x</li> <li>5. Herbicide in rows + hoeing in interrows 1x</li> <li>6. Weeding in rows 1x + cultivator in interrows 1x</li> <li>7. Weeding in rows 1x + cultivator in interrows 2x</li> <li>8. Weed flaming + cultivator in interrows 1x</li> </ol> | <ol style="list-style-type: none"> <li>9. Flaming + cultivator in interrows 2x</li> <li>10. Weeding in rows 1x + brush hoe in interrows 1x</li> <li>11. Weeding in rows 1x + brush hoe in interrows 2x</li> <li>12. Flaming + brush hoe in interrows 1x</li> <li>13. Flaming + weed brush in interrows 2x</li> <li>14. Weeding in rows according to need + brush hoe in interrows 2x</li> <li>15. Weeding in rows according to need + cultivator in interrows 2x</li> </ol> |
|--|---|

Figure 1. Weed cover according to life form groups in inter-rows in 2001.

Higher weed occurrence was observed in all cases when the cultivator or brush hoe was combined with flaming in inter-rows. When looking at these findings, inter-row flaming seems not feasible.

Because of extremely variable climatic conditions, it is very difficult to extract consistent results. On overall we can conclude that there is no definite and completely reliable method for physical weed management in carrot. Farmers’ skills and good judgements of the actual need for weed control play the most important role. According to our results, traditional hand weeding of weeds in the rows and hoeing against inter-row weeds appear to be the best combination in terms of weeding effectiveness and preserving carrot yield.

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OTKA T 030346 supported experiment

# Mechanical control of *Cirsium arvense* in organic farming

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## Introduction

*Cirsium arvense* L. (Scop) is a troublesome weed wherever it is found in agro-ecosystems (Donald, 1994; Skinner *et al.*, 2000). In conventional agricultural systems, targeted use of herbicides during the growing season can provide satisfactory control, whereas the problem tends to increase in organic growing systems. A very expansive root system and the ability to form new aerial shoots from root buds, facilitate the formation of dense patches only a few years after establishment (Bakker, 1960). Furthermore, the high presence of labile carbohydrates in the roots of *C. arvense* enables the plant to regenerate even from root fragments 10 mm long (Hamdoun, 1972). It has been suggested that the amount of labile carbohydrates varies across the season (McAllister & Haderlie, 1985) and further that minimum regrowth of underground regenerative organs, hence the time when the plant is most susceptible to removal of aboveground plant tissue, occurs when the aerial shoot has approximately eight expanded leaves (Gustavsson, 1997). Here, it is hypothesized that the use of repeated mechanical control events, removing aboveground biomass, are likely to deplete the carbohydrates of the root system.

## Materials and methods

A two-factor experiment was conducted on a sandy loam in two consecutive periods from 2000-2002 and 2001-2003 at two adjacent experimental sites, hereafter called EXP1 and EXP2, respectively. The first factor consisted of three levels of mechanical weed control and untreated control plots. The second factor included plots sown with a grass/white clover mixture and plots with unsown stubble of spring barley, making up a total of 32 plots including four replicates.

The mechanical treatment consisted of three different numbers of mowings, two, four and six respectively, carried out from mid May until end of July in the second experimental year. Mowing was carried out whenever the majority of the shoots of *C. arvense* had reached a height of 10 cm. After ending the treatments the field was left untouched until November, when the field was ploughed. The following spring, i.e. in the third experimental year, the plots were re-established from fixed points along the field. All plots were fertilised with pelleted chicken manure, corresponding approximately to 70 kg N ha<sup>-1</sup>, just before sowing of spring barley. In the third experimental year no control of *C. arvense* was carried out. Just before harvest, the number of above ground shoots of *C. arvense* within the plots was counted and their biomass estimated.

## Results and discussion

In EXP1 a regression line fitted to data, explained 87.6% and 89.5% of the variation in the aboveground biomass of *C. arvense* when mowed in unsown stubble and in the white clover/grass mixture, respectively (Fig. 1). In EXP2 none of the suggested regressions lines fitted significantly to the data, even though the maximum number of mowings (six) reduced the above ground biomass of *C. arvense* by 64% and 69% in unsown stubble and in grass/white clover mixture respectively, when compared to no mowing (Fig. 1). A power-test showed values below 0.8, thus the null hypothesis – no treatment effect – should be accepted



cautiously. A comparison of the two regression lines within each experiment showed that, in both cases, there was no difference between the regression coefficients, whereas the intercepts differed significantly from each other, suggesting that the presence of a competitive crop, here a grass/white clover mixture, is likely to suppress the growth of *C. arvensis* significantly.

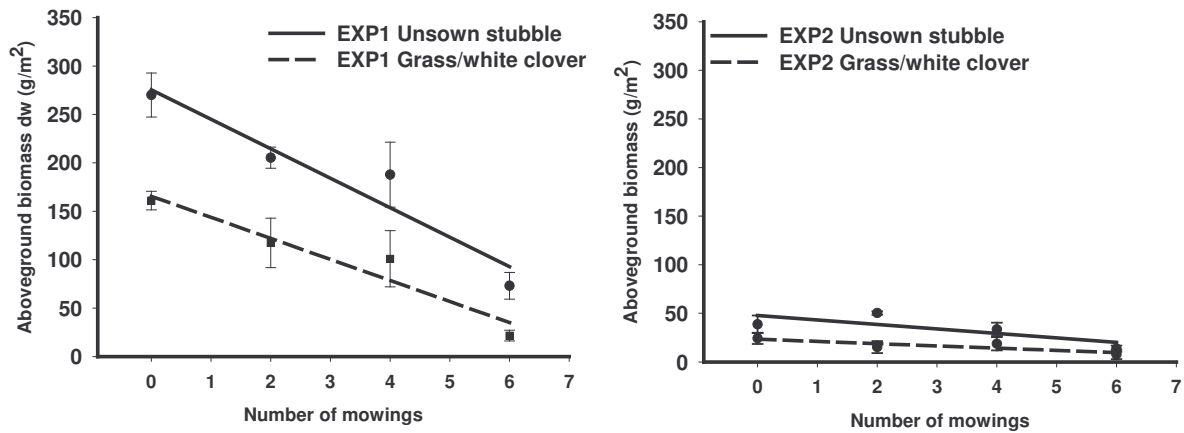


Fig. 1. Relationship between number of mowings and aboveground biomass of *C. arvensis* in the subsequent year. Treatments were carried out in plots with a grass/white clover mixture and in plots with unsown stubble.

Our results support the hypothesis that a continuous depletion of carbohydrates from the root system, caused by mowing and/or the use of a competitive crop, will diminish the regrowth capacity of the plant. Assuming that the root energy reserves attain a minimum during springtime (Gustavsson, 1997), any control strategy would be most effective at this time of year. However, Bourdot *et al.* (1998) reported that late season mowing had the most severe impact on root biomass. It is likely though, that the number of treatments confounded the effect of timing, as it was the case in the present study.

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# Tolerance to weed harrowing in spring barley genotypes

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## Introduction

Controlling weeds in spring cereals grown under organic conditions is mostly done by post-emergence weed harrowing, where spring tines of the weed harrow control weeds by uprooting and/or covering small weed plants with soil. In situations with relatively large weed plants and relatively small crop plants, there are increased risks of crop damage by soil cover or other mechanical damages to the crop leaves. These damages are increasing with increasing weed control intensity, and result in reduced crop growth immediately after weed harrowing. There are risks that the reduced growth reduces final crop yield too. However, there is some evidence that there are varietal differences in the tolerance to weed harrowing and that tolerance is negatively correlated with competitiveness against weeds (Rasmussen *et al.*, 2004). The aim of this study was to estimate the damages by weed harrowing in four pure genotypes and three two- or one three-component mixtures of spring barley, and to analyze if there were differences in tolerance to weed harrowing between the genotypes and mixtures.

## Materials and methods

Four pure genotypes, three two-component mixtures, and one three-component mixture of spring barley genotypes were examined for differences in tolerance to weed harrowing in field trials at Research Centre Flakkebjerg in 2003 and 2004. The field trials were designed as a split-split-split-plot-design in combination with an  $\alpha$ -plan. Every whole plot contained combinations of two levels of mechanical weed control (with and without a pre-emergence harrowing and one post emergence weed harrowing); two levels of pesticide treatment (with and without herbicides and fungicides) and two levels of nutrient level (40% or 80% of the recommended nutrient rate).

Tolerance to weed harrowing was measured as an immediate effect (how much of the plant was covered with soil after weed harrowing), a short-term effect (growth rate after harrowing), and a long-term effect (effect on yield). To estimate the degree of soil covering and crop growth after weed harrowing, reflectance measurements were conducted immediately prior and after the post emergence weed harrowing with a CropScan MSR16R instrument (CropScan Inc., Rochester MN 55906 USA). In the following three weeks, four measurements were conducted to measure the barley re-growth after the harrowing. Red Edge Inflexion Point (REIP) was estimated from the reflectance measurements and growing degree days (GDD) was used as the time-scale in the re-growth analysis. The reason for using REIP is the close correlation to chlorophyll content in the crop (Gitelson *et al.*, 1996). Soil covering was estimated as the difference between REIP immediate before and after weed harrowing ( $\Delta$ REIP). The growth rate in the following weeks were estimated as a linear correlation between REIP and time (GDD).

## Results and discussion

Results from the two-year field studies showed that there were varietal differences in the tolerance to mechanical weed control in the immediate effect as well as the short term effect, however there were marked differences in the immediate and short term effects between the two years (Fig. 1). In this figure there was, for 2004, a correlation between the degree of soil cover ( $\Delta$ REIP) due to weed harrowing, and the growth rate after weed harrowing, which indicate that varieties which are covered more by weed harrowing are able to compensate by

an increasing growth after harrowing. However the same correlation was not found in 2003. Regarding the long-term effect of weed harrowing on yield, there was no significant differences in 2003 but in 2004, cv. Brazil and the three-component mixture suffered significantly from weed harrowing while cv. Modena, cv. Otira and the cv. Modena + Orthega mixture seemed to benefit from weed harrowing, although not significantly. These differences are probably due to differences in growth habit at the time for weed harrowing.

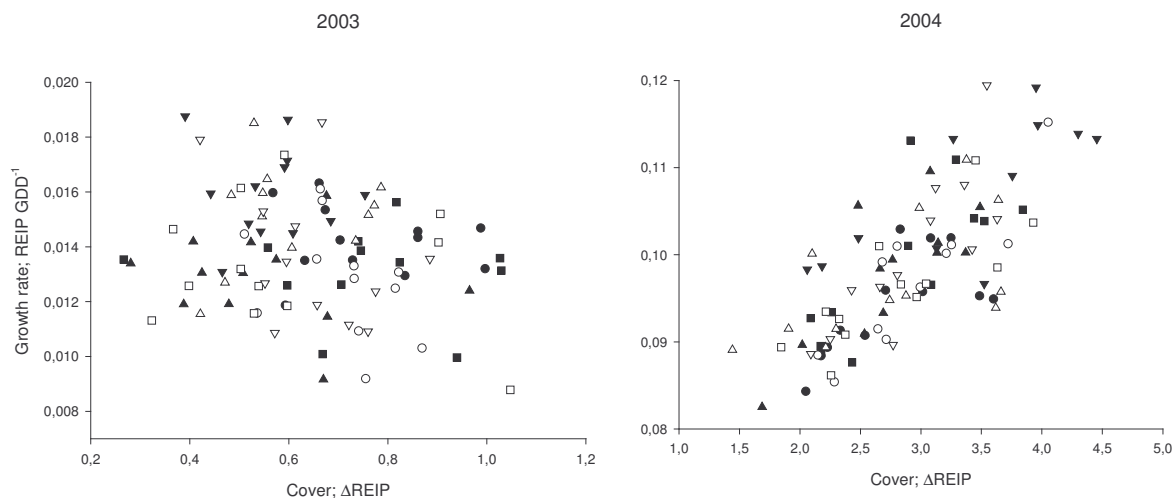


Fig. 1. Immediate effect measured as reduction in Red Edge Inflexion Point ( $\Delta$ REIP) versus growth rate of Modena ( $\bullet$ ), Otira ( $\blacksquare$ ), Orthega ( $\blacktriangle$ ) Brazil ( $\blacktriangledown$ ), 50% Modena + 50% Otira ( $\circ$ ), 50% Modena + 50% Orthega ( $\square$ ), 50% Modena + 50% Brazil ( $\triangle$ ) and 33% Modena + 33% Otira + 33% Orthega ( $\nabla$ ) in the 3-weeks period after weed harrowing in chemically-treated plots. Please note different scales on the x and y axes.

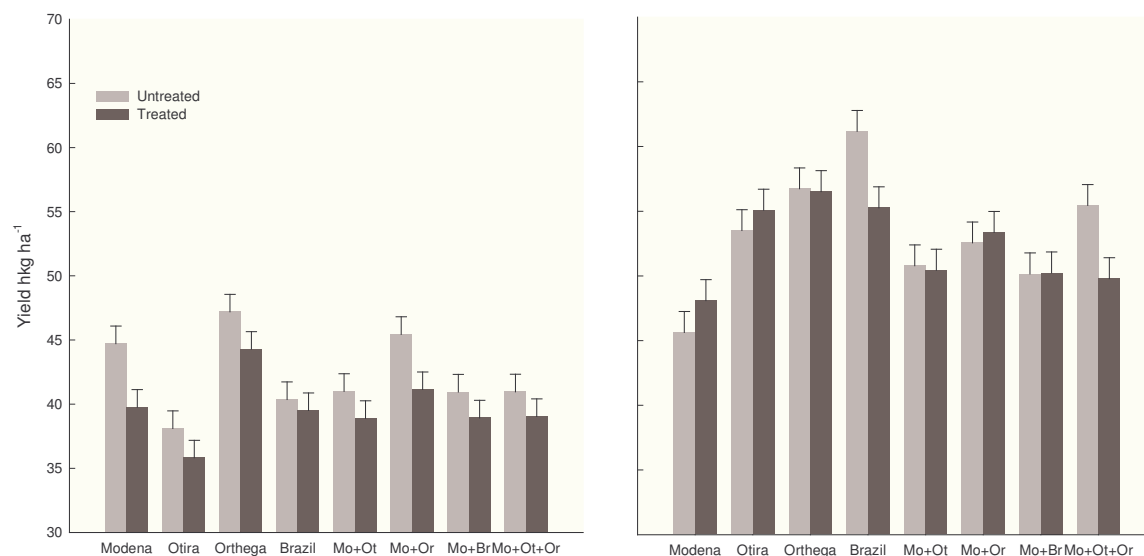


Fig. 2. Yield of the genotypes and mixtures in 2003 and 2004 in chemically-treated plots (i.e. no influence from weed competition). Light grey bars indicate mean yield from two levels of slurry application in plots *without* weed harrowing and dark grey bars indicate plots *with* weed harrowing. Please note a general yield *decrease* in weed harrowed plots in 2003, and a yield *increase* in harrowed plots in 2004 with cv. Modena and Otira and the cv. Modena + Orthega mixture (not significant) and an *decrease* in cv. Brazil and the cv. Modena + Otira +Orthega mixture ( $P < 0.01$ ).

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## Weed management under system of rice intensification

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### Introduction

The ecological system of rice intensification (SRI) developed in Madagascar has given remarkably good rice yields of 6 to 10 t ha<sup>-1</sup> in irrigated conditions (Stoop *et al.*, 2002). Fourteen day-old seedlings are normally transplanted at one seedling hill<sup>-1</sup> under wider spacing in the system of rice intensification (SRI) to exploit the full yielding potential of the rice crop (Rabenandrasana, 1999). Moreover, judicious use of water (alternate wetting and drying) is practiced under SRI for better input use efficiency; the above situation, however, favours weed growth (Uphoff, 2002). Hence, weed management under SRI is more crucial than under the normal system of planting.

### Materials and methods

Field experiments were conducted at the Experimental Farm, Department of Agronomy, Annamalai University to evolve an effective weed management practice under the System of Rice Intensification (SRI). The study was conducted in two seasons during 'Kuruvai' (June to October 2003) and 'Navarai' (November 2003 to March 2004). The experiment consisted of three main treatments, *viz.* 20x20 cm (M<sub>1</sub>), 25x25 cm (M<sub>2</sub>), and 30x30 cm (M<sub>3</sub>) spacings and five sub treatments, *viz.* weedy check (S<sub>1</sub>), hand weeding twice on 20 and 40 days after transplanting (DAT) (S<sub>2</sub>), weeding three times using conoweeder on 15, 30 and 45 DAT (S<sub>3</sub>), butachlor at 1.5 kg a.i. ha<sup>-1</sup> 3 DAT + hand weeding 30 DAT (S<sub>4</sub>) and pyrazosulfuron- ethyl at 30 g a.i. ha<sup>-1</sup> 3 DAT + hand weeding 30 DAT (S<sub>5</sub>).

### Results and discussion

Weeds in the experimental field included *Echinochloa colonum*, *Leptochloa chinensis*, *Cyperus difformis*, *Cyperus rotundus*, *Sphenoclea zeylanica* and *Bergia capensis*. Other weeds, *viz.* *Cyperus iria*, *Marselia quadrifolia* and *Eclipta alba* were also observed. In terms of weed control efficiency, among the main treatments the 20 x 20 cm (M<sub>1</sub>) spacing showed the highest weed control efficiency in both seasons. Among weed control measures, weeding three times using conoweeder on 15, 30 and 45 DAT showed the maximum weed control efficiency (84.9 and 84.7% in the 'Kuruvai' and 'Navarai' seasons, respectively).

Plant spacing had a significant influence on the yield components of rice. M<sub>2</sub> (25x25 cm) spacing showed the highest number of panicles hill<sup>-1</sup> (16.9 and 14.0), number of grains panicle<sup>-1</sup> (91.5 and 88.7), 1000 grain weight (22.6 and 22.3 g) and number of filled grains panicle<sup>-1</sup> (87.5 and 78.0) in Kuruvai and Navarai seasons, respectively. This might be due to optimum plant density, which might have resulted in less competition among the plants for nutrients at wider spacing. SRI seedlings are transplanted singly with a spacing of 25 x 25 cm so that there is no competition among plant roots to inhibit growth.

With respect to weed control measures, application of pyrazosulfuron ethyl at 30 g a.i. ha<sup>-1</sup> at 3 DAT + hand weeding at 30 DAT (S<sub>5</sub>) resulted in the highest number of panicles hill<sup>-1</sup> (16.7 and 14.7), number of grains panicle<sup>-1</sup> (97.6 and 92.4), 1000 grain weight (22.8 and 22.7 g), and number of filled grains panicle<sup>-1</sup> (90.3 and 81.5) in the Kuruvai and Navarai seasons. Good weed control by pyrazosulfuron-ethyl during the early period of rice crop and control of late emerging weeds by hand weeding reduced weed pressure, which in turn promoted crop growth.

Crop spacing significantly influenced the grain yield of rice. In both seasons, the highest grain yield was achieved with M<sub>2</sub> (4.9 t ha<sup>-1</sup> in Kuruvai and 4.1 t ha<sup>-1</sup> in Navarai; Tab. 1). Optimum plant population and increased specific leaf weight influenced the photosynthetic rate of transplanted rice. This finding was in line with the report of Neelamkumar Chopra and Nisha Chopra (2000). Among the weed control measures, application of pyrazosulfuron-ethyl at 30 g a.i ha<sup>-1</sup> at 3 DAT + hand weeding at 30 DAT (S<sub>5</sub>) showed the highest grain yield (5.5 and 4.7 t ha<sup>-1</sup> in the Kuruvai and Navarai seasons, respectively).

Table 1. Effect of crop spacing and weed control measures on grain yield of rice in the Kuruvai and Navarai seasons.

Treatments	Grain yield (t ha <sup>-1</sup> )	
	Kuruvai	Navarai
<i>Crop spacing</i>		
M <sub>1</sub> (20 x 20 cm)	3.75	3.19
M <sub>2</sub> (25 x 25 cm)	4.93	4.07
M <sub>3</sub> (30 x 30 cm)	3.90	3.22
SED (P < 0.05)	0.04	0.03
<i>Weed control measures</i>		
S <sub>1</sub> (weedy check)	2.71	2.28
S <sub>2</sub> (hand weeding twice at 20 & 40 DAT)	3.79	2.58
S <sub>3</sub> (weeding three times using conoweeder at 15, 30 & 45 DAT)	4.87	4.22
S <sub>4</sub> (butachlor 1.5 kg a.i. ha <sup>-1</sup> at 3 DAT + hand weeding at 30 DAT;	4.14	3.82
S <sub>5</sub> (pyrazosulfuron-ethyl 30 g a.i. ha <sup>-1</sup> at 3 DAT + hand weeding at 30 DAT)	5.46	4.71
SED (P < 0.05)	0.05	0.04

Fig. 1 clearly points out the monetary advantage associated to the different treatments. Spacing of 25x25 cm along with application of pyrazosulfuron ethyl at 30 g a.i. ha<sup>-1</sup> at 3 DAT + hand weeding at 30 DAT (M<sub>2</sub>S<sub>5</sub>) resulted in the highest return (rupee invested), equal to 2.89 and 2.37 in the Kuruvai and Navarai seasons, respectively.

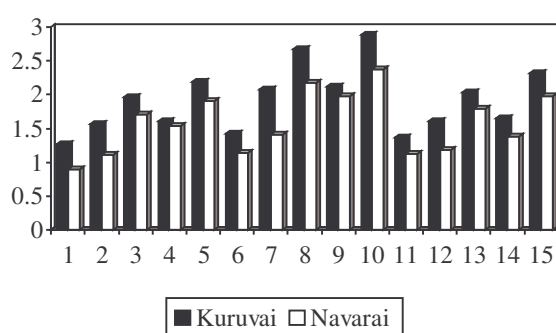


Fig. 1. Return per rupee invested in 'Kuruvai' and 'Navarai' seasons. 1 - M<sub>1</sub>S<sub>1</sub>; 2 - M<sub>1</sub>S<sub>2</sub>; 3 - M<sub>1</sub>S<sub>3</sub>; 4 - M<sub>1</sub>S<sub>4</sub>; 5 - M<sub>1</sub>S<sub>5</sub>; 6 - M<sub>2</sub>S<sub>1</sub>; 7 - M<sub>2</sub>S<sub>2</sub>; 8 - M<sub>2</sub>S<sub>3</sub>; 9 - M<sub>2</sub>S<sub>4</sub>; 10 - M<sub>2</sub>S<sub>5</sub>; 11 - M<sub>3</sub>S<sub>1</sub>; 12 - M<sub>3</sub>S<sub>2</sub>; 13 - M<sub>3</sub>S<sub>3</sub>; 14 - M<sub>3</sub>S<sub>4</sub>; 15 - M<sub>3</sub>S<sub>5</sub>.

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## Integrated weed management of *Ambrosia artemisiifolia* L.

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### Introduction

On the basis of the Fourth National Weed Survey in wheat and maize crops in Hungary (1996-1997 years), common ragweed (*Ambrosia artemisiifolia*) seems to be the most important weed species, with an average weed cover of 4.7% (Tóth, 2003). The weed has a considerable competitive ability, e.g. it caused 38% yield losses in white lupine (*Lupinus albus* L.) at a density of 18 plants m<sup>-2</sup> (Béres, 1985). In a study to assess *A. artemisiifolia* competition in maize, one plant m<sup>-2</sup> of *A. artemisiifolia* reduced the yield by 0.24 t ha<sup>-1</sup> (Varga, 2002). A triazine resistant biotype of *A. artemisiifolia* has also been found in Hungary (Tóth & Hartmann 1995). The plant or its pollen cause dermatitis and allergy in some people, and the importance of such diseases increase year after year. *A. artemisiifolia* is also known as an allelopathic weed. Its water extracts influence the germination of higher plants (Béres *et al.* 2002) and always decrease the propagation of some phytopathogen fungi (Brückner, 2001). *A. artemisiifolia* seeds germinate mainly from 2.6 to- 3 cm soil depth. As much as 20% of the seeds from the upper 20 cm soil layer germinate in a year or lose their viability; this, coupled with prevention of seed formation and ripening by soil cultivation may considerably reduce the number of viable *A. artemisiifolia* seeds in the soil seed bank already after 5 years (Béres, 1994). *A. artemisiifolia* shows high sensitivity to more soil and leaf herbicides. A great choice of effective and selective herbicides are available for its control in field crops.

The aim of our investigations was to study the effect of some post-emergence herbicides and mowings against *A. artemisiifolia*.

### Materials and methods

*A. artemisiifolia* seeds were sown at 2 cm soil depth in plastic pots (100 seeds pot<sup>-1</sup>) under glasshouse conditions in the spring of 2004. At 2-4 leaves stages post-emergence treatments were applied in four replications, using 500 l ha<sup>-1</sup> water. The control treatments applied are listed in Tab. 1.

Tab. 1. Herbicide treatments investigated for *A. artemisiifolia* control.

Commercial name	Active ingredient	Dose (g ha <sup>-1</sup> )
Athos	75% sulfosulfuron	20
Chikara 25 WG	25% flazasulfuron	200
Glean 75 DF	75% chlorsulfuron	15
Granstar 75 DF	75% tribenuron-methyl	20
Merlin WG	75% izoxaflutol	120
Pledge 50 WP	50% flumioxazin	80
Refine 75 DF	75% tifensulfuron-methyl	15

In another experiment, carried out in the same spring, seeds of *A. artemisiifolia* were sown in small plots (60 seeds m<sup>-2</sup>) under field conditions. Hand-mowing was made at 3 cm above the soil surface when *A. artemisiifolia* height was 12, 46 to 55 and 65 to 72 cm, respectively (on 12 May, 21 June, and 12 July).



## Results and discussion

*A. artemisiifolia* showed high sensitivity to applied sulfonylurea, phthalimide and phenylketone herbicides. All of the examined herbicides gave excellent (97 to 100%) post-emergence weed control effect. In Hungary, these herbicides are allowed for use in field crops and in grapevine. Therefore, selective and effective herbicides are available in these crops against *A. artemisiifolia*.

The time and frequency of mowings influenced greatly the vegetative development and the number of male flowers and seed production of *A. artemisiifolia*. Mowing three times gave the best result, reducing the number of male flowers by more than 90%, compared to the control. No reduction in pollen production was observed when mowing occurred on 12 May. If only one mowing is possible, the best time for doing it is just before flowering, i.e. at the beginning of July. Reduction in male flowers was 49.5 and 87.7%, with mowings on 21 June and 12 July, respectively. Mechanical control should be preferred and considered a viable alternative to herbicide application for *A. artemisiifolia*, especially on waste and uncultivated lands, conservation and inner-city areas.

## Acknowledgements

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# Weed control in the public area: combining environmental and economical targets

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## Introduction

An actor-participative project on sustainable weed control on pavements was started in 2000 in the Netherlands. The aim of the project was to develop a new concept of weed management that provides cost-effective and environmental sound weed control (Kortenhoff et al., 2001; Kempenaar et al., in prep.; [www.dob-verhardingen.nl](http://www.dob-verhardingen.nl)). Participants in the project are a water purification board (ZHEW), the Dutch association of drinking water producers (VEWIN), the producer of the mostly-used pavements herbicide today (Roundup Evolution<sup>®</sup>, Monsanto), Wageningen UR (PRI and Alterra), municipalities and weed control contractors. They all have a mutual interest in reducing emission (surface runoff) of glyphosate. Figure 1 shows results of a Life Cycle Assessment (LCA) study on the environmental effects of different weed control systems for pavements (Saft & Staats, 2002). The LCA assumed a 50 % runoff factor for glyphosate, resulting in relatively large effects on aquatic and sediment ecotoxicity.

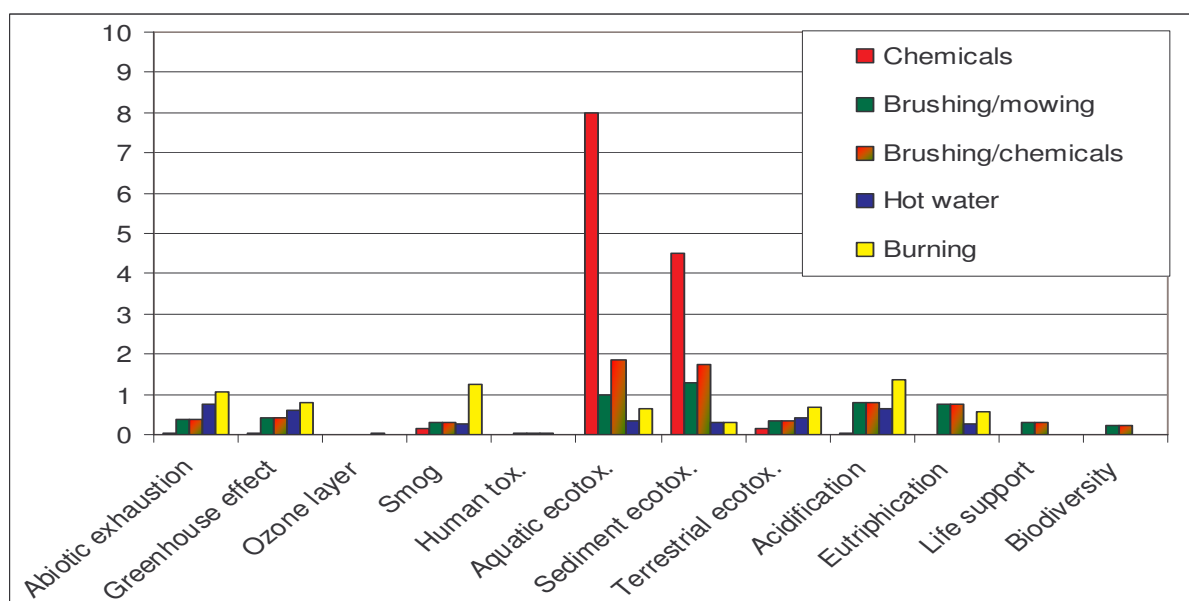


Figure 1. Relative impact scores on environmental themes of five weed control systems. The vertical axis is a relative scale allowing comparison and ranking of effects of different system.

## Materials and methods

Practical weed control guidelines were listed early 2002 to support decision making by managers of pavements and contractors of weed control. Details on the guidelines, the studies and results are given on [www.dob-verhardingen.nl](http://www.dob-verhardingen.nl) (also Kempenaar et al, in prep.). The guidelines are mainly focused on reduction of herbicide use and emission, with main objective to reduce surface runoff of glyphosate. For example, the concept forbids herbicide use on places and moments sensitive to herbicide runoff. In addition, the concept stimulates professional organization, weed prevention and registration of management activities. The guidelines and the new concept were tested in three seasons (2002 - 2004) in residential quarters (5 – 25 ha) in nine Dutch municipalities. A large emission monitoring program was carried out, including flow rate proportional sampling. Both sewage water and surface water was assessed on herbicide residues. Efficacy and costs of weed control were also determined.

## Results and discussion

Glyphosate use in the test quarters was circa 400 gram a.i. per ha per time weed control was done (generally two times per season per quarter). The emission monitoring program yielded a large amount of data (see reports on [www.dob-verhardigen.nl](http://www.dob-verhardigen.nl)). Glyphosate concentrations were higher in the sewage water than in the surface water. Surface water concentrations were often (>80 % of the cases) below the detection limit, and on average circa a factor 100 less than the ecological threshold concentration of 77 µg per liter. Flow rate proportional sampling data were used to estimate emission factors (which is the amount of glyphosate emitted from the quarter divided by the amount of glyphosate used in the quarter). Estimated emission factors for glyphosate are shown in Figure 2 (on average: 2 %). The longer the period between application and rain, the smaller the emission factor. If observed emission factors are projected on Figure 1, the ecotoxicity bars for chemicals are circa 90 % lower.

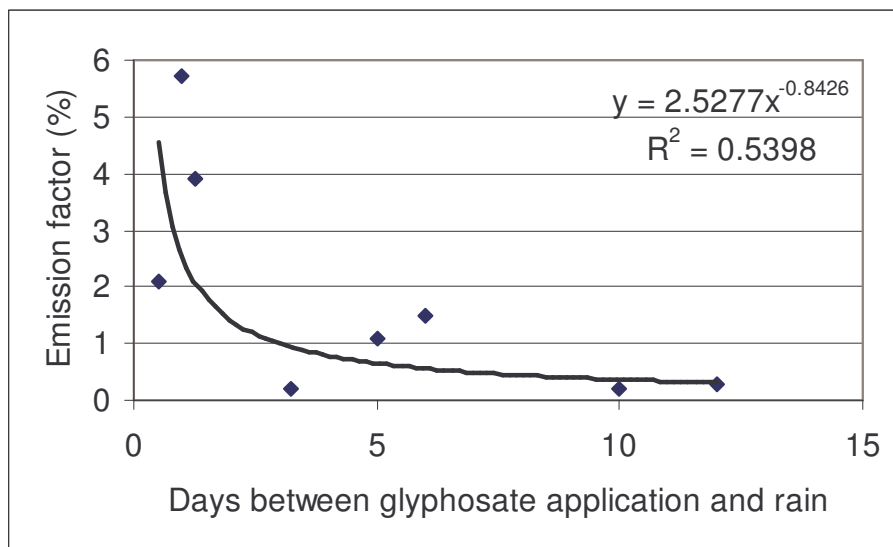


Figure 2. Emission factors via the sewage water system in 2002 and 2003.

Efficacy of weed control was good in the test quarters. The level of weed development did not exceed pre-set maximum levels. Costs of weed control by the new concept was calculated and were a little higher than standard practice chemical weed control, but much lower compared with non-chemical weed control systems. Costs ranged from 0.05 to 0.15 € per m<sup>2</sup> per year. Overall, it was concluded that the new concept allowed meeting both economical and ecological objectives related to weed management of pavements. The new concept reduced glyphosate runoff in a way that the ecological thresholds were not exceeded by far. A specific guideline is included to protect surface waters that have to be safeguarded from pesticides according to the new EU water framework directive (drinking water criterion 0.1 µg a.i. per liter). Broad dissemination of the new concept is planned, and will be supported by the EU Life program (SWEEP project, <http://europa.eu.int/comm/environment/life/home.htm>).

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# The influence of tillage system, fertilization and crop protection level on spring barley weed infestation on a light-textured soil

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## Introduction

Higher number of weed species along with lower plant density has been found under ploughing than under non inversion tillage (Jędruszczak *et al.* 1997; Kraska & Pałys, 2004). Mineral fertilization and chemical crop protection has been found to limit weed infestation in a spring barley crop (Deryło, 2004).

The present work was conducted to determine the impact of ploughing and non inversion soil tillage systems, under two fertilization and two chemical crop protection levels, on weed density, species composition and biomass in spring barley cultivated on a light-textured soil.

## Materials and methods

The investigations were run in the period 1998-2000. A two-factor field experiment was set up within a randomized block design with four replications on loamy sand.

Spring barley was cultivated in a potato-spring barley-winter rye rotation. The experimental plan included two factors: I. tillage systems – 1) conventional tillage (autumn ploughing at 25 cm) and 2) reduced tillage (chisel ploughing at 30 cm); II. two crop intensification levels, given by the combination of mineral fertilization and crop chemical protection: a) basic – 150 kg ha<sup>-1</sup> NPK, herbicide application at the tillering stage (417.5 g a. i. ha<sup>-1</sup> of 2,4-D and 32.5 g a. i. ha<sup>-1</sup> of dicamba); b) intensive – 310 kg ha<sup>-1</sup> NPKMg, herbicide application at the tillering stage – 417.5 g a. i. ha<sup>-1</sup> of 2,4-D and 32.5 g a. i. ha<sup>-1</sup> of dicamba + 69 g a. i. ha<sup>-1</sup> fenoxaprop-P-ethyl and 75 g a. i. ha<sup>-1</sup> mefenpyr-dietyl (safener); fungicide application at the late tillering stage – 125 g a. i. ha<sup>-1</sup> flusilazol and 250 g a. i. ha<sup>-1</sup> carbendazym, at the heading stage – 375 g a. i. ha<sup>-1</sup> tridemorph and 125 g a. i. ha<sup>-1</sup> epoxiconazol; growth regulator – at the shooting stage – 610 g a. i. ha<sup>-1</sup> chlormequat chloride and 310 g a. i. ha<sup>-1</sup> ethephon.

P, K and Mg were applied at seedbed preparation. Nitrogen was applied in three times, the first before sowing (30 kg ha<sup>-1</sup> in the basic level and 50 kg ha<sup>-1</sup> in the intensive level), the second (30 kg ha<sup>-1</sup>) at the tillering stage and the third (20 kg ha<sup>-1</sup>, only in the intensive level) at the heading stage. Spring barley cv. Start was sown at 400 seeds m<sup>-2</sup> at an inter-row distance of 12 cm. Prior to spring barley harvest the assessment of canopy infestation was made with quantitative-weighting method. Weed density was counted by species and weed biomass was sampled in two randomly located sampling areas of 1 x 0.5 m in each plot. Data were statistically analysed by ANOVA, and the mean values were compared with a Tukey test at P < 0.05).

## Results and discussion

Contrary to crop intensification level, soil tillage did not affect weed density of spring barley (Tab. 1), while weed biomass was unaffected by both experimental factors. Pałys *et al.* (1997) obtained similar results on a rendzina soil. Intensive level of fertilization and chemical crop protection significantly decreased density of monocots and total density of weeds (Tab. 1). In conventional tillage, 19 dicot and 5 monocot species were recorded, *Echinochloa crus-galli* being the most abundant one. High abundance of *E. crus-galli* in spring barley was also found by Pałys *et al.* (1997). Under reduced tillage, 25 weed species (20 dicots and 5 monocots) were detected. The higher number of dicots (26) was found in the basic crop intensification

level. *E. crus-galli* was the prevailing monocot also under reduced tillage. A lower number of species (23) was detected in the chemically-intensive treatment. Under conventional tillage, density of *Geranium pusillum*, *Galinsoga parviflora*, *Stellaria media*, *Chenopodium album* and *Poa annua* decreased while that of *Chamomilla suaveolens* increased. Bujak (1996) demonstrated that in spring barley less intensive autumn cultivation increased especially the density of *Chenopodium album*. Intensive fertilization and crop protection levels reduced the occurrence of *Echinochloa crus-galli*, *Poa annua*, *Galinsoga parviflora* and *Chenopodium album* but increased that of *Chamomilla suaveolens* (Tab. 1).

The results of the present study confirm that weed infestation in spring barley do not depend on soil tillage. In fact, higher levels of mineral fertilization and crop protection limited weed density under both conventional and reduced tillage.

Table 1. Weed density (plants m<sup>-2</sup>) before harvest in spring barley as influenced by tillage system and level of fertilization and crop chemical protection (means of the period 1998-2000).

Species	Tillage system		Level of fertilization and crop protection	
	Conventional	Reduced	Basic	Intensive
<b>Dicotyledonous</b>				
<i>Chamomilla suaveolens</i> (Pursh) Rydb.	3.1	0.2	0.2	3.1
<i>Geranium pusillum</i> Burm. F. Ex L.	2.0	4.2	4.0	2.2
<i>Galinsoga parviflora</i> Cav.	0.8	5.5	5.0	1.2
<i>Stellaria media</i> (L.) Vill.	0.7	2.8	2.1	1.4
<i>Chenopodium album</i> L.	0.1	4.7	4.3	0.5
Other species	9.1	10.4	10.1	9.5
Total dicotyledonous	15.8	27.8	25.7	17.9
Number of dicotyledonous species	19	20	21	19
<b>Monocotyledonous</b>				
<i>Echinochloa crus-galli</i> (L.) P. Beauv.	55.7	55.8	105.3	6.2
<i>Poa annua</i> L.	2.7	13.3	11.3	4.7
Other species	0.6	1.9	1.7	0.8
Total monocotyledonous*	59.0	71.0	118.3	11.7
Number of monocotyledonous species	5	5	5	4
Total number of weeds**	74.8	98.8	144.0	29.6
Number of species	24	25	26	23

\*, \*\*significant at P < 0.05 and 0.01 levels respectively. LSD (P < 0.05) for crop intensification level: 40.4 and 42.3 respectively.

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# Cover crops in organic farming systems: exploring and optimizing their contribution to ecological weed management

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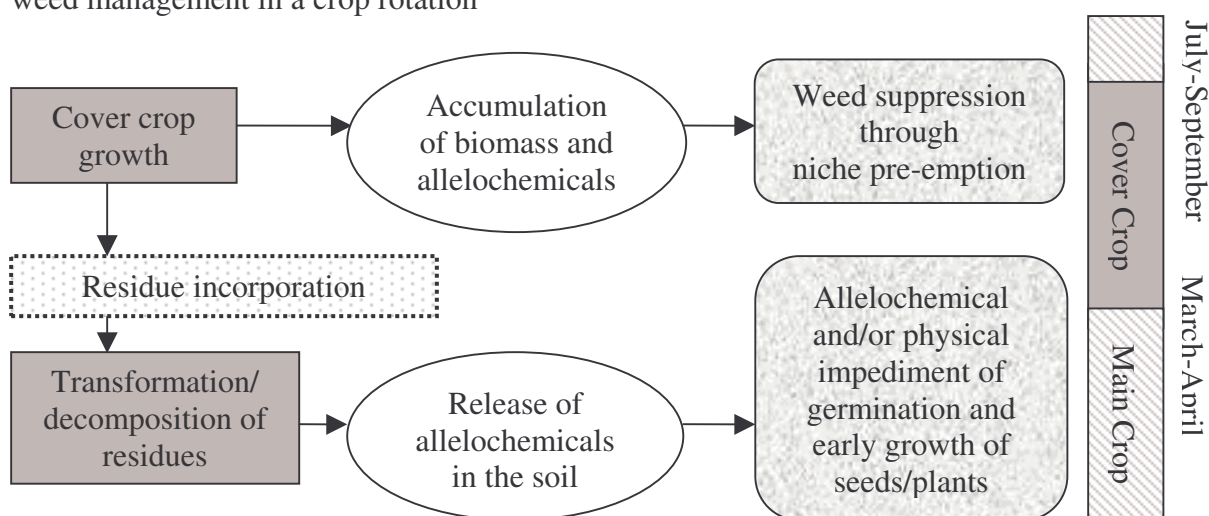
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## Introduction

Cover crops have potential to form an important part of a systems-oriented approach to weed management. One strategy for using cover crops is to grow them during the period when the main crop is absent. Inclusion of cover crops in crop rotations introduces two important mechanisms through which the development of weed populations might be hampered. In late summer and autumn the successful introduction of cover crops prevents growth, development and, most importantly, seed production of weeds that remain in the stubble. In springtime cover crop residues, either incorporated in the upper layer of the soil or left as a mulch on the soil, might suppress or retard weed emergence and growth due to allelopathic and/or physical effects (Fig. 1). In 2003, a 4-year research project was started with the aim to explore the potential of cover crops to contribute to the ecological management of weed populations in organic farming systems.

Fig. 1. Framework showing the mechanisms through which cover crops might contribute to weed management in a crop rotation



## Cover crop growth

Weed suppressive ability of cover crops in autumn is studied in field experiments. In the first year of experimentation six cover crop species were evaluated. Their weed suppressive ability was assessed through determination of the growth of natural weeds and the growth of a model weed (*Vicia sativa* L.). Different morpho-physiological characteristics of the cover crop species were measured in order to elucidate differences in weed suppressive ability. For example, we found that for all cover crop species, except for lucerne, a faster soil coverage coincided with a lower dry weight per *V. sativa* plant (Tab. 1). In the second year of experimentation the field experiment was repeated with the winterhard species (winter rye, winter oilseed rape and lucerne) sown at three different densities.

In order to effectively reduce weed emergence and growth in spring it is important to optimize the amount of allelochemicals in the cover crop material, which depends both on the biomass and on the concentration of allelochemicals in the cover crop at the time of residue



incorporation. The concentration of allelochemicals in the cover crop is genetically determined, varies with development stage/age (e.g. Burgos *et al.*, 1999), and is influenced by abiotic factors, such as light, nutrient and water availability (e.g. Mwaja *et al.*, 1995) and biotic factors, such as herbivore/microbial attack and competition (e.g. Siemens *et al.*, 2002). Whole plants from different cover crop species, different developmental stages and different cover crop densities by nutrient levels are harvested from the field and subsequently freeze-dried and ground. This material will be tested for allelopathic potential by chemical analysis and by means of bioassays with lettuce (*Lactuca sativa* L.) as a test species. Furthermore the influence of mechanical damage on the concentration of allelochemicals in winter rye and winter oilseed rape will be assessed.

Tab. 1. Time to reach 50% soil cover ( $t_{50\%}$ , das) and per plant dry weight of the model weed *V. sativa* (W; g plant<sup>-1</sup>) in the 2003 field experiment (Wcontrol = 6.013 g plant<sup>-1</sup>).

	Winterhard	$t_{50\%}$	W	Not winterhard	$t_{50\%}$	W
<i>Poaceae</i>	Winter rye	33	0.795	Italian ryegrass	50	2.539
<i>Fabaceae</i>	Lucerne	46	4.121	White lupin	53	2.322
<i>Brassicaceae</i>	Winter oilseed rape	28	0.222	Fodder radish	27	0.269

### Residue management

In the Netherlands, cover crop residues are commonly incorporated in the soil by ploughing. However, when aiming at weed suppression, other ways of residue management are likely to be more effective. The inhibition of weed germination and growth by the cover crop residues depends on the contact of the allelochemicals with the weed and crop seeds. This in turn depends on the distribution of the residues in the soil and on the nature of allelochemicals, i.e. whether they are water-soluble or volatile. The way cover crop residues are pre-treated (e.g. cutting, crushing) might influence the release rate of the allelochemicals and therefore their effect on weed and crop seeds. Residue management of two different cover crop species, winter oilseed rape (volatile and non-volatile allelochemicals; Brown & Morra, 1995) and winter rye (merely non-volatile allelochemicals; Barnes *et al.*, 1986), will be compared in an experiment.

### Transformation/decomposition of residues

The uptake of allelochemicals by the seed(ling)s depends on the presence of the allelochemicals in the soil, which in turn depends on three different processes; 1) the release rate of allelochemicals from the residues, 2) the transformation/degradation of the allelochemicals and 3) the movement of allelochemicals out of the soil. By repeated introduction of lettuce seeds, the presence of allelochemicals in the soil in the period following residue incorporation of winter rye, winter oilseed rape and lucerne will be monitored. To take advantage of allelochemical effects of cover crop residues for weed suppression in agro-ecosystems, we must target weed species and avoid negative effects on the crop. Therefore the effect of the allelochemicals released by the residues of the above-mentioned species will be assessed for different crop and weed species.

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## Management of grass-weeds in overwintering stubbles

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### Introduction

The over-wintering stubble option of agri-environment schemes requires farmers to maintain their stubbles over winter, with minimal management interventions, as a feeding site for birds. However, it is widely documented that reduction in cultivation operations increase the potential for grass-weed build up, particularly on heavy soils. Hence, there is a danger that leaving an over-wintering stubble could amplify the level of grass-weed infestation in subsequent crops.

The grasses chosen for this research are all serious weeds of autumn-sown cereals, causing yield loss and/or are of increasing prominence in the UK. As recurrent herbicide applications are used to combat these weeds, the risk of herbicide resistance is increased, particularly in the case of *Alopecurus myosuroides* and *Lolium multiflorum*. Therefore, an objective of the research reported here was to investigate natural depletion of grass-weed seeds in over-wintering stubbles subjected to straw residue management or disc cultivation, without the use of herbicides.

### Materials and Methods

Straw management trials were conducted at Morley Research Centre, Norfolk between August 2003 and February 2004 in two stubble fields of winter wheat on either heavy or light soil. Seeds of *A. myosuroides*, *Anisantha sterilis*, *Bromus commutatus* and *L. multiflorum* were broadcast on the soil surface of 2 m<sup>2</sup> plots at a rate of either 500 or 1000 seeds m<sup>-2</sup>. Half the plots were left bare whilst the other half were covered with straw and chaff at a rate equivalent to 10 tonnes ha<sup>-1</sup>. Seedling emergence was monitored regularly until the plots were harvested in mid February. Micro-meteorological measurements were taken.

A second trial was conducted at The University of Reading Experimental Farm, Sonning, Berkshire from October 2003 to January 2004 in barley stubble. Two m<sup>2</sup> plots were surface sown with seeds of contrasting size (*A. sterilis* and *A. myosuroides*) at 500 seeds m<sup>-2</sup>. Plots were left undisturbed or subjected to a single or double pass with medium disc harrows on 10 October 2004 to a depth of 5 cm. Seedling emergence was recorded at regular intervals.

### Results

Although not always statistically significant, where straw was retained on the soil surface, the results showed a trend for depletion of *A. sterilis* and *B. commutatus* through seed germination. Conversely, there was a tendency for greater diminution of *A. myosuroides* and *L. multiflorum* seed by germination from an exposed soil surface (Tab. 1). The microclimatic effect of a mulch simulates a layer of soil, excluding light and reducing extremes of temperature and humidity. Germination of *A. sterilis* is promoted by darkness (Hilton, 1982). Although germination of *B. commutatus* is not photo-inhibited (Chancellor & Froud-Williams, 1986), it is strongly influenced by water supply (Mortimer *et al.*, 1993), which maybe enhanced under a mulch. In contrast, for many small-seeded species, (*A. myosuroides* and *L. multiflorum*), light serves as a germination stimulus (Andersson *et al.*, 2002). These species may also be sensitive to the allelochemicals produced by straw residues (Kati & Froud-Williams, 1999).

Soil disturbance by disc cultivation stimulated emergence of *A. sterilis* although there was no significant difference between one or two disc passes. In contrast, whilst discing initially promoted emergence of *A. myosuroides*, over time there was no significant difference between the treatments (Fig. 1).

Tab. 1. Seedling establishment in the presence/absence of a straw mulch (arc sin transformed mean percentage of seeds sown. LSD values compare straw\*density interactions).

Heavy soil	500 seeds m <sup>-2</sup>		1000 seeds m <sup>-2</sup>		LSD
Species	+ straw mulch	- straw mulch	+ straw mulch	- straw mulch	
<i>A. sterilis</i>	40.8	29.1	39.1	36.9	4.95
<i>B. commutatus</i>	41.4	31.4	31.7	35.2	16.52
<i>A. myosuroides</i>	9.14	12.01	2.5	8.1	2.87
<i>L. multiflorum</i>	n/a	n/a	n/a	n/a	
Light soil	500 seeds m <sup>-2</sup>		1000 seeds m <sup>-2</sup>		LSD
Species	+ straw mulch	- straw mulch	+ straw mulch	- straw mulch	
<i>A. sterilis</i>	51.9	44.1	48.7	43.3	1.28
<i>B. commutatus</i>	40.0	31.6	n/a	n/a	6.71
<i>A. myosuroides</i>	17.8	19.0	14.8	18.3	3.87
<i>L. multiflorum</i>	30.6	33.7	29.9	34.7	3.06

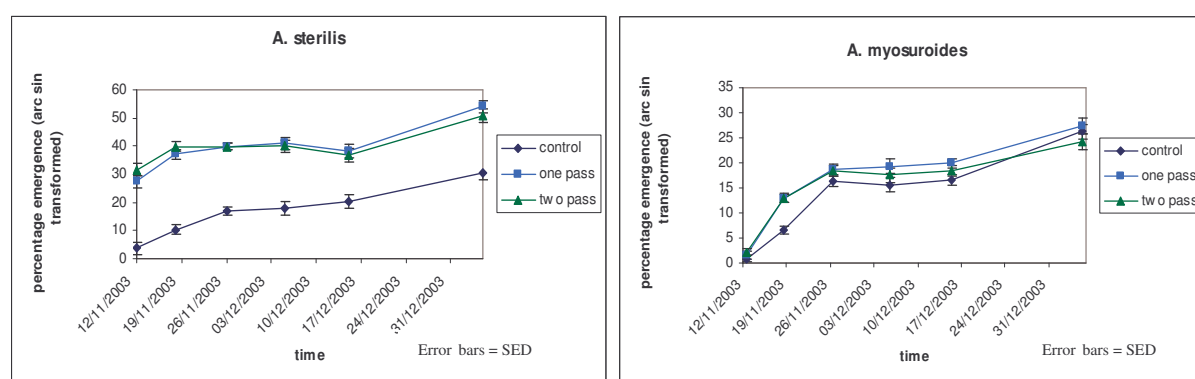


Fig. 1. Effects of disc cultivation on *A. myosuroides* and *A. sterilis* seedling emergence.

It is likely that covering *A. sterilis* seed with soil by discing promoted its germination through light exclusion. Due to the exceptionally dry autumn, the increase in seed/soil contact may also have been important in provision of sufficient moisture for germination. The same is true for *A. myosuroides*; however this species showed very little germination overall, which may account for lack of significant difference between the treatments. At the onset of the trial, low moisture availability and direct exposure to wetting/drying cycles through dew formation in the control treatment may have inhibited germination compared to the disced treatments, overriding the influence of light.

This research demonstrated that some grass-weed seeds can be encouraged to germinate during over-wintering stubbles with simple management practices. As a result, besides seedbank depletion, there will be an opportunity for seedling destruction before seed shedding in the spring. However, employing these management techniques alone will likely be inadequate to sufficiently prevent seedbank replenishment; furthermore, as individual species show differential responses to management, any management intervention must be targeted to the particular field and its associated weed flora.

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# Competitiveness of erect, semi-erect, and prostrate cowpea (*Vigna unguiculata*) genotypes with sunflower and purslane

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## Introduction

The growth habits of the crop and competing weed species are important determinants of crop-weed competition. To compete with weeds, a spreading growth habit that rapidly forms a canopy and covers bare soil to shade weeds is thought to be ideal for a cover crop. However, erect crop varieties have a height advantage that might be more effective in suppressing weeds and maintaining yield in the face of weed competition. In addition, erect varieties (for example, erect cowpea varieties) can be mechanically cultivated later into the season, allowing growers to achieve a better overall weed control. Identifying competitive cowpea varieties and specific traits related to competitive ability would provide breeders with useful traits and information that could be used to develop a crop ideotype for highly competitive cowpea cover crop varieties. We compared the relative competitiveness of three cowpea genotypes with prostrate, erect, or semi-erect growth habits and identified the characteristics that contribute to differences in ability to compete with purslane or sunflower.

## Materials and methods

Three cowpea [*Vigna unguiculata* (L.) Walp.] genotypes with similar vegetative vigor but different growth habit were assessed for their relative competitiveness with two weed species. 'Iron-Clay' (IC) grows erect, 'IT89KD-288' (288) is semi-erect, and 'UCR 779' (779) is prostrate. Common purslane (*Portulaca oleracea* L.), a short statured weed, and common sunflower (*Helianthus annuus* L.), a tall species, were planted within the cowpea rows.

Cowpea canopy height, canopy width, and light intensity above and below sunflower and cowpea canopies were measured weekly from 21 days after planting until final biomass harvest. One meter of row (0.76 m<sup>2</sup>) was harvested at each sampling date by cutting plants at ground level. The numbers of cowpea and weed plants were counted, leaves were separated from stems, and the leaf area of fresh green leaves was measured with an optical leaf area meter. Leaves and stems were dried at 70°C with ventilation until a constant weight was reached.

## Results and discussion

Sunflower reduced the leaf area, amount of light received, and biomass of all cowpea genotypes. Cowpea reduced the amount of light received by purslane, purslane grown with IC and 779 received less light than purslane grown with 288 (Fig. 1). Purslane reduced the leaf area of 779 and the biomass of 288 and 779, but the biomass and leaf area of IC was not affected. The presence of sunflower increased the height of IC and 288, but the presence of purslane decreased the canopy height of 779. IC reduced sunflower biomass, while IC and 779 reduced purslane biomass. IC and 288 reduced sunflower leaf area, while IC and 779 reduced purslane leaf area. The growth analysis of biomass, leaf area, and canopy height of cowpeas and weeds showed similar results. IC was the most competitive genotype, suggesting that an erect growth habit may be more effective in suppressing weeds than semi-erect or prostrate growth habits. Results were used to develop a simulation model based on INTERCOM to help develop competitive crop varieties.

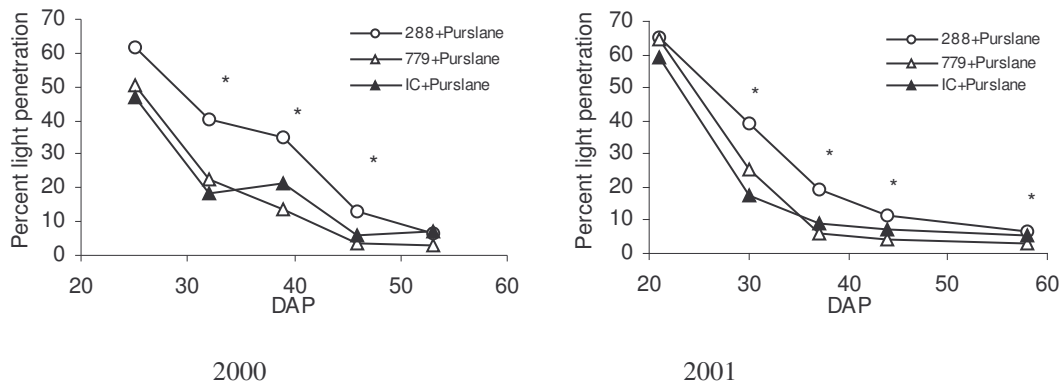


Fig. 1. The effect of cowpea genotypes on the percent light reaching purslane at various days after planting (DAP) in 2000 (top) and 2001 (bottom). Asterisks indicate differences in the percent light reaching purslane when grown with cowpeas, as determined by Fisher's Protected LSD at the 5% level.

# Integration of *Elymus repens* control and post-harvest catch crop growing in organic cropping systems

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## Introduction

*Elymus repens* (L.) Gould constitutes a major perennial weed problem in organic cropping systems in many parts of Denmark with great negative impact on crop yield and quality. *E. repens* infestations are traditionally controlled by repeated stubble cultivation in the post-harvest period from harvest to ploughing, either early autumn before sowing a winter-sown crop or in late autumn. However, in organic farming, post-harvest tillage is undesirable due to the need for retaining nutrients, particularly nitrogen, in the cropping system. Thus, the soil is often cropped with catch crops, autumn-sown crops or perennial crops in that period, limiting opportunities for post-harvest tillage.

In this paper, a new control strategy (strategy I) against *E. repens* is presented that merges the objectives of achieving a significant reduction of *E. repens* while having the soil covered with plants during most of the post-harvest period. Strategy I contains an integration of rhizome fragmentation by soil cultivation within one or two days after harvest in early August with subsequent catch crop growing in late summer and autumn to suppress shoot growth from the weakened rhizome fragments. Strategy I is discussed in relation to another strategy (Strategy II) that also includes catch crop growing in late summer and autumn but is preceded by a mid-summer fallow period lasting 4-6 weeks where repeated soil cultivations are conducted to fragment, weaken and desiccate the rhizomes.

## Materials and methods

*Strategy I*: Two field experiments were established on a coarse sandy soil just after harvest of spring barley in August, (a) in 2002 and (b) in 2003. Both experiments contained three factors. Factor 1: mechanical disintegration of rhizomes, 5 levels: 1. untreated, 2. disintegration and loosening by mouldboard ploughing to only 10 cm soil depth, 3. strong disintegration by PTO-driven rotary cultivation, 4. disintegration and loosening by stubble cultivation, 5. loosening and uprooting by a newly developed Danish implement, the “Kvik-Up”, based on a cutting and loosening tool element followed by a rotating tool element for uprooting. Factor 2: catch crop growing, 3 levels: 1. no catch crop, 2. mixture of fodder radish (*Raphanus sativus* L.) and westerwolds ryegrass (*Lolium multiflorum* Lam. var. *westerwoldicum* Mansh.), 3. mixture of red clover (*Trifolium pratense* L.) and winter vetch (*Vicia villosa* Roth). Factor 3: timing of mouldboard ploughing, 2 levels: 1. late autumn, 2. spring. Factor 1 was conducted within two days after harvest and straw removal, and the catch crops were sown subsequently so that both rhizome disintegration and catch crop establishment were accomplished no later than a week after harvest. Spring barley was sown in the following spring and the overall effects on *E. repens* including barley yield were assessed at harvest.

*Strategy II* was conducted by the Danish Agricultural Advisory Service, National Centre, Crop Production ([www.lr.dk](http://www.lr.dk)). The mid-summer fallow period was started around first of July by shallow mouldboard ploughing to 10 cm soil depth following the harvest of a whole crop for silage. Then tine cultivation once a week was conducted till early August where the fallow period was ended by mouldboard ploughing to 20 cm soil depth. A competitive catch crop (mixture of red clover, fodder radish, winter rye (*Secale cereale* L.) and winter vetch) was established to suppress *E. repens* shoot growth and to take up nutrients during autumn. The



effect was assessed the following year in spring barley. Results from a total of 5 experiments on fine or coarse sandy soils are reported in this paper.

## Results and discussion

Rotary cultivation in strategy I caused the highest level of rhizome disintegration with an average rhizome length of 11.7 cm as compared with 30.7 cm on average for all the other treatments that did not differ significantly. Catch crop growth during the autumn of 2002 and 2003 suffered from very dry weather conditions resulting in poor establishment and canopy development. The radish/ryegrass mixture significantly suppressed weed growth in Exp. (a) but none of the catch crops affected weed growth in Exp. (b). Mechanical disintegration reduced growth of *E. repens* significantly in Exp. (a). Rotary cultivation gave the highest growth reduction compared with untreated (Fig. 1A), but compared with the infestation level prior to starting the strategy, the reduction was only roughly 40%. Catch crop growing affected the crop's ability to compete with *E. repens* in the subsequent year as shown in Fig. 1B, where barley grain yield is related to the amount of *E. repens* biomass that followed the treatments (1-5) under factor 1. The two regression lines have significantly different slopes ( $P < 0.05$ ), which show that the radish/ryegrass mixture had strengthened crop growth more than no catch crop and the clover/vetch mixture. Timing of ploughing had generally no effect and none of the three factors affected *E. repens* in Exp. (b) because of wet and cold weather conditions during most of the summer of 2004, presumably promoting *E. repens* growth to an extent where the effects of previous year's treatments were eliminated.

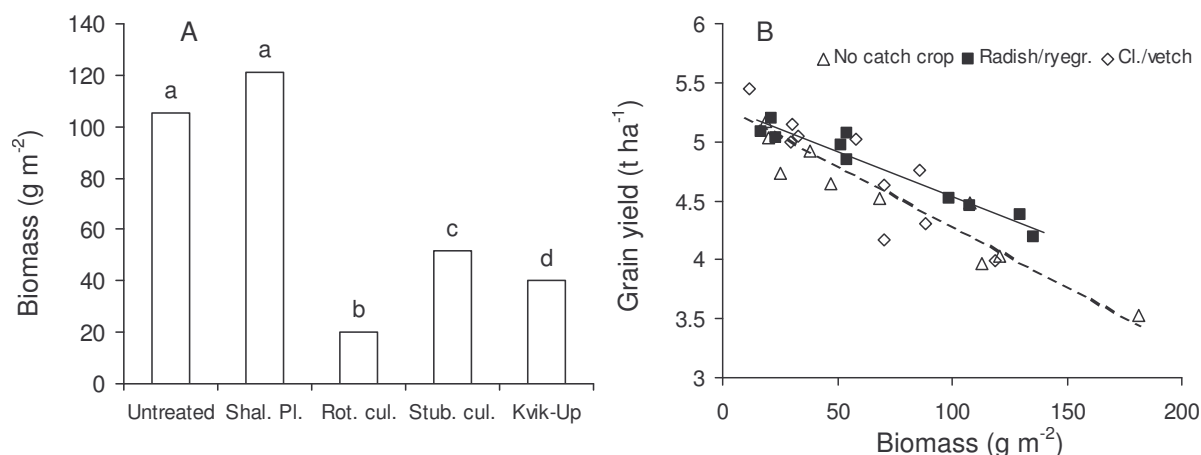


Figure 1. A: Aboveground *Elymus repens* biomass assessed close to barley harvest following previous year's mechanical treatments in Exp. (a) (data averaging factor 2 and 3), columns with different letters are significantly different ( $P < 0.05$ ). B: Regression analyses of barley yield related to *E. repens* biomass in the year after conducting treatments 1-5 under factor 1 in Exp. (a). (---) is no catch crop and clover/vetch, and (—) is radish/ryegrass.

Strategy II gave very high and consistent reductions of *E. repens* infestations, leading to 91-99 % efficacy compared with the infestation level prior to starting the strategy. A stronger weakening and desiccation of rhizomes took place during the mid-summer fallow period in strategy II than in the short-term mechanical treatment in strategy I. However, strategy II has been tested in long-term organic crop rotation experiments where high effects attained in the first year tended to decline rapidly in a few years' time. Also the rather long mid-summer fallow period may lead to some nutrient loss, (but still lower than with traditional *E. repens* control), through leaching after heavy rainfall events, which subsequently may give rise to crops that are less competitive against *E. repens*. In addition, the grower will have to desist from growing a full-season cash crop. These aspects should be counterbalanced against the urgency for *E. repens* control and other possible control options.

## Mechanical weeding in potato

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### Introduction

Efficacy of new weeding strategies using fewer herbicides than the standard practices was tested by Agro-Transfert between 1998 and 2003 in potatoes. The experiments were aimed at estimating the effectiveness of mechanical weeding in order to provide new recommendations for farmers (Van Loon, 1993). In the Picardie region of France, there is a lack of references, and recommendations are confined to pre-emergence herbicide application. Mechanical weeding is only used by organic farmers.

\* Agro-Transfert is a non for profit organization with a partnership between Chambre Régionale d'Agriculture de Picardie and INRA.

### Materials and methods

Seven trials were carried out between 1998 and 2003 on silty loams. Potato was cropped according to the Picardie standard practices. The first part of the experiments (1998-2001) was aimed at estimating the efficacy of mechanical weeding by hilling. In the second part (2002-2003), combinations of herbicides at low doses and hilling were investigated. The cultivator was the farmer's tool (Hebert), and herbicide treatments were made with a hand-carried sprayer. Every year, the first hilling and the pre-emergence herbicide treatment were usually made around the 15 May two weeks after planting. The other treatments were made by the end of June, according to the weather conditions and before covering the soil with potato foliage. Weed counts were done before defoliation around 20 August. Efficacy was calculated as : % efficacy =  $100 \times (1 - \text{NWT} / \text{NWC})$  (NWT = Number of Weeds in the plots after Treatment, and NWC = Number of Weeds in the Control plots). Table 1 contains information about treatments, weed density in the control plot, efficacy on weeds, and precipitation.

### Results and discussion

Weed density was between 2 and 109 plants m<sup>-2</sup> in the control plots, depending on year and field. Major weeds were *Chenopodium album* L. and *Solanum nigrum* L. except in 2003. They were typical weeds found in potatoes in Picardie. From 1998 to 2001, results showed that herbicides on average were more efficient than 2 passes of hilling, and 2 passes of hilling were more effective than one pass. These results are similar to those achieved in other countries (e.g. Bellinder *et al.*, 2000). Irla (1995) and Kilpatrick (1995) observed that rain lowers the efficacy of mechanical weeding. As shown in Table 1, herbicides were more efficient, if considerable rain had fallen within 6 days after spraying. Wet soil conditions are assumed to promote the root uptake of herbicides. Moreover, our observations showed us that weeds might re-grow after mechanical weeding under wet soil conditions. On the contrary, when a dry period followed the treatment, mechanical weeding by hilling (2 passes) was as effective as herbicides. Results obtained in 2002-03 with combining mechanical weeding and 2 herbicide applications at low doses showed on average that this practice gave the same efficacy as standard recommendations. It seems that split-application of herbicides gives more consistent results, because some rainfall is likely to occur when applying herbicide twice.

Table1. Main results of the trials conducted between 1998 and 2003.

	1998	1999	2000	2001	2002	2003	2003
Weed density in the control plots (plants/m <sup>2</sup> )	2	13	97	82	109	14	6
Main weeds (0)	Am, Fh	Am, Fh	Am, Fh	Am, Fh	Am, Fh	Am, Bh, Bb	Fh, Bb
Efficacy (% control)							
Chemical (1)	80%	100%	98%	100%	96%	79%	97%
Mechanical 1 (2)	80%	78%	82%	98%	68%	88%	98%
Mechanical 2 (3)	9%	69%	88%				
Mechanical and chemical (4)					86%	91%	95%
Efficacy variation (2)-(1)	0%	-20%	-17%	-1.6%	-28%	-9%	+1.8%
Efficacy variation (4)-(1)					-12%	+12%	-3%
Rain fall after spraying (mm water) (5)	2	18.5	11.5	4	22	12	12
Number of days with rain (6)	1	2	5	1	2	4	4
May rainfall (mm water)	9	34	54.5	28	42	52.5	52.5

(0) = Am=*Mercurialis annua* L., Fh = *Chenopodium album* L. Bn= *Solanum nigrum* L., Bb = *Fallopia convolvulus* L.. (1): Hilling followed by prosulfocarb 3600g ha<sup>-1</sup> + metribuzin 210 g ha<sup>-1</sup> (pre emergence) in May = standard practice. (2) Pre emergence hilling in May, and post emergence hilling before covering the soil with potato foliage. (3) No pre-emergence hilling but post emergence hilling before covering the soil with potato foliage. (4) Pre emergence hilling + metribuzin 140 g ha<sup>-1</sup> and post emergence hilling before covering the soil with potato foliage + metribuzin 70 g ha<sup>-1</sup>. (5) precipitation within 6 days after pre emergence spraying. (6) number of days with precipitation within the 6 days period.

The results should be evaluated under a wider range of cropping conditions (soil and climate). Moreover, better weather forecast for a 5-6 day period might save the second herbicide application (see (4) in table 1), if the first application has been made on wet soil resulting in high weed efficacy.

In conclusion, it is possible to reduce the amount of herbicides from 3200 g ha<sup>-1</sup> to 210 g ha<sup>-1</sup>. However, in Picardie there is a shortage of workforce, necessary for one or two extra passes, and an implementation of more labour demanding mechanical weeding in potato has to be analysed in relation to a decreasing work force in French agriculture. This is the aim of a new study of Agro-Transfert dealing with integrated weed management.

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# Mechanical weed control in sunflower

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## Introduction

In row crops, although weeds between the crop rows can normally be controlled by ordinary inter-row cultivation (e.g. hoeing, harrowing), weeds in the crop row constitute a major problem. For intra-row weed control, most mechanical methods are based on old principles but new implements and improved versions have emerged lately (e.g. finger-weeder, torsion-weeder) (Ascard & Bellinder, 1996; Pannacci & Covarelli, 2004). There are, however, limited reports on the effects of these implements. The aim of this study was to evaluate the weed control effect of several mechanical inter- and intra-row weed control methods in sunflower (*Helianthus annuus* L.), one of the major arable row crops in central Italy.

## Material and methods

In 2002 and 2003, two field experiments with sunflower were carried out in central Italy (Tiber valley, Perugia, 165 m a.s.l.) on a clay-loam soil (24.8% sand, 30.4% clay and 0.9% organic C). In both years, soft winter wheat was the preceding crop and the seed bed was prepared by ploughing to 0.4 m soil depth, disc harrowing, rotary harrowing and spike-tooth harrowing. Sunflower (var. Sambro) was sown on 15.04.2002 and 16.04.2003, with an inter-row spacing of 0.5 m. After emergence, sunflower seedlings were thinned to a final density of 5 plants m<sup>-2</sup>. The experimental design was a randomized block with three replicates and a plot size of 45 m<sup>2</sup> (3 m width). Different mechanical weed control methods were compared: 1) hoeing; 2) hoeing-ridging; 3) split-hoeing; 4) finger-weeding; 5) split-hoeing + finger-weeding; 6) pre-emergence herbicides (metobromuron 750 g a.i. ha<sup>-1</sup> + pendimethalin 1250 g a.i. ha<sup>-1</sup>) in band applications on the row (50% of total surface) + inter-row hoeing. A pre-emergence broadcast application (same herbicides as above) and untreated plots were added as checks. Mechanical weed control treatments were applied on 16.05.2002 and 14.05.2003 when the crop had developed 6 true leaves, broadleaved had developed 2 to 8 true leaves and grass weeds from 3 leaves to tillering. Hoeing was carried out with a powered rotary hoe (5-6 cm depth, driving speed 4 km h<sup>-1</sup>), leaving 12-cm untilled strip in the crop rows. Hoeing-ridging was carried out with the same rotary hoe as mentioned above, but fitted with ridging wings to bury weeds along the row. Split-hoeing close to the row was performed with a Asperg Gartnereibedarf (Asperg, Germany) split-hoe, leaving a 10-cm untilled strip in the crop rows, at a working depth of 3-4 cm and a driving speed of 3 km h<sup>-1</sup>. Finger-weeding close to the row was carried out with a Kress (Tamm, Germany) finger-weeder at a working depth of 1-3 cm and a driving speed of 5 km h<sup>-1</sup>. Pre-emergence herbicides were always applied two days after sowing with a plot sprayer delivering 300 L ha<sup>-1</sup> spray solution at 200 kPa pressure. Sunflower was grown according to normal agronomic practice in central Italy. Four weeks after mechanical treatments the following parameters were assessed both intra-row and inter-row weeds: weed density, weed fresh weight, weed dry weight and weed ground cover (%). At harvest, crop production was determined. All data were analyzed by ANOVA and means were separated by protected LSD at p=0.05.

## Results and discussion

In 2002 and 2003, the weed flora in untreated check (Table 1) was mainly *Amaranthus retroflexus* L., *Chenopodium album* L., *Solanum nigrum* L. and *Echinochloa crus-galli* L. In both years, chemical weed control alone or integrated with inter row hoeing gave the best

weed control and showed an efficacy ranging from 91 to 100% (Table 2). Considering mechanical weed control methods, the best results were obtained by hoeing-ridging showing an efficacy not significantly lower than chemical weed control. Hoeing alone did not show a good weed control, especially in 2002: this low efficacy was due to the lower weed reduction of intra-row weeds than those in the inter-row. Split-hoeing showed good results with an efficacy ranging from 70% to 88%. Finger-weeding showed a satisfactory efficacy only

Table 1. Weed density in the untreated checks. Standard errors are in parentheses.

Weed species	Density (n. m <sup>-2</sup> )	
	2002	2003
<i>Amaranthus retroflexus</i> L.	21 (1.2)	63 (29.1)
<i>Solanum nigrum</i> L.	10 (2.5)	- -
<i>Echinochloa crus-galli</i> L.	9 (3.2)	8 (2.9)
<i>Chenopodium album</i> L.	7 (1.6)	3 (2.7)
Other species	10 (2.4)	11 (1.9)
Total	57 (4.6)	85 (22.2)

against weeds at early growth stages (2-4 true leaves or less) and did not control the weeds at larger growth stages. As a consequence, its weed control efficacy was rather low in both years. However, finger-weeding following split-hoeing caused a significant weed control increase compared to split-hoeing alone. Thus the ranking among mechanical weed control methods based on weed control efficacy was hoeing-ridging > split-hoeing + finger-weeding > split-hoeing > hoeing > finger-weeding. The weed control methods gave different results in terms of weed control but only slightly affected crop yield due to high crop tolerance. All weed control treatments showed a good selectivity to the crop; likewise, differences in crop yield were very small owing to high sunflower tolerance and competitiveness. However, it should be noted that several mechanical methods gave yield levels comparable or higher than herbicide treatments.

Table 2. Effects of different weed control methods in sunflower on weed ground cover, weed density, weed biomass and crop yield (2002-03).

Weed control treatments	Weed reduction (%)						Crop yield (t ha <sup>-1</sup> )	
	Ground cover		Density		Total dry mass			
	2002	2003	2002	2003	2002	2003	2002	2003
Untreated check	-	-	-	-	-	-	3.89	3.81
Herbicide broadcast	99	91	100	95	100	98	4.19	3.91
Herbicide on row + inter-row hoeing	100	98	100	97	100	99	4.39	4.14
Hoeing	75	76	43	79	46	72	4.11	3.98
Hoeing-ridging	96	93	95	90	95	90	4.25	3.95
Split-hoeing	88	85	73	81	70	85	4.26	3.77
Finger-weeding	56	68	46	73	56	61	4.21	3.87
Split-hoeing + finger-weeding	94	91	89	87	90	85	4.44	4.04
<i>LSD</i> ( <i>p</i> =0.05)	5	9	10	16	19	24	0.27	0.22
Untreated check	122	124	57	85	62	55		
	(%)		(plants m <sup>-2</sup> )		(g m <sup>-2</sup> )			

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## Physical weed control in organic carrots in the Fucino Valley (Italy)

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### Introduction

Weed control is a major constrain to organic carrot cropping. A low seed vigour gives the crop a very slow emergence resulting in a low competitive ability against weeds at the early growth stages. The long growth period and the low canopy cover of the soil also contribute to a challenging weed management. Weed control by physical methods seems to be the most important measures to obtain good yields in organic carrot cropping. Brush weeding and flame weeding are the most used methods for weed control, and they are often combined with preventive methods like the stale seedbed technique and night time soil cultivation (Folgerberg, 1998, 1999; Radics *et al.*, 2002). However, although some new techniques and implements can be used for non-chemical weed control in organic carrot, hand weeding is still necessary to achieve sufficient control of intra-row weeds (Ascard, 1990 and 1995).

The aim of this study, carried out in the Fucino Valley (Italy) from 2001 to 2003, was to evaluate the applicability and the efficacy of new strategies for physical weed control in organic carrot (Peruzzi *et al.*, 2002). This paper reports the results obtained in the three years experimental period.

### Materials and methods

In each experimental year, two cropping systems for organic carrot were evaluated: Traditional organic (TO) management and Innovative Organic (IO) management. In the TO system, carrot was sown at a  $2.5 \times 10^6$  seeds  $\text{ha}^{-1}$ ; seeds were randomly distributed within 7 cm wide strips, with a distance of 30 cm between strips. Conventional inter-row hoeing and intra-row hand weeding were used for post emergence weed control. In the IO system, carrot was sown in single rows by a precision drill at the same seed rate as TO but with a distance of 20 cm between single rows. An innovative precision hoe (Peruzzi *et al.*, 2002), characterized by a steering guidance system and two additional tools working in the row (torsion weeders and vibrating tines), was used for post emergence weed control together with hand weeding. In the application of the innovative system, quick hand weeding was made before hoeing to increase the effectiveness of hoeing, which otherwise could be difficult to conduct because of large weeds clogging the hoe. A stale seedbed technique was applied in both cropping systems. Spring tine harrowing was done 1 or 2 times just before sowing. Ten days after sowing, early emerging weeds were flame weeded in both systems.

Carrot (2001-2003) and weed (2002-2003) density were sampled just before and after each weed control treatment. Time consumption for each physical and manual weeding treatment was recorded. The experimental design was a randomised complete block design with 4 replicates. Each plot was  $50 \text{ m}^2$  (2 x 25 m).

Carrots were harvested in a  $1 \text{ m}^2$  area for yield assessment in each plot. The carrots were weighed, counted and graded into four marketable classes according to the EC Regulation n° 730/1999.

### Results

Weed density level varied among years, depending on the climatic conditions. In 2003, the stale seedbed techniques only had a low effect: very little precipitations and cold weather during the sowing period resulted in very little weed emergence. Thus weed competition was



very severe and consequently hand-weeding became very laborious. However, weed effectiveness of innovative hoeing was high in both years (63% and 85% respectively) (Table 1). In 2003 a supplementary hand-weeding before hoeing increased the effectiveness so that the innovative techniques gave a notable lower requirement for hand weeding (- 29%) (Table 2). Time consumption to operate the implements only covered a small amount, usually from 3 to 12%, of the total time consumption in all three years.

Table 1. Weed density (plants m<sup>-2</sup>) counted at different times during the cropping season. In the same row and for the same year, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's Multiple Range test).

	2002		2003	
	TO	IO	TO	IO
Before stale seedbed	100	100	35	35
After stale seedbed	28 b	51 a	0 ns	0 ns
After flame weeding	19 ns	25 ns	0 ns	0 ns
Before first hoeing	157 ns	127 ns	504 ns	488 ns
After first hoeing	18 b	48 a	223 a	73 b
At harvest	4 ns	6 ns	6 ns	4 ns

In 2002 and 2003, crop density at harvest was 28% higher in the innovative system. It could be due to the different spatial patterns of the two systems: in the traditional system, where carrot is sown in strips, it is common that some carrot plants are picked up as well during hand weeding. The higher crop densities at harvest also gave a higher total yield in 2002 and 2003 (+23%), whereas in 2001 no significant differences were seen. In 2003, crop yield was extremely reduced by a long lasting drought. The innovative system also resulted in a better carrot quality in 2002 and 2003 (Table 2).

Table 2 – Carrot yield as fresh roots (total and graded into four classes) obtained with the two different cropping systems (TO Traditional organic, IO Innovative Organic) in 2001, 2002 and 2003. In the same row and year, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's Multiple Range test)

Characteristics	2001		2002		2003	
	TO	IO	TO	IO	TO	IO
N° carrots (plants m <sup>-2</sup> )	135 ns	129,0 ns	140,0 b	179,0 a	139,0 b	178,1 a
Total yield (t ha <sup>-1</sup> )	77.5 ns	75.9 ns	91.8 b	119.0 a	65,3 b	85,1 a
Extra carrots (t ha <sup>-1</sup> )	2.1 a	0.0 b	3.0 ns	2.9 ns	0,0 b	3,1 a
1 <sup>st</sup> cl.carrots (t ha <sup>-1</sup> )	26.6 ns	27.5 ns	22.0 ns	25.3 ns	6,6 ns	14,1 ns
2 <sup>nd</sup> cl.carrots (t ha <sup>-1</sup> )	35.5 ns	35.7 ns	50.9 b	73.5 a	33,7 b	49,3 a
Non-marketable (t ha <sup>-1</sup> )	13,3 ns	12,7 ns	16.0 ns	17.6 ns	25,0 a	18,5 b
Total labour time (h ha <sup>-1</sup> )	215	135	168	166	658	476

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# The rolling harrow: a new implement for physical pre- and post emergence weed control

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## Introduction

Mechanical weed control after a false seedbed is usually made by implements, such as spring tine harrows. However, weed seedlings may survive the treatment, as they are often included in small lumps that are left on the soil after passage of the implement. The same problem has often been detected with hoes equipped with elastic and/or rigid tines for inter-row weed control (Peruzzi, 2003). These problems are obviously more important when the mechanical treatments are made in wet and plastic crusty soils. We observed such problems during four years with experimental tests of physical weed control in organic carrot, spinach, fennel and chicory in different regions of Italy (Peruzzi et al., 2004a and 2004b).

Aiming at solving these very important problems, a new implement, called "rolling harrow" was projected, built, tested and optimized at the Centro Interdipartimentale di Ricerche Agro-Ambientali "E.Avanzi" of the University of Pisa in the two years period 2003-2004. During the construction of the implement, we were inspired by other innovative implements, such as the "basket weeder" (Bowman, 1997). We focused at developing an implement that was able to effectively control weeds even on very bad soil conditions by creating a very shallow tillage but still with a significant crumbling of the soil. This new implement has recently been patented by the authors (Peruzzi et al, 2004c).

## Materials and methods

Two prototypes of the rolling harrow were developed during 2003 and 2004. The first prototype is 1,4 m wide, especially adapted to organic spinach production under Italian conditions, while the second prototype is 2 m wide and meant for organic carrot, fennel and chicory production in the Fucino Valley. Both prototypes are equipped with specific tools. The tools are spike discs placed at the front and gage rolls mounted at the rear. The front and rear tools are connected to one another by a chain drive with a ratio equal to 2 (Fig.1).

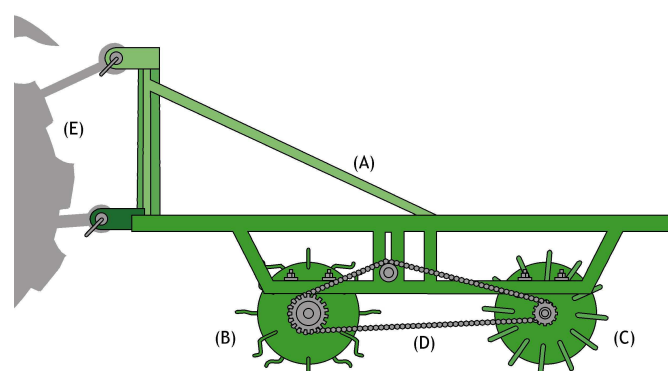


Figure.1 – Scheme of the rolling harrow: (A) frame; (B) front axle equipped with spike discs; (C) rear axle equipped with gage rolls; (D) chain drive; (E) three points linkage.

The discs and the rolls can be placed differently on the axles:

1. close arrangement in order to create a very shallow tillage (3-4 cm) of the whole treated area (for seed-bed preparation and non-selective mechanical weed control after false seed-bed) (Fig.2a);

2. spaced arrangement in order to create efficient selective post-emergence weed control (for precision inter-row weeding) (Fig.2b).

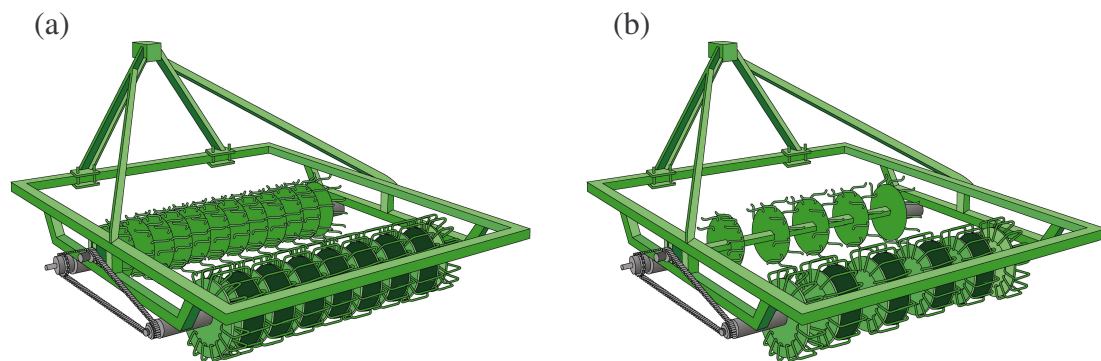


Figure 2 – Scheme of the two arrangements of the tools on the rolling harrow: (a) close arrangement for non-selective treatments; (b) spaced arrangements for precision inter-row weeding.

The action of the rolling harrow is characterized by the passage of the spike discs (that till the top 3-4 cm of the soil ) followed by the passage of the gage rolls that work at a higher peripheral speed, (as the rear axle is driven by the front axle by means of an overdrive with  $\tau=2$ ), tilling and crumbling the 1-2 cm top soil. Moreover, the rolling harrow can be equipped with pairs of elastic tines (working as both vibrating tines and torsion weeders) with the aim to control weeds in the rows.

The new implement was tested in some organic vegetable crops making both a false seedbed and later precision hoeing. The rolling harrow was compared with both spring tine harrows and steerage hoes equipped with rigid tools.

### Results and discussion

The results of the tests made on fields with spinach in the Serchio Valley in Tuscany and with carrot, fennel and chicory in the Fucino Valley in Abruzzo were very good and promising. The rolling harrow performed better than both the spring tine harrow (false seedbed technique) and the steerage hoe equipped with rigid tines (inter-row weeding treatment). In the realization of non-selective treatments, the operative performances of the rotating rolls harrow were always of the same order of those detected for the spring tine harrow. In the realization of inter-row weeding treatments, the performances of the new implement were markedly better with respect to those of the hoeing machine equipped with rigid tools. The good results obtained allow us to suggest this implement for mechanical weed control in other herbaceous and horticultural crops. In this respect, the working width can be varied from 1m and up to 18 m. For hoeing treatments, the rolling harrow can be adjusted to cultivate different inter-row widths, varying from 15 cm and up 90 cm.

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# Physical weed control in organic spinach in the Serchio Valley (Italy)

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## Introduction

Spinach (*Spinacia oleracea* L.) represents one of the most important vegetable crops in the coastal area of Tuscany. In order to produce spinach characterized by high qualitative standards, a reduction of chemical inputs – particularly herbicides - represents a very interesting opportunity for local farmers. Unfortunately, spinach is a relatively poor competitor against weeds, thus specific non-chemical weed control strategies are required. Previous experiments (Peruzzi et al, 2004) showed that innovative weed control strategies, suitable for the local organic farmer, gave significant reductions in weed numbers and weed biomass with a substantial reduction in working time required for direct weed control as compared with traditional organic weed management.

## Materials and methods

Field experiments in the period 2002-2004 were carried out on an organic farm located in the Serchio Valley near Pisa. Spinach was cultivated on ridged soil, the common soil preparation for spinach crop for fresh consumption. Ridges were 1,4 m wide.

Physical weed control was conducted with a specific spring tine harrow, a flame weeder and a precision hoe in spinach under normal cropping conditions typical for farmers in the Serchio Valley, (Peruzzi et al, 2004). Furthermore, a new implement, the rolling harrow (Peruzzi et al, 2005), was built and tested for pre-sowing and post-emergence weed control. Three different mechanical methods for false seed-bed technique were compared: spring tine harrowing, flame weeding and rolling harrowing (Peruzzi et al, 2004). Post-emergence precision hoeing was applied 1 or 2 times. Experiments were designed as a split-plot design with randomised blocks with four replicates. Weed biomass, spinach fresh biomass and number of marketable plants recorded at harvest were analysed according to design: main-plot factor was represented by pre-sowing weed control strategies; sub-plot factor was the number of precision hoeing. In all experiments working times were recorded. Weed density was counted just before each treatment in a 0,225 m<sup>2</sup> sample area in each plot. In this paper, only the results from 2004 obtained in spring spinach will be presented and discussed.

## Results and discussion

Weed density observed in the course of the cultural cycle is presented in Table 1. After the false seed-bed technique, flame weeding and rolling harrowing showed a significant reduction in weed number compared with spring tine harrowing. After sowing and just before first hoeing in plots where flame weeding had been applied, higher weed emergence was observed than with rolling and harrowing. On average soil disturbance caused by mechanical weeding reduces post crop sowing weed presence by 50% when compared to flame weeding. Significant differences between spring tine harrowing and rolling harrowing may be a consequence of a more effective uprooting ability of the new implement.

Weed reduction obtained by means of the first hoeing, carried out at the first true leaves stage, was significantly higher where flame weeding had been applied. Probably subsequent weed flushes following crop sowing showed a different intra and inter-row spatial distribution in relation to the different weeding strategies applied following the false seed-bed technique.

Table 1. Weed density observed during spinach crop cycle and weed biomass present at harvest. In the same row numbers followed by different letters are significantly different at  $P<0.05$  (Duncan's Multiple Range Test).

False seed bed technique Number of hoeing		Spring tine harrow		Flamer		Rolling harrow	
		1	2	1	2	1	2
Before false seed-bed	$n^{\circ} m^{-2}$			537			
After false seed-bed	$n^{\circ} m^{-2}$		12 a	1 b		3 ab	
Before first hoeing	$n^{\circ} m^{-2}$	169 b		311 a		133 c	
After first hoeing	$n^{\circ} m^{-2}$	30 a		12 b		25 a	
Before second hoeing	$n^{\circ} m^{-2}$	135 b		161 a		127 b	
After second hoeing	$n^{\circ} m^{-2}$		49 ab	65 a		22 b	
Weed biomass at harvest	$g m^{-2}$	36.2	32.6	29.1	19.6	43.6	32.3

After the second precision hoeing weed number was reduced more than 65%. Thus, the second post-emergence mechanical intervention seems to have acted more efficiently in plots where rolling harrowing was applied (84% of weed number reduction).

On average, total weed biomass at harvest (Table 1) was significantly reduced when precision hoeing was applied twice in comparison to a single hoeing (about by 30%). As a matter of fact, the fresh marketable produce (Table 2) was significantly higher ( $P<0.05$ ) where precision hoeing had been applied twice. Number of marketable plants showed the same trend.

Table 2. Working times required for physical weed control, fresh marketable produce and density of marketable spinach plants at harvest.

False seed bed technique Number of hoeing		Spring tine harrow		Flamer		Rolling harrow	
		1	2	1	2	1	2
Pre sowing	( $h ha^{-1}$ )		2	2.5		2.2	
Precision hoeing	( $h ha^{-1}$ )	11.8	23.6	11.8	23.6	11,8	23.6
Total working time	( $h ha^{-1}$ )	13.8	25.5	14.3	26.0	13.9	25.7
Fresh product	( $t ha^{-1}$ )	14.2	15.7	14.7	16.1	13.9	15.8
Marketable plants	( $n^{\circ} m^{-2}$ )	29	33	30	34	29	33

Effectiveness of pre-sowing rolling harrow application was associated with a very moderate labour requirement. Moreover, the total working time was obviously strongly reduced when only a single precision hoeing was applied. Results from the other years confirm that physical weed control in organic spinach can be performed by means of the new mechanical implements with very good results. False seed-bed technique always reduced weed numbers considerably leading to substantial savings in labour time. False seed-bed strategies with repeated application of a course of different implements (i.e. rolling harrow and flamer) should be investigated to clarify when secondary flushes of weed emergence occur. More knowledge on this aspect could result in further reductions in labour time for post-emergence weed control.

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## Weed control by steam and compounds causing an exothermic reaction

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### Introduction

A new system for soil disinfection (including weed seed control) by means of steam injection following the incorporation of compounds (KOH, CaO, etc.) in the soil that cause an exothermic reaction was developed during a six year period (1998-2004) by the Celli firm in co-operation with researchers from the University of Pisa (Peruzzi *et al.*, 2002). This system was tested under various environmental and operational conditions and against different plant pathogens and pests with very promising results, both in terms of control (e.g. of fungi and nematodes) and of increased growth of several vegetable crops.

Moreover, the system seems to have potential for weed control, because it can reduce weed seedling emergence considerably (Bàrberi *et al.*, 2002, Moonen *et al.*, 2002). A specific experiment was carried out in 2003 under controlled conditions to evaluate the effects of this system on seeds of both winter- and summer annual weeds.

### Materials and methods

The experiment was carried out on plastic cages in which steam was injected at a depth of 15 cm through a specific dispenser. The amount of steam used was the same as that commonly used under field conditions. Four doses (corresponding to 1000, 2000, 3000 and 4000 kg ha<sup>-1</sup>) of each of two compounds (KOH and CaO) causing an exothermic reaction were compared to steaming only and to an untreated control. The compounds were mixed into a sandy soil down to a depth of 15 cm, while weed seeds were placed in permeable small plastic bags (100 seeds/250 cm<sup>3</sup>) resistant to high temperatures and to chemical impact. The bags were placed at 7.5 cm soil depth. Soil temperature was recorded throughout the experiment. The effect of the treatments was evaluated on seeds of three winter annuals (*Alopecurus myosuroides* Hudson, *Matricaria chamomilla* L., and *Raphanus raphanistrum* L.) and four summer annuals (*Amaranthus retroflexus* L., *Echinochloa crus-galli* L., *Bilderdykia convolvulus* L. and *Setaria viridis* L.). After the treatments, soil was extracted from the bags and placed in plastic tubs, which were arranged on a table according to a completely randomised design and watered regularly. Weed seedlings were counted daily until no further emergence occurred. The effects of the different treatments were determined both in terms of number of emerged seedlings (density) and seedling density reduction relative to the untreated control. Data were subjected to ANOVA according to a completely randomised experimental design (for comparison of the 10 treatments altogether) or a factorial design (for testing substance by dose interactions).

### Results and discussion

Treatment effects differed among weed species (Table 1); however, species in the two weed groups (winter annuals and summer annuals) showed some common responses. All species from the first group showed a higher sensitivity to KOH than to CaO with reductions in seedling emergence > 90% for all the three species when treated with steam + 4000 kg ha<sup>-1</sup> KOH. Except from a fairly good control exerted on *A. myosuroides*, the use of steam alone slightly stimulated germination of *M. chamomilla* and *R. raphanistrum*. Apparently, *M. chamomilla* was the species most difficult to control with this system, since it was also less sensitive to CaO than the other two winter species. In contrast, *A. myosuroides* was the most sensitive species, for which even steam alone or steam + 1000 kg ha<sup>-1</sup> KOH resulted in high



seedling emergence reductions. A significant interaction between substance and dose was found only in the case of *R. raphanistrum*.

Treatment effects on summer annuals showed a similar response for *A. retroflexus* and *E. crus-galli* (less sensitive) and for *B. convolvulus* and *S. viridis* (more sensitive). Emergence of the latter two species was significantly reduced even by steam only, while that of *A. retroflexus* and *E. crus-galli* was not.

Table 1. Mean number of seedlings emerged after the steaming treatments for the two winter annual and the four summer annual species. Within a column, values sharing the same letter are not significantly different (Duncan's Multiple Range test at  $P \leq 0.05$ ).

Treatment	Winter annual species			Summer annual species			
	ALOMY	MATCH	RAPRA	AMARE	ECHCG	POLCO	SETVI
Control	77,0 a	82,0 a	87,0 a	69,8 a	89,5 a	83,0 a	80,3 a
Steam	17,5 b	91,0 a	90,3 a	59,8 ab	78,0 ab	46,5 b	49,3 b
KOH 1000	10,8 de	37,0 bc	24,0 c	50,0 b	74,0 abc	26,8 c	39,5 bc
KOH 2000	8,8 def	23,0 cd	10,5 d	27,0 c	56,0 bcd	22,8 cd	34,0 bcd
KOH 3000	5,5 fg	11,0 de	8,8 d	25,0 c	49,8 cde	18,8 cd	28,8 cd
KOH 4000	4,0 g	3,3 e	6,0 d	22,5 c	17,5 f	14,5 de	21,8 d
CaO 1000	15,8 bc	49,3 b	41,3 b	23,5 c	73,2 abc	26,5 c	42,8 bc
CaO 2000	12,3 cd	44,5 b	18,0 c	17,0 c	56,0 bcd	23,5 cd	29,0 cd
CaO 3000	7,8 efg	36,0 bc	9,5 d	12,5 c	46,8 de	15,3 de	28,5 cd
CaO 4000	7,0 efg	20,3 d	7,8 d	11,8 c	30,5 ef	7,8 e	17,8 d

Treatment doses are in  $\text{kg ha}^{-1}$ . ALOMY, *Alopecurus myosuroides*; MATCH, *Matricaria chamomilla*; RAPRA, *Raphanus raphanistrum*; AMARE, *Amaranthus retroflexus*; ECHCG, *Echinochloa crus-galli*, POLCO, *Bilderdykia convolvulus*; SETVI, *Setaria viridis*.

Steaming still allowed part of *E. crus-galli* seeds to emerge even when high doses of compounds were used. For winter annual species, the type of compound did not seem to influence seedling emergence to a great extent. Of the species tested, only *A. retroflexus* showed a higher sensitivity to CaO than to KOH. There was a clear tendency of increased seedling reduction with increased doses for all summer annuals and, in general, emergence of all the tested species was considerably reduced with  $4000 \text{ kg ha}^{-1}$  of both compounds. The interaction between substance and dose was not significant for any of the tested summer species.

In conclusion, the effect of steaming with the addition of activating compounds on the preventive control of selected weed species is encouraging, thus confirming that this system is able to considerably reduce weed seedling emergence. More research is needed to test the sensitivity to the system of other weed species, as well as the potential control exerted by other compounds able to cause an exothermic reaction.

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# Weeds and weed management of dry peas in Finland

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## Introduction

Pea (*Pisum sativum* L.) is a minor field crop grown only on around 5,000 hectares in Finland. In 1999, 59% of the total pea area was under organic production. A research program “Pea as a source for domestic protein” was launched in 2002 in Finland to improve yield and quality stability and economics of pea production through better crop management. As a sub-project of the program, a survey of weeds in pea fields was carried out in the main dry pea production area in southwestern Finland in 2002-2003 (Salonen *et al.*, 2005).

The objective of the survey was to compare the species composition of weed communities in pea fields under organic and conventional cropping and to explore the relative importance of different cropping measures as well as other factors explaining the variation in species composition.

## Materials and methods

The number of pea fields examined was 93 in 2002 and 90 in 2003. In both years 32 fields were under organic production. The majority of pea fields were pure stands, but 29 fields in 2002 and 23 fields in 2003 were mixed stands, predominantly (87%) pea with oats (*Avena sativa* L.). Dry peas were grown for food, feed or seed.

The occurrence of weeds was assessed in the first week of July from five 1 m<sup>2</sup> (1.0 m x 1.0 m) sample quadrats randomly located in each field (median size 3.8 ha). Weed cover was visually assessed and recorded using a scale of 0-3 (0 = not present, 1 = less than 5% cover, 2 = 5 to 25% cover and 3 = more than 25% cover) by species. The weed cover data from five sample quadrats were pooled and the sum was applied as a measure of weed abundance. The term frequency refers to the proportion of fields where the species was found in quadrats.

Data on factors involved in each field were collected through observation, measurement or by interviewing the farmer. The data on weeds and a set of explanatory variables were analysed using the Redundancy Analysis (RDA) procedure of the CANOCO software.

## Results and discussion

A total of 76 weed species were recorded, of which 29 exceeded 10% frequency. The average number of weed species per field was 10 (min 3, max 21) under conventional farming and 18 (min 8, max 31) under organic farming.

The most frequent weed species in organically cropped pea were *Chenopodium album*, *Galeopsis* spp. and *Stellaria media*. In conventionally cropped fields the three most frequent species were *Viola arvensis*, *C. album* and *S. media* (Fig. 1). In addition, *Elymus repens* and *Erysimum cheiranthoides* were abundant in organic production as indicated by observations on ground cover.

In the RDA analysis, the main gradient in the variation in species composition along the first ordination axis was the difference between conventionally and organically cropped fields. Under conventional cropping the most important factors explaining the variation in the species composition were the crop stand characteristics (e.g. height, median 45 cm) and chemical weed control, which indicated the selective pressure of different herbicides. Under organic cropping, the most significant factors were respectively the age of crop stand from sowing to assessment and the field location but not e.g. intercropping with cereals.

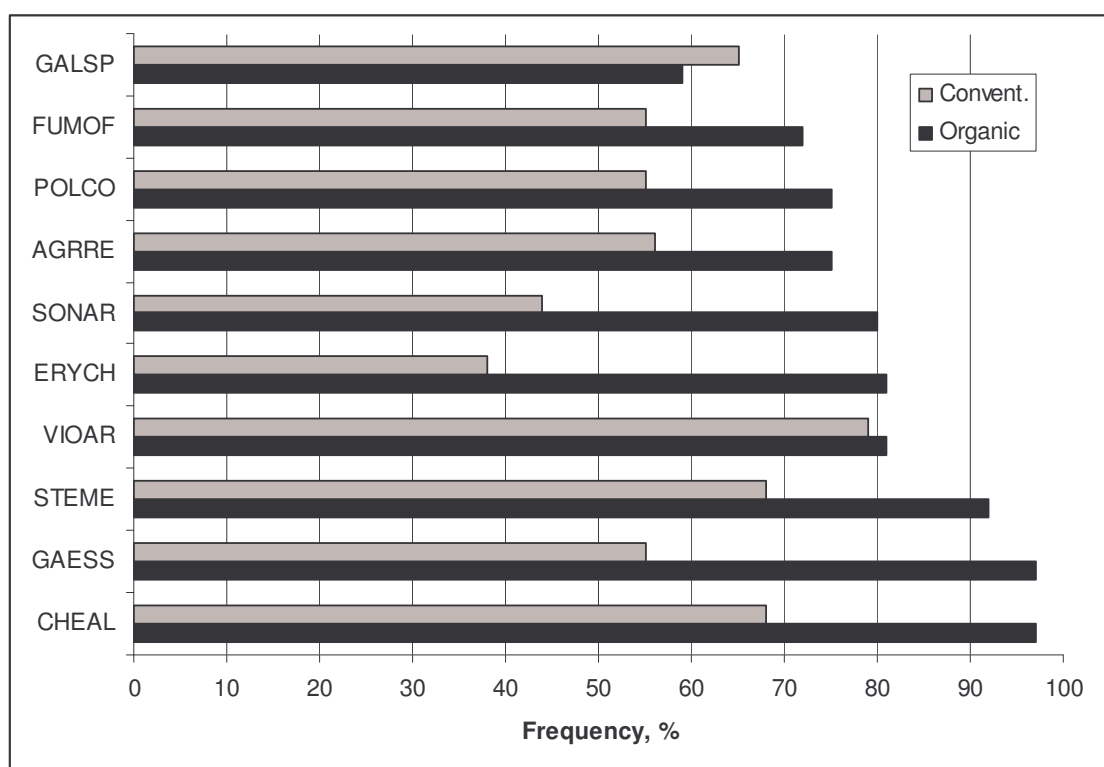


Fig. 1. Frequency of occurrence of the most common weed species in pea fields under conventional (119 fields) and organic (64 fields) cropping.

In conventional production herbicides were applied in 92% of fields and were effective for weed control but at high cost (30 to 95 € ha<sup>-1</sup>). Bentazon and metribuzin were the most frequently applied herbicides. Tank mixtures of various active ingredients (including MCPA and acetonitrile) were common. Characteristic weed species recorded after herbicide application included *Galium spurium*, *Fumaria officinalis* and *Viola arvensis*. Weeds were controlled mechanically only in five organic fields reflecting the slight implementation of physical weed control methods in Finland in general.

Pea was grown in cereal-dominated crop rotations on most farms. Obviously this explains the similarity in species composition between the surveyed pea fields and the spring cereal fields surveyed in the late 1990s (Salonen *et al.*, 2001). Similarities in weed flora of pea fields were recorded even at the European level (Uludag *et al.*, 2003).

Weeds were effectively controlled with herbicides in conventional pea cropping, but they represented a significant problem in organic cropping. Mixed cultivation of pea with cereals is recommended, particularly in organic cropping, as it favours competition of semi-leafless pea varieties against weeds.

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# Control of swallow wort (*Cynancum acutum* L.) in apple orchards

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## Introduction

Swallow wort (*Cynancum acutum* L., SW) is a perennial vine from *Asclepiadaceae* family. It climbs fruit trees and if not controlled it virtually strangles them to death. This weed has been imported to Iran via the northern borders and has rapidly invaded orchards in the northern half of Iran. Some herbicides have been reported to control this weed to a certain extent. Bahat (1987) has recommended glufosinate ammonium (Basta 20% SL) at 20 l ha<sup>-1</sup> to control SW. Soters *et al.* (1983) believes that dicamba, 2,4-D, and glyphosate can adequately control SW. Badaly (1997) has reported nicosulfuron to control SW in maize fields. Coble & Slife (1970) have reported that atrazine or 2,4-D can only control shoots of SW, and not its perennial roots. Meister (2002) recommends isoxaben, and glyphosate for the control of *Cynancum laeve*. This research was conducted in Ghazvin, 100 km west of Tehran city in an apple orchard which was heavily infested with SW. All available physical and chemical methods were tried to find the best way of controlling this invasive and dangerous weed.

## Materials and methods

This experiment was conducted in a Randomised Complete Block Design with 4 replications and 10 treatments. Each plot included two trees. All treatments were applied at the 10-15 cm height of SW. The experimental plots were fixed for all the three years of experiment. The treatments were as follows: (1) paraquat (20% SL) at 3 l ha<sup>-1</sup> + a second treatment of glyphosate (41% SL) + 8 kg ha<sup>-1</sup> of ammonium sulphate at the regrowth of SW; (2) two applications of glyphosate at the same rate of treatment 1; (3) one application of nicosulfuron at 1.5 l ha<sup>-1</sup>; (4) one application of paraquat at 3 l ha<sup>-1</sup> + diuron (80% WP) at 2 kg ha<sup>-1</sup>; (5) twice soil level cutting of SW, at the third regrowth, application of glyphosate as in the other treatments; (6) Same as treatment 5 but with only one cutting of SW; (7 & 8) two or three soil level cuttings of SW, respectively; (9) weedy check; (10) one application of diuron + glyphosate mixture at the same rates used in the other treatments. Percent cover of SW as compared to weedy check was estimated periodically in each spring before treatments were applied. All data was statistically analysed and treatments were compared using Duncan's Multiple Range Test.

## Results and discussion

As shown in Tab. 1, during the first year of the experiment, and at the end of growing season (3 months after the application of first treatments), the best treatments were 2 and 6, followed by 1, 4, 7 and 8. In the next spring, effect of last season's treatments was observed in the plots, but the lowest number of SW was detected in treatments 1 and 2. In second and third years, only treatments 1 and 2 (especially the latter) resulted in the lowest number of SW. Finally, in the spring next to the third year only treatment 2 had the lowest number of SW. These results indicate that it is not possible to control SW permanently in orchards, the remedy being to prevent it from reaching the tree and climbing on to it. The best chemical treatment was application of 6 l ha<sup>-1</sup> of glyphosate + 8 kg ha<sup>-1</sup> ammonium sulphate at the 10 to 15 cm height of SW, and repeating it once the weed has regrown to the same height. Mechanical control of SW may prevent it from reaching the tree, but it stimulates new stems emission.

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## Weeds and weed management in cabbages - a review

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The EWRS Working Group "Weed Management Systems in Vegetables" was established with the aim of collecting and disseminating information and results on weeds and weed control strategies in vegetables, identifying gaps in knowledge and defining new research projects. So far the working group has published reviews on onions (Tei et al., 1999a), tomatoes (Tei et al., 1999b; 2002b), carrots (Tei et al., 2002a) and peas (Uludag et al., 2003a; 2003b).

Information about key weeds, new weeds or species that have recently become problematic, effect of competition, weed management programmes in integrated and organic production, approved herbicides and those currently undergoing registration for use in cabbages grown under field conditions in Croatia (HR), Finland (FIN), Germany (D), Hungary (H), Italy (I), The Netherlands (NL), Poland (PL), Portugal (P), Slovenia (SLO), Spain (E), Switzerland (CH) and United Kingdom (UK) was collected.

In 2003, the world production of cabbages (i.e. headed cabbages, savoy cabbage, cauliflower, broccoli, Brussels sprouts and Chinese cabbage) was 66 million tons on 3.2 million hectares. In the surveyed countries, cabbages were cropped on 436000 ha (PL 47000 ha, I 37000 ha, E 34000 ha, UK 33000 ha, D 19370 ha, HR 11000 ha, NL 10800 ha, P 8500 ha, H 6200 ha, FIN 1320 ha, SLO 1000 ha and CH 500 ha).

Information on organic production is very scarce: about 5-10% of total production in H, less than 5% in SLO, about 3% in FIN and a few hundred hectares in PL and UK.

Cabbages are mostly transplanted but Chinese cabbage is commonly direct seeded. The season of planting is extensive involving sequential cropping from spring through to autumn. Row distance is 0.50 to 0.80 m with a planting density 2 to 6 plants m<sup>-2</sup> depending on species and cultivar. Horticultural brassicas are usually grown on moisture retentive as well as well-drained soils (i.e. silty to silty-loam soils) but they can also be grown on heavy calcareous clay soils.

The weed communities are commonly very species rich and their composition is highly variable in relation to climate, soil and crop period. The most important and frequent species are *Digitaria* spp., *Echinochloa crus-galli*, *Elymus repens*, *Setaria* spp., *Amaranthus* spp., *Chenopodium* spp., *Datura stramonium*, *Mercurialis annua*, *Polygonum* spp., *Portulaca oleracea* and *Solanum nigrum* in spring-summer crops and *Stellaria media*, *Cirsium arvense*, *Matricaria* spp., *Senecio vulgaris*, *Sonchus* spp., *Fumaria officinalis* and cruciferous weeds in autumn-winter crops.

*Chenopodium* spp. and *Galinsoga parviflora* are key weeds in H, PL, CH and I, *Urtica urens* in NL and UK (locally). *Cruciferae* species (i.e. *Capsella bursa-pastoris*, *Diplotaxis erucoides*, *Sinapis arvensis*, *Thlaspi arvense*) are dominant species in FIN, UK, I and E. Further key weeds are: *Galium aparine* in some areas of the UK; *Sonchus* spp. in CH; *Cyperus esculentus*, *C. rotundus* and *Rumex* spp. in P; *P. oleracea* and *C. rotundus* in E; *Alopecurus myosuroides*, *Lolium* spp., *Papaver rhoeas*, *Veronica* spp. in I.

IWM can control most weeds but some species are becoming important: *C. album* and *Polygonum* spp. in FIN; *U. urens* in NL; *Ambrosia artemisiifolia* in H; *Rorippa sylvestris* in CH; *Abutilon theophrasti*, *Panicum* spp., *Setaria viridis*, *Xanthium strumarium* in HR; *Amaranthus* spp., *C. arvense*, *Convolvulus arvensis*, *Cruciferae* in SLO; *C. album*, *Cuscuta* spp., *G. parviflora* in P; *Calystegia sepium*, *Rumex* spp., *Sorghum halepense* in I.



Competitive ability of cabbages is generally greater than other vegetable crops (e.g. carrots, onions, lettuces...). Research has shown that no critical period of competition can be determined: a single weeding at 3-5 weeks after transplanting resulted in crop yield similar to those for weed-free cabbage.

Herbicides approved for use in the different cabbage species are very different among the countries. Taking into consideration only the “key” active ingredients, clopyralid is authorised in FIN, I, PL and UK; metazachlor is approved in all the countries except in H and P; napropamide in HR, H, I, PL, SLO, CH, and under registration in FIN; oxyfluorfen in HR, I, PL, P and SLO; pendimethalin in HR, D, H, I, PL, P, SLO and E; pyridate in FIN, D, SLO, E and UK; propachlor in I, PL, E, CH, and UK; trifluralin in all the countries except in FIN, NL and SLO. A wide range of post-emergence graminicides is registered in all the countries except in PL for cauliflower, broccoli and Chinese cabbage.

Conventional weed control involves a soil incorporated pre-emergence/pre-transplanting treatment (trifluralin, napropamide) or non-incorporated pre-emergence/pre-transplanting treatment (pendimethalin, oxyfluorfen, propachlor, metazachlor) followed by one to two post-emergence/post-transplanting treatments (metazachlor, pyridate, clopyralid, propachlor, graminicides).

Trifluralin has a narrow spectrum of activity and it requires moisture for optimum activity; moreover in dry seasons crop phytotoxicity may occur. Propachlor fails to control *Polygonum aviculare* and *Chenopodium album*. Pyridate may be applied for the control of *Galium aparine* in cabbage and Brussels sprouts while clopyralid controls *Cirsium arvense*, *Matricaria* spp., and *Sonchus* spp.

However, mechanical weed control (harrowing, hoeing, ridging) is often used to compensate for poor herbicide efficacy.

IWM generally involves: 1) false seedbed technique followed by shallow harrowings or by glyphosate or glufosinate-ammonium application; 2) pre-emergence or pre-transplanting herbicide application; 3) post-emergence inter-row hoeing or rotary cultivation combined with ridging for in-row weed control.

Both in conventional and integrated weed control, very early head cabbages and cauliflower can be grown in the open field under perforated polyethylene plastic flat cover or non-woven polypropylene to improve crop earliness and to control insects.

Common strategy for organic production is: 1) false seedbed technique followed by shallow harrowing; 2) transplanting; 3) repeated inter- and intra-row cultivation through the growing season sometime combined with ridging; 4) hand-weeding. Some growers also flame weeds under the cabbage leaves when the crop plants are big enough. False seedbed for early cabbage production or for very late varieties seems not feasible. In organic growing of cabbages the use of plastic covers is more common than in conventional and IWM systems but it stimulates weed emergence and growth and costs are high because it must be removed before each mechanical weed control. In Spain black plastic mulching combined with drip irrigation is widely used in summer crops.

Further and more detailed information can be found in the WG web site [www.agr.unipg.it/ewrsveg/](http://www.agr.unipg.it/ewrsveg/)

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# Risks of weed infestations from manures fermented in a biogas plant

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## Introduction

Weeds interfere with crop production by e.g. lowering yield and quality, increasing production costs, being a transmitter of numerous plant diseases, and decreasing soil fertility as they remove large amount of nutrients from the soil. The large supply of viable weed seeds from manures lowers the benefits of such fertilizers. Moreover, manures promote weed seed production including their competitive ability against the crop.

Weed seeds preserve their viability over long periods, particularly when they remain dormant. Seed dormancy can be broken by scarification, light and temperature (stratification).

## Materials and methods

We determined that feedstuffs from hay products, cattle excretal and semi-liquefied manures were the main sources causing weed seeds to enter into a biogas plant. The samples were selected according to common methodical recommendations with assistance from other guidelines as well (further information can be given on request). In every sample, weed seeds were identified and their viability was tested.

As the amount of weed seeds that enters with the manure into the biogas plant, and because their specific relation may vary in the course of the year, it was decided to determine the loss of seed viability in the plant according to the working regime prevailing under summer-autumn and winter-spring conditions, respectively. We developed a special method to attain a more objective evaluation of how the fermentation conditions influenced weed seed kill. So, a lot of 25 of the most common and important weed seeds packed in nylon net bags were placed inside the reservoir and before the manure was fermented. Seeds of these weed species were collected from fields and had been kept under conditions close to those in the storehouse. Seed viability was 72-99% before putting them into the manure.

When the fermentation cycle was completed, weed seeds were removed and tested for their germinating ability under laboratory conditions at an optimum temperature of 22-24°C.

For comparison seeds of the same weed species were placed in a manure pit and then composted for the same period as in the biogas plant. When the storage period had ended, the seeds were removed and their viability was determined.

## Results and discussion

It was determined that 1 kg of hay product used for cattle feeding contained 10125 seeds of all the species found, among which 5425 or 53,6% were viable. Weed species found were *Chenopodium album* (70%), *Stellaria media* (5%), *Matricaria inodora* (3%), *Echinochloa crus-galli* (3%) and *Polygonum scabrum* (2%).

One kg of horned cattle excrements taken from a farm contained 235 weed seeds where 115 or 48,9% were viable. The proportion of individual species was 31% for *S. media*, *C. album* 30%, *M. inodora* 8%, and *E. crus-galli* 5%.

There were only 126 seeds per kg in seed stocks of a semi-liquid manure of which 51 seeds per kg or 40,5% were viable. Weed species such as *C. album* 30%, *E. crus-galli* 10%, *S. media* 10%, *P. scabrum* 4%, and other grass weeds 20% will enter into a biogas installation with semi-liquid manure.

The results showed that weed seeds with viabilities up to 30-31% may enter a field following an anaerobic manure fermentation process in a biogas plant both under winter-spring and summer-autumn conditions (Fig. 1).

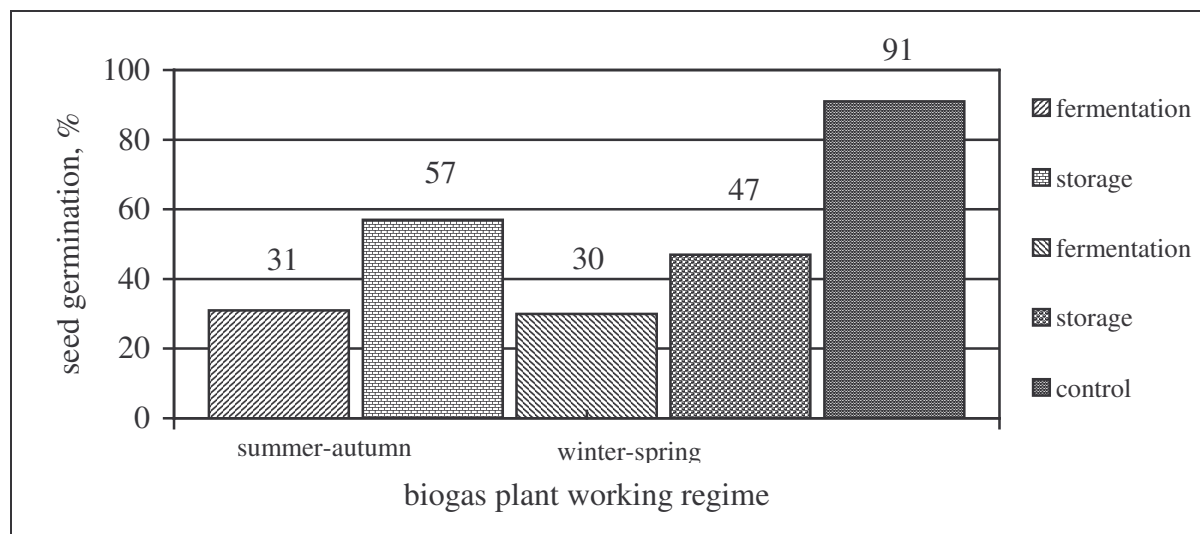


Figure 1. Influence of anaerobic manure fermentation on weed seed viability.

After summer-autumn fermentation, the following species may enter the field: *Amaranthus retroflexus* viable for 90%, *Galeopsis* spp. 88%, *Polygonum convolvulus* 71%, *C. album* 68%, *Rumex confertus* 51%. And after winter-spring fermentation: *Galeopsis* spp. 88%, *Melandrium album* 80%, *R. confertus* 72%, *S. media* 67%, *C. album* and *P. convolvulus* 66% may enter.

Fermented manures are completely free of species such as *M. inodora*, *Taraxacum officinale*, *Agrostis*, *Plantago lanceolata*, and species from the *Brassicaceae* family.

Anaerobic fermentation of manures in a biogas plant is in our opinion one of the most perspective technologies to obtain organic fertilizers of high quality. However, complete removal of weed seed viability in the manures will require longer fermentation cycles or an increase in the temperature during the fermentation process.

## Innovation in mechanical weed control in crop rows

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### Introduction

Reasons for increasing interest in non-chemical weed control are: concerns for pollution of ground and surface water with herbicides, human health risks due to herbicides, site effects on the flora and fauna, and development of herbicide resistance. In organic farming, weed control, especially in the crop rows, is one of the main problems. Especially in slow growing and non-competitive crops, the labour required for hand weeding is expensive and difficult to organize. The input of manual weeding in crop rows varies from around 45 h ha<sup>-1</sup> for planted vegetables to 175 h ha<sup>-1</sup> for direct-sown onion under Dutch field conditions. Different implements are used and/or in development for controlling weeds in the crop rows. During the last decade, research has focused successively on harrowing, torsion and finger weeding, weeding with high pressured air (Pneumat) and hoes that move weeding tools out of the crop row for every crop plant. Possibilities vary with crop type, growth stage and environmental conditions.

### Materials and methods

In several European countries, trials have been done in several crops with different machinery. Guidelines for such experiments have been made available by Vanhala *et al.* (2004) as an activity of the EWRS working group *Physical and Cultural Weed Control*. Also a glossary with pictures of the implements and experimental results are presented by this working group ([www.ewrs.org/pwc](http://www.ewrs.org/pwc)). Here, we use this information and currently unpublished results from other sources to indicate the possibilities but also the challenges and shortcomings of mechanical weed control in the crop rows.

### Results and discussion

Harrowing is a long-standing method for mechanical control in the crop rows. Although the harrows were modernized, especially in terms of possibilities to adjust tines and working capacity (up to 24 m wide and driving speed up to 12 km h<sup>-1</sup>), still the selectivity at early crop growth stages is limited. By harrowing only small weeds (until the first true leaves are visible) can be effective but frequent treatments are often needed for sufficient control during the entire growing season. Spring tine harrows can be used in cereals, maize, potatoes, peas, beans, many planted vegetables and relatively sensitive crops such as sugar beet. In sensitive crops, harrows cannot be used at the young crop growth stages (e.g. before 4 true leaves in sugar beet), and manual weeding is the only option in organic crops, while additional use of herbicides is an option in integrated production. Besides the risk of crop damage and high cultivation frequency, dependence on favourable weather (dry periods) and soil conditions (loose soil and few stones) are major drawbacks to mechanical control in the crop rows.

Finger weeders and torsion weeders have been imported and modified from implements operating in the United States. Compared to the harrow, the finger and torsion weeders have the disadvantage that they need a very accurate and reliable steering to work as close as possible in the crop rows, and thus their working capacity is relatively low. However, they are gentler to the crop and can easily be combined with hoeing. The finger and torsion weeders operate from the sides of the crop row and beneath most of the crop leaves. As with intrarow brush weeding, finger weeders also cause relatively more uprooting of the weeds and move them away from the crop rows. Finger weeders are more effective against weeds with true

leaves, but still also for this weeder, the weeds need to be small and/or easy to uproot. The amount of manual weeding can be reduced by 40 to 70% using finger or torsion weeders (Table 1). The tools can be used in many transplanted vegetables, beans, spring seed rape, seeded onions (from 2 leaves stage), red beet and sugar beet (from 2-4 leaves), carrots (from 2 leaves stage). The pneumat weeder controls also larger weeds by blowing them out of the crop row. Some results indicate additional advantages of using the pneumat in broad crop rows such as tulip.

Table 1. Number of hours ha<sup>-1</sup> of manual weeding to control remaining weeds after mechanical weed control with different implements in direct-sown or transplanted onions.

Implement	Direct-sown	Direct-sown	Direct-sown	Transplanted
	2002	2003	2004	2004
Hoeing	188	108	79	30
+ Finger weeding	78	56	41	9
+ Torsion weeding	99	38		
+ Finger/Torsion		39	42	
+ Pneumat				8
+ Sarl Radis				7

Similar to the harrows, finger weeders, torsion weeders and the Pneumat still treat both the crop and the weeds. Selectivity based on difference in anchorage strength, leaf area and/or plant height is needed to prevent crop damage. Intelligent weeders are needed for more sophisticated control of even larger weeds without treating the crop plants. One of the first commercially available new intelligent weeder, the Sarl Radis, has a simple crop detection system based on light interception and moves a hoe in and out of the crop row around the crop plants. Currently, several small companies in cooperation with research develop other intelligent weeders that use computer vision to recognize the crop plants with the aim of guiding weeding tools in and out of the crop row. Denmark (Melander, 2004) and Germany (Gerhard, 2003) are focusing on developing sensors or cameras to distinguish between crop and weed plants. The University of Sweden has a prototype working in sugar beet (<http://www2.hh.se/staff/bjorn/mech-weed>). In the Netherlands, we develop and test together with Danish, German and Dutch companies two different intelligent intrarow weeders.

We seem to be at the start of a new era of innovation in physical weed control, currently driven by available technology and the high dependence and costs for manual weeding in organic farming. However, it is desired that these new innovations can be commercialized and partly become true alternatives to herbicides.

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# Assessing common vetch-cereal intercrops for suppression of wild oat

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## Introduction

Common vetch (*Vicia sativa* L.) and winter cereals are cultivated as monocultures for hay production which is used as livestock food. Unfortunately, vetch or cereal monoculture hay yield is not satisfactory (Osman & Nersoyan 1986). This fact is attributed to cereal low quality, particularly in protein and vetch low yield and low concentration in carbohydrates. Vetch and cereals can also be cultivated as intercrop in order to improve the quality of produced hay (Thompson *et al.*, 1992), but selective herbicides for this intercrop do not exist. Moreover, cereal species, seed proportion, as well as their competition affect yield and quality of the final product (Droushiotis, 1989; Caballero *et al.*, 1995). The objectives of this research were to assess the influence of common vetch and four cereal monocultures as well as eight vetch-cereal intercrops on wild oat (*Avena sterilis* L.) emergence and growth and to evaluate common vetch and cereal tolerance to postemergence applied imazamox.

## Materials and methods

Common vetch 'Melissa', wheat (*Triticum aestivum* L.) 'Yecora', triticale (Triticosecale) 'Thisvi', barley (*Hordeum vulgare* L.) 'Thessaloniki', and oat (*Avena sativa* L.) 'Pallini' monocultures as well as vetch-cereal intercrops, in two percent seed ratios (65:35 and 55:45), were planted at 170 kg seed ha<sup>-1</sup> in a sandy loam soil with pH 7.0 and organic matter 1.0% at the Farm of Aristotle University of Thessaloniki, Greece. The planting date was 21 November 2003. A split-plot design was deployed in a randomized complete block layout with four replicates. Plot size was 5 by 12 m. In each plot four subplots of 5.0 by 3.0 m were created. The 13 monocultures or intercrops were the main plot factor levels and the nontreated vs. herbicide-treated at 12 weeks after planting were the subplot factor levels. Wild oat control in these herbicide treated subplots was achieved with 0.03 or 0.05 kg a.i. ha<sup>-1</sup> of imazamox and 0.03 + 0.56 kg a.i. ha<sup>-1</sup> of imazamox + diclofop applied post-emergence. Wild oat plants were harvested in a 1 m<sup>2</sup> area in the centre of each untreated subplot 0, 3, 6, and 9 weeks after completion of the cereal tillering stage. Wild oat stem number as well as fresh weight was measured at each sampling date. Data for wild oat stem number and fresh weight were regressed against time (weeks after completion of tillering). In these regression equations, stem number and fresh weight were the dependent variables (y) and time the independent variable (x).

## Results and discussion

Both stem number and fresh weight of wild oat were significantly affected by crop species, duration of interference, and their interactions. Therefore, the crop species x duration of interference interaction means are presented (Fig. 1 and 2). Regression equations of wild oat stem number and fresh weight on time (weeks after completion of tillering) indicated that, in most cases, the quadratic equation ( $y = a + bx + cx^2$ ) provided the best fit (data not shown). Wild oat emergence and growth was reduced in common vetch monoculture as compared with cereal monocultures (Fig. 1 and 2). Similarly, Mas & Verdú (2003) reported that winter pea (*Pisum sativum* L.) crop had the least weed infestation as compare to winter cereal crops. Also, vetch-cereal intercrops were not effective for wild oat reduction with the exception of vetch-oat mixtures. Imazamox applied postemergence alone or along with diclofop gave effective control of wild oat 4 weeks after application, but reduced dramatically growth of vetch (50-85%), cereals (> 95%), and their mixtures (data not shown). The results showed



that post-emergence applied imazamox cannot be used in vetch-cereal intercrops. In addition, vetch monoculture as well as vetch-oat intercrops showed significant competitive advantage over wild oat.

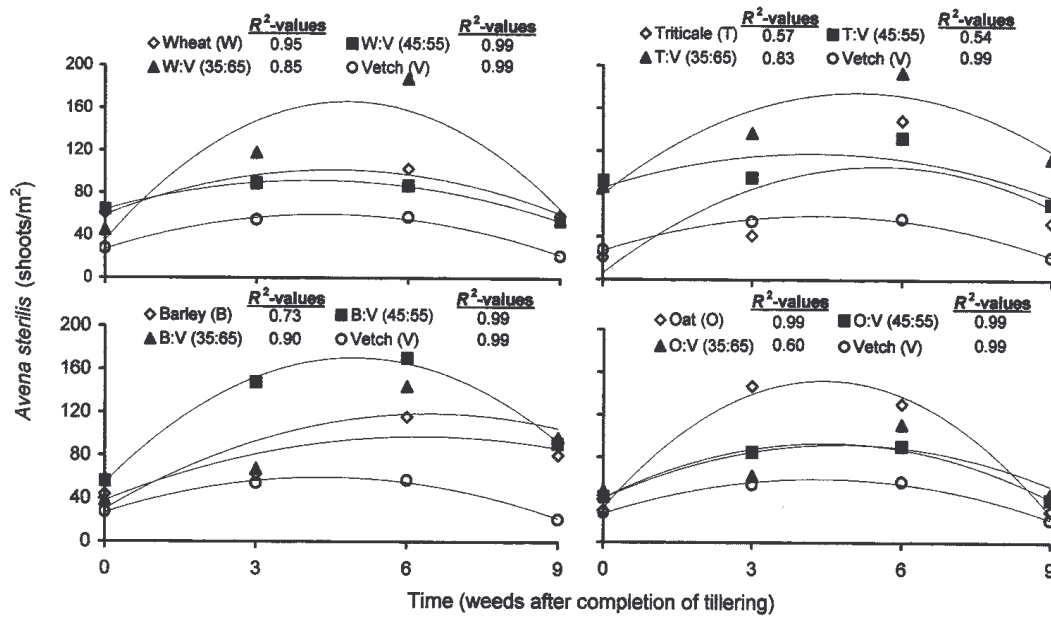


Fig. 1. Temporal pattern of shoot number of wild oat grown in vetch, four winter cereals, and their mixtures. Lines describe quadratic regression equations.

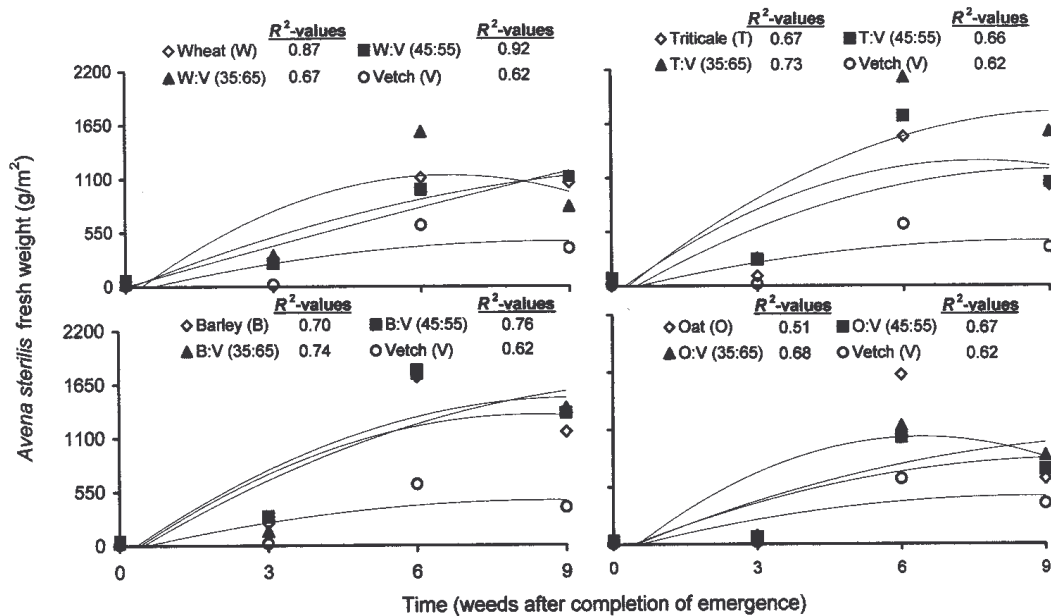


Fig. 2. Temporal pattern of fresh weight of wild oat grown in vetch, four winter cereals, and their mixtures. Lines describe quadratic regression equations.

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## Different weed control systems in tomato

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### Introduction

In the latest years several physical methods have been developed for weed control. These methods can be interesting in organic farming and for integrated production. In this context, thermal weeding (selective heat employment for the elimination of weeds), mechanical weed control with horizontal brush weeder between the crop lines, and mulching with black plastic or crop residues are accepted weed control methods by the regulations of organic farming. Nevertheless, there is little empirical evidence about advantages of these weed control systems (Leroux *et al.*, 2000; Bàrberi, 2002). Suso *et al.* (2003) compared the above mentioned weeding systems for tomato crop with two trials located at Logroño and Zaragoza, Spain. Results of one year showed that the plastic mulching was a better weeding system, which in addition gave higher yield. However, to confirm these results, it is necessary to repeat the trial several years. In this work, different weed control systems were compared in three trials to analyse the efficiency in weed control and the effects on yield.

### Materials and methods

Three trials were performed in Montañana (Zaragoza, Spain) in 2002, 2003 and 2004. The experimental design was a randomised block with five treatments and four replications. Table 1 shows the description of the five treatments tested every year.

Tab. 1. Treatments description in trials for every year. HW: hand-weeded. DAT: days after transplanting.

Treatment	2002	2003	2004
1) Check with herbicide	Glyphosate (36%) (impregnation)	Metribuzin (70%) 0.5 l ha <sup>-1</sup> + glyphosate (36%) (impregnation)	Metribuzin (70%) 0.5 l ha <sup>-1</sup> + rimsulfuron (25%) 30 g ha <sup>-1</sup>
2) Horizontal brush weeder	2 times (22 and 34 DAT)	1 time (13 DAT) + 1 HW (16 DAT)	1 time (19 DAT) + 1 HW (21 DAT)
3) Flame weeder	5 times (13, 21, 27, 36, 44 DAT)	2 times (10 and 24 DAT) + 1 HW (16 DAT)	3 times (21, 28 and 41 DAT) + 1 HW (51 DAT)
4) Black plastic mulch	Polyethylene 15 μ + glyphosate (36%) (impregnation)	Polyethylene 15 μ + glyphosate (36%) (impregnation)	Polyethylene 15 μ + glyphosate (36%) (impregnation)
5) <i>Artemisia absinthium</i> straw mulching	2 HW +2 applications (1.6 + 1.6 kg m <sup>-2</sup> , 10 and 26 DAT)	2 HW + 2 applications (6.6 + 5.4 kg m <sup>-2</sup> , 2 and 30 DAT)	2 HW + 2 applications (3.4 + 1 kg m <sup>-2</sup> , 8 and 29 DAT)

Treatment 2 was performed with a horizontal rotary brush which operated between the crop lines. Thermal weed control was applied between the crop lines with a manual flame weeder (propane burner of 37 x 13 cm). Glyphosate dose for impregnation in treatments 1) and 4) was on average 9.9 ml a.i plot<sup>-1</sup> (12 linear m). The transplant was done in single rows 1.5 m apart. Tomato cv. 'Perfectpeel' was used and planted at 20 cm between plants. The dates of transplanting were 21

May, 10 June and 26 May in 2002, 2003 and 2004 respectively. Fertilisation was done preplant with foliar fertiliser. Every year several assessments of weed density were done in 3 m<sup>2</sup> of each elementary plot. In 2002, weed plants were counted three times during the crop season (17, 34 and 52 DAT), in 2003 only one time (17 DAT) and in 2004 two times (27 and 51 DAT). The crop was harvested ca. 120 DAT. For yield determination 40 linear m per treatment were harvested.

## Results

**Weed control:** best weed control was generally achieved with black plastic mulch (Tab. 2). The representative weed species were: *Cyperus rotundus*, *Convolvulus arvensis*, *Chenopodium album*, *Portulaca oleracea*, *Amaranthus blitoides* and *Amaranthus retroflexus*.

**Yield:** the yield in 2002 differed between treatments being highest for black plastic mulch and *Artemisia* mulch. In 2003 yield was lowest for these two treatments due to high temperatures and excessive *Artemisia* straw application, respectively. Tomato yield was insignificant in treatment 4. In 2004, no significant yield differences were found (Tab. 2).

Tab. 2. Average density of *Cyperus rotundus* and annuals weeds (plants m<sup>-2</sup>) and commercial tomato yield (t ha<sup>-1</sup>) per treatment and year. Numbers refer to treatments described in Table 1.

Treatment	2002			2003			2004		
	<i>Cyperus</i>	Annuals	Yield	<i>Cyperus</i>	Annuals	Yield	<i>Cyperus</i>	Annuals	Yield
1	92 b	22 a	77.4 bc	58 ns	3 ns	76.6 a	26 ns	2 ab	65.4 a
2	114 ab	22 a	57.6 c	51 ns	2 ns	64.6 a	34 ns	7 ab	74.5 a
3	161 a	11 a	61.8 bc	27 ns	6 ns	72.6 a	22 ns	9 a	78.5 a
4	49 b	2 a	98.1 a	29 ns	0 ns	-	22 ns	1 b	69.7 a
5	55 b	15 a	81.8 ab	34 ns	6 ns	30.6 b	49 ns	7 ab	67.8 a

In each year, treatments with different letters are significantly different (LSD test at P < 0.05), ns = not significant.

## Conclusions

The best treatment for weed control was achieved with the black plastic mulch, which was very effective on annual weeds but not on *C. rotundus*, which perforated it. Effectiveness of the other treatments was erratic. The best yield was also obtained with black plastic mulch, but only during 2002 with an unusually cold summer. In contrast, results of the second year show that this system is not to be recommended with high temperatures (as found by Radics & Székelyné Bognár, 2002). It was also found that *Artemisia* straw application should be done carefully as too big quantities apparently delayed tomato growth, resulting in a significant lower yield. Under ordinary weather conditions (as in 2004), all the treatments gave similar yields, so that weather conditions strongly affect the choose of any tested control method. Disadvantages of the brush weeder and the flame weeder use are the need of a precise labour to avoid damaging the crops. With regard to *Artemisia* use it is necessary to properly calculate the quantity (biomass) and manpower requirements to implement it.

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## **A software to compute early weed competition from cover images analysis**

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### **Introduction.**

A better description of the status of a crop and its associated weed flora at early stages all over a field is needed for an unbiased estimation of potential weed competition in that field and – in the context of Precision Agriculture – for a spatially modulated weed control. One prerequisite is the development of integrated tools where image analysis techniques are combined with an estimation of the crop-weed interaction effects at a small scale. Next, spatial integration at larger scale would be conducted by a spatial interpolating technique as kriging. We present an integrated software devised to compute competition effects between maize and the pool of weeds present at early stages of growth. Although species identification within the weed pool has not yet been completely done by vision technique (Manh et al., 2001), it would be introduced by the user as a priori knowledge of the main weed species present in the field.

### **Image acquisition**

Prior to crop sowing, several annual weeds were sown in order to simulate a patchy weed flora. Color images were acquired by a standard 35 mm camera held on an aluminum frame in that maize field at 2-3 leaf stage of the crop. Colour slides film were used, then digitized and coded in TIF format. Each image represented 113 x 76 cm of soil surface and the resolution was 0.05 cm per pixel.

### **Image & leaf segmentation.**

A few image samples were used to determine color statistics of the vegetation (mean & covariance matrix in the RGB space). Then each image was segmented using these color statistics, obtaining binary images in which the vegetation pixels were separated from the background.

### **Binary image analysis & leaf classification.**

Binary objects (i.e. groups of connected pixels) do not always correspond to individual plants, especially in areas of higher weed density. Therefore, specific pattern recognition criteria were designed to separate group of weeds from maize plants: tip density (tips were counted all along the binary object contour), ratio object area versus tip density, maximum length of included segment, object area, contour length, etc. These criteria were used to feed a K-Nearest Neighbour classification module. Five output classes were used: 1) individual or group of maize plants, 2) individual or group of maize + individual weeds, 3) individual or group of maize + group of weeds, 4) individual weed, 5) group of weeds. At the moment, no distinction between weed species is integrated in the software.

### **Graphical output.**

In user-defined mesh size (about 4 x 4 cm), 5 parallel zones are constructed, the 2 maize-rows (north and south) and 3 crop-free zones (north, inter-row and south). From previous module, cover estimation for the 5 classes were computed for each zone. A symbolic image can be obtained with a grey level scaling for plant cover (Figure 1).

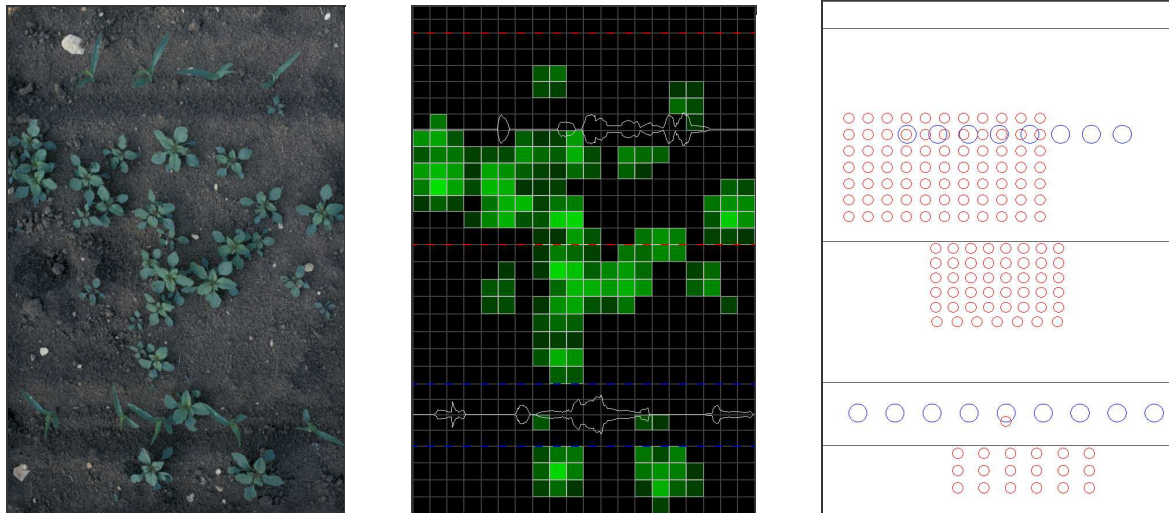


Figure 1. Original colour image (113 x 76 cm), symbolic image and its translation with plant mock-up.

### Competition.

From spatial information of cover for the 5 classes of vegetation in the 5 zones, an explicit model of plant competition for light was used (Assémat, 1998; Sinoquet & Bonhomme, 1992); the radiation interception between plants was computed from the reconstruction of the vegetation by mock-up that simulate shape and size in 3D space for maize and main weed species at various development stages. Competition indices were built from the relative reduction of light interception by maize due to weeds, for the whole image or for separate zones.

### Results & discussion.

Different validations are possible. First, the usefulness of such software relies of its flexibility (easiness in changing parts of modules independently) and the validity of each step can be checked independently. Then modules to validate main parts has been devised (leaf classification, cover reconstruction, light regime estimation within cover, ..). But an overall validation is also possible when initial estimate of competition indices at early stages and final status of the crop (i.e yield) are correlated. From an experiment done in 2002 where 253 images quadrats were systematically sampled, these different aspects were studied. Higher plot densities gave unsatisfying results, mainly because of bad maize rows identification by image analysis; but in these very weedy parts of the field the decision to control weeds is less problematic.

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# Influence of sampling accuracy on parameters describing the aggregation of weeds in arable fields

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## Introduction

In order to answer the question of whether or not weeds in arable fields are distributed in irregularly spread patches, several statistical parameters have been applied in weed science. On the basis of aggregation indices such as *Lloyds* patchiness index (PI) (Lloyd, 1967), conclusions are drawn from the results of spatial sampling concerning the characteristics of weeds spatial appearance. In this study, a simulation experiment was conducted applying a custom geoinformation - software (SAMPLING SIMULATOR) in order to investigate the performance of this parameter in relation to sampling grid resolution and counting quadrat size.

## Materials and methods

A weed density of 2.0 plants per square metre distributed uniformly inside of a rectangular arable field was simulated in this study. Applying the above mentioned software package (Backes *et al.*, 2004) it was possible to determine the number of weeds theoretically counted in relation to the size of a counting quadrat and the sampling grid resolution applied. Besides several further measures, the resulting PI value and the standard deviation (SDV) could be calculated for every possible combination of sampling grids and counting quadrat sizes.

## Results and discussion

The results of this study clearly indicate that the commonly applied measures such as *Lloyds* PI are not capable of reliably drawing conclusions on the distribution of weeds in arable fields.

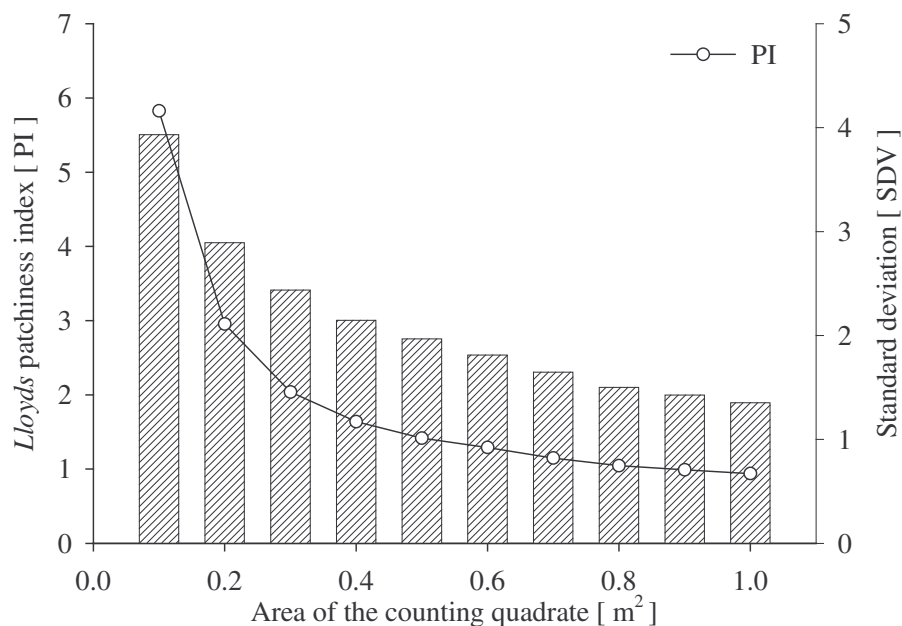


Figure 1. *Lloyds* patchiness index (PI) and the standard deviation of weeds counted in relation to the area of the counting quadrat used. The weeds in this simulation (2.0 plants per m<sup>2</sup>) were distributed uniformly in the arable field.



In Figure 1 the above described uniform distribution of weeds was used in order to simulate the value of *Lloyds* PI in relation to the area of a counting quadrat. It is obvious that a small counting quadrat area leads to the impression of a patchy distribution whereas a larger counting area seems to prove the opposite on the same arable field. According to several authors (e.g. Hamouz *et al.*, 2004) a PI value of  $> 1$  indicates a patchy distribution. In this particular case a value of equal to or below 1 would be correct. In the case of small sampling areas, the SDV is sensitive to this variation (Fig. 1). Consequently, the variation obtained with small counting quadrates is not caused by the spatially patchy distributed weeds but by random effects caused by the size of the counting quadrat. This clearly indicates that none of these values is capable of producing adequate results for the interpretation of weeds spatial distribution. The influence of the sampling grid resolution is demonstrated in Figure 2 for the same simulated population. Using a  $1 \text{ m}^2$  counting quadrat, the PI values obtained were generally close to 1, exceeding this value in only one case. This indicates a slightly patchy distribution of weeds in the arable field. Nevertheless, if we use the  $PI_r$  index (Indicating the PI calculated from the weeds in the equidistant rectangular area surrounding each sampling quadrat - the reference area  $r$  -) a completely different conclusion is reached. This difference is probably resulting from the SDV demonstrated in Figure 1 for the respective counting quadrates area. Further investigations on new measures for the description of weeds spatial distribution should reveal more reliable results.

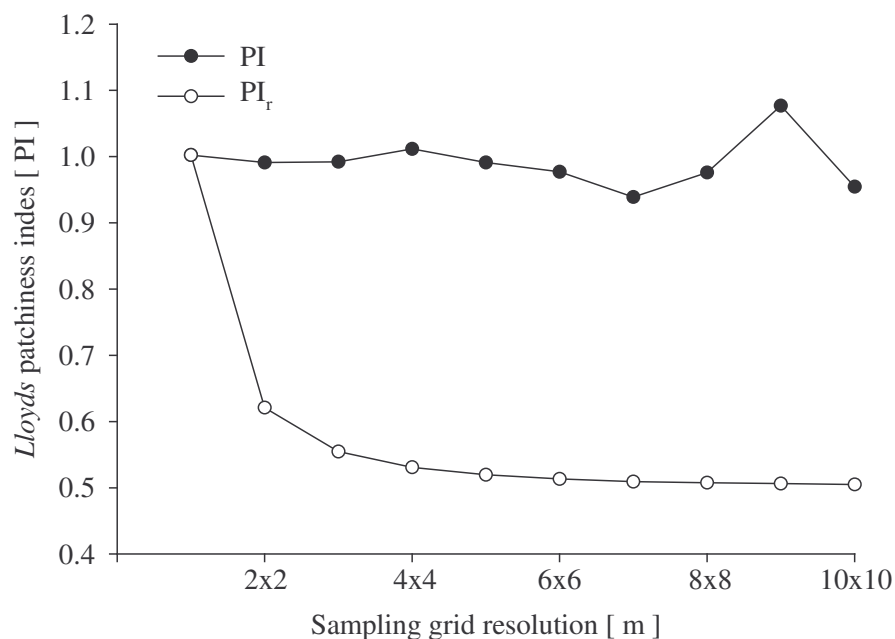


Figure 2. *Lloyds* patchiness index (PI) and  $PI_r$  which is the PI calculated from the underlying simulated distribution in relation to sampling grid resolution applied. The weeds in this simulation ( $2.0 \text{ plants per m}^2$ ) were distributed uniformly in the field.

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# Practical experiences with a system for site-specific weed control using real-time image analysis and GPS-controlled patch spraying (TURBO)

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## Introduction

Weed seedling populations have been found to be spatially and temporally heterogeneous within agricultural fields. Weeds often occurred in aggregated patches of varying size or in stripes along the field borders and along the direction of cultivation (Marshall, 1988; Gerhards *et al.*, 1997; Dieleman & Mortensen, 1999). Progress in Information technologies including digital image analysis, Geographic Information Systems and GPS-controlled patch spraying allow to take this heterogeneity into account for weed management practices.

## Materials and methods

For automatic weed detection, 3 digital bi-spectral cameras were mounted in the front of the sprayer (Gerhards *et al.* 2002). With each bi-spectral camera, a differential image (NIR-VIS) was taken in real-time. The advantage of the bi-spectral camera was that high quality images with a strong contrast between green plant and soil, mulch and stones were achieved even under variable illumination and soil moisture conditions. Artificial lightning was not necessary. The cameras were triggered with an exposure time of 1/4000 s to get well focused images at a speed of 7-8 km/h. Approximately every 2 m a set of 3 images was taken and stored on an on-board computer of the vehicle together with the location co-ordinates of the images. Plant species in the images were identified based on shape analysis. Average identification rate was 80% when plant species were grouped into 5 different classes according to their sensitivity to herbicides and competitiveness. Weed distribution maps were created from analysed images and compared to manual weed distribution assessments (Figure 1). Herbicide selection in winter wheat, winter barley, spring barley, sugar beet and maize was based on regional guidelines in Germany for the range of species and plant size observed. A multiple sprayer with 3 separated hydraulic circuits was built for this study (Figure 2). This sprayer allows varying the herbicide application on-the-go. Each of the 3 sprayer circuits has a boom width of 21 m, divided into 7 sections of 3 m. During the herbicide application, the spray control system was linked to an on-board computer loaded with the weed treatment maps for three weed classes (e.g. grass weeds, *Galium aparine* and other broad-leaved species) A differential mode GPS was used for real-time location of the patch sprayer. The on-board computer compared the actual position of the sprayer with the information in the weed treatment maps and signals were transmitted to the control unit via a data bus to open each individual solenoid valve when herbicide application was warranted (Figure 2).

## Results and discussion

In a six-year study, herbicide use with this map-based approach was reduced in winter cereals by 60 % for herbicides against broad-leaved weeds and 90 % for grass weed herbicides. In sugar beet and maize, average savings for grass weed herbicides were 78 % in maize, and 36 % in sugar beet. For herbicides against broad-leaved weeds, 11 % were saved in maize and 41 % in sugar beet. Savings in spring barley ranged from 20 to 80 % for herbicides against broad-leaved species and 18 – 90 % for grass weed herbicides. The efficacy of weed control in all fields was high enough that average weed density did not increase in the six-year period of study. Only in one field density of *Galium aparine* has increased within the six-year study. Therefore, site-specific weed control offers a great potential for herbicide use in arable fields.

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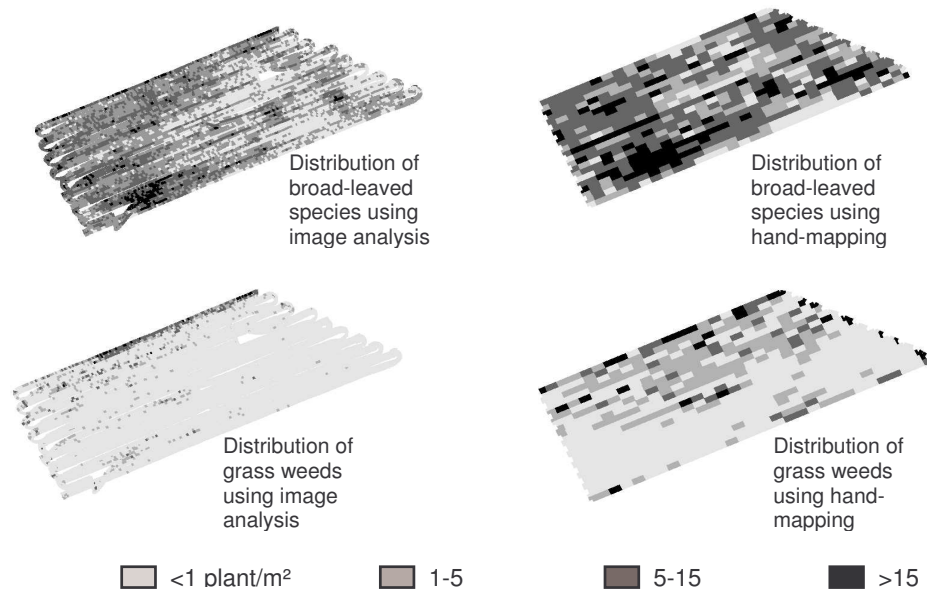


Figure 1. Distribution maps for grass weeds and broad-leaved weed species on a 5.8 ha sugar beet field at Dikopshof Research Station; camera sampling was performed on 2003-04-24; manual sampling was done on 2003-05-08.

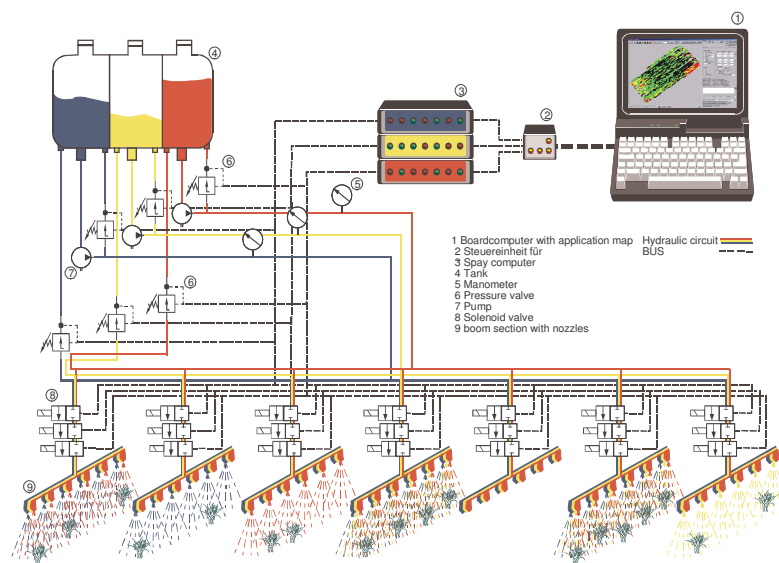


Figure 2. Schematic description of the GPS-controlled multiple patch sprayer with three hydraulic circuits for the variation of herbicide mixture on the go

# **SIMCE: A knowledge-based system for identification of weed seedlings in cereals**

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## **Introduction**

A knowledge-based system is a computer program that uses knowledge to solve problems that are difficult enough to require human expertise. Knowledge-based systems have been developed for many kinds of applications within agriculture, involving diagnosis, predictions, consultation, control, etc. (Gonzalez-Andujar & Recio, 1996; Knight, 1997). In weed science, most knowledge-based developments have mainly focused on weed control, particularly for herbicide selection (Castro-Tendero & García-Torres, 1995; Stigliani *et al.*, 1993). Only a few knowledge-based systems have been reported to be used for weed identification (Gonzalez-Andujar *et al.*, 1990; Olmos y Recasens, 1995; Pasqual, 1994). In this paper we describe a knowledge-based system developed to provide farmers and extension specialists with information for identification of the main cereal seedling of weed species in Spain. The computer program has an image support system which helps non-weed specialists to key in the weed species. Moreover, the system can be used for educational purposes.

## **Materials and methods**

Two steps were used in the development of SIMCE:

### *Use of printed and visual materials*

We acquired textual and graphical information from the literature, such as extension booklets, literature, etc. The printed material allowed familiarization with the subject and a more effective communication with the experts.

### *Interviewing the expert*

The interview methods allowed us to take out the heuristic knowledge, which was not present in the printed material.

Unstructured and structured interviews were used. The unstructured interviews were used to define the familiar tasks involved in the process of identification, to obtain an initial understanding of the range of complications involved, and to define specific problems for later discussion. The questions were more or less spontaneous, and notes were taken for discussion. These methods were completed with structured interviews. In the structured interviews, we made in-depth revision and discussion of familiar tasks in order to clarify questions and to establish a hierarchical process of identification. Through this method, the knowledge can be entered directly at the level appropriate to the input and output relationship of the system. It provided a trustworthy procedure for knowledge acquisition.

### *Knowledge organization*

The hierarchical classification (Schulthess *et al.*, 1996) was used in the development of the SIMCE identification system. In hierarchical classifications, knowledge is organized in a decision tree, with nodes at different levels. The knowledge is distributed over several nodes. At the most general level, distinction may, e.g., be made between a grass or a broadleaf weed, while at the lowest level node, the weed species may be represented individually. At each

level we used a combination of text and pictures (in JPG format) to improve the identification process.

The knowledge base contains 41 weed seedlings commonly found in cereals in Spain and 128 digital colour images.

### **Program requirements**

Two versions of SIMCE have been developed. One is a PC-version that operates under Windows 98 and Windows XP, requiring 16 Mb of RAM and 40 Mb of hard disk, while the second one is a web-based system.

### **System evaluation**

The evaluation process was carried out in the following two steps: verification and validation (Harrison, 1991). In the verification step, we determined the possible errors in the knowledge-system and ensured that the system performed as intended. The system was verified periodically throughout the development process by different weed specialists.

The second step of the evaluation was the validation. In this process, we used the methodology validation by the end users or live testing (Mosqueira-Rey & Monet-Bonillo, 2000). The validation team was a group of 14 students from agricultural courses. All the students were computer users and non-experts in weed identification. Each student was asked to identify a group of selected previously identified cases. Samples were obtained from nearby fields. A total of 123 identifications were performed. Students with system help were able to identify 70% cases correctly. Prior to the validation process, comments in writing on applicability, interface design, functionality and output were requested.

On the basis of the test results, recommendations for improving the expert system were identified. In order to improve the system, suggestions provided by the participants concerning the interface and more pictures were taken into consideration.

### **Acknowledgement**

We are greatly indebted to the Spanish Ministry of Education and Science (R+D project AGL 2002-3801) for their financial support.

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# Evaluation of weed distribution and weed spatial stability for precision farming

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## Introduction

Weeds occur non-uniformly across the arable land. Intensity of weed infestation is often spatially heterogeneous even within one field. (Clay *et al.* 1999, Gerhards, R. *et al.*, 2000). This variability has been ignored by weed management decisions in the praxis. Herbicides are mostly applied in one dose on the whole field. Site-specific weed control allows reduction of herbicide dose in areas with low weed infestation and so diminishes the environmental contamination. To determine whether use of site-specific weed management is appropriate, the level of weed spatial variability in the field must be known. However, manual weed mapping is time consuming. Werner and Garbe (1998) suggest, that because of the stability of weed patches, it is not necessary to carry out mapping every year and historical maps for the forecasting of weediness can be used. This paper discusses the spatial variability and stability of weed populations in various fields.

## Materials and methods

Weed mapping was carried out on 3 fields with total area of 188 ha in central Bohemia to characterize the spatial structure and temporal stability of weed populations over six years (1999 – 2004). A rectangular grid 40 x 40 m or 20 x 40 m was established with use of GPS on trial fields. Number of plants for each species and total weed coverage were investigated on the grid points. To determinate the weed occurrence heterogeneity, the Lloyd's Patchiness-Index (PI) was calculated. For individual weed species, maps of infestation were created and the Pearson's correlation coefficient between years was calculated.

## Results and discussion

At the Klučov site the weed patches were situated mainly in the lower part of the field. *Galium aparine* was distributed patchily. The area with a density below the threshold (0.2 plants per m<sup>2</sup>) represents up to 72.6 % of the total area depending on the year. A high aggregation level was also found in *Cirsium arvense*, which only formed small aggregations across the field, and *Tripleurospermum maritimum* which tended to occur mainly in the field margins. In contrast, *Viola arvensis* was observed at high infestation levels in most years and showed a lower variability. In 2000 it was present on 82 % of the sampling points. The Trebově I site was characterized by an accumulation of weeds in the western part of the field. An aggregated spatial pattern was found in *G. aparine*, which was detected on 52.3 % of sampling points in 2001 and 27.9 % in 2003. *Viola arvensis* and *Lamium amplexicaule* only occurred in the NW-part of the field and likewise showed a high Patchiness-index. A strong patchy distribution was also shown for *Fumaria officinalis* and *T. maritimum*. The concentration of these species was found on the field headland. Most homogeneity occurred in *Stellaria media*, which was present at approximately 70 % of points in both years. The Trebově II site presented generally low weed infestation (Fig. 1). Weed coverage exceeded the threshold of 1 % on 3.6 % of field area only. *G. aparine* occurred on 13.2 % of sampled points on this field. For important species, the mean values of weed density and PI in selected years are summarized in Table 1. Correlation analysis shows, that the weed patch stability is largely dependent on the specific conditions of the site. At Klučov, a satisfactory correlation coefficient was only obtained in some years for *G. aparine* ( $r = 0.19 - 0.64$ ) and *V. arvensis* ( $r = 0.22 - 0.60$ ). In other weeds the correlation was poor.



Table 1: Mean weed density (plants/m<sup>2</sup>) and Patchiness-Index (PI) for surveyed fields.

Weed	Field Klucov				Field Trebovle I				Field Trebovle II	
	2000		2002		2001		2003		2004	
	Mean	PI	Mean	PI	Mean	PI	Mean	PI	Mean	PI
<i>Galium aparine</i>	1.28	7.43	0.55	7.70	1.93	4.68	0.32	9.97	0.08	26.8
<i>Viola arvensis</i>	3.86	2.14	4.23	2.36	4.35	6.99	2.82	8.07	-	-
<i>Cirsium arvense</i>	0.34	9.31	0.21	15.6	-	-	-	-	-	-
<i>Veronica persica</i>	0.83	11.9	-	-	2.08	3.67	0.22	5.87	0.29	19.7
<i>Stellaria media</i>	0.13	14.9	0.06	42.3	1.65	2.37	2.29	3.09	-	-
<i>Tripleurospermum maritimum</i>	0.14	11.0	0.13	217.3	0.15	16.8	0.08	51.5	0.1	25.3
<i>Apera spica-venti</i>	-	-	0.54	9.52	-	-	5.11	12.3	-	-
<i>Fumaria officinalis</i>	-	-	-	-	4.19	5.99	0.63	13.9	2.00	7.04
Weeds total	20.90	1.46	9.04	1.87	18.68	2.50	23.4	2.44	2.98	4.17
Weed cover	3.46	2.5	0.38	2.57	5.29	3.21	2.97	2.54	0.22	9.76

At Trebovle I, high correlation coefficients were found between 2001 and 2003 for *G. aparine* ( $r = 0.68$ ), *V. arvensis* ( $r = 0.68$ ), *Capsella bursa-pastoris* ( $r = 0.68$ ), *L. amplexicaule* ( $r = 0.68$ ), *S. media* ( $r = 0.68$ ), and *F. officinalis* ( $r = 0.68$ ).

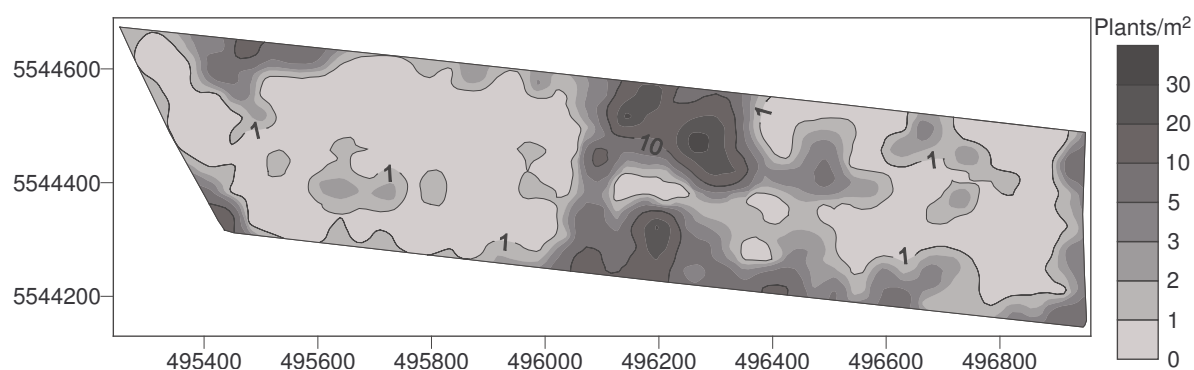


Figure 1. Total weed infestation in the field Trebovle II (2004)

Results show large differences among fields and document heterogeneous occurrence of weeds. *C. arvense*, *G. aprine*, *T. maritimum* and *F. officinalis* showed mainly irregular distribution whereas *V. arvensis* and *S. media* were distributed more uniformly. *G. aparine* and *V. arvensis* were characterized by relatively high spatial stability. Patchy weed distribution offers large potential for using site-specific weed control on some fields.

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# Species specific spatial relations between weed patterns and soil characteristics

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## Introduction

Densities of weed species have been found to vary greatly within arable fields (Christensen *et al.*, 1999). Weed patterns are often spatially aggregated, thus offering possibilities for site specific weed management (SSWM). One of the factors which can possibly explain patterns of weeds in arable fields is the spatial pattern of soil characteristics. Spatial relations between weed species and soil properties were investigated in several studies (Andreasen *et al.*, 1991; Walter *et al.*, 2002; Dille *et al.*, 2002; Dieleman *et al.*, 2000; Heisel *et al.*, 1999). Ultimately, if consistent spatial relations between weed species and soil properties can be established, the presence of weeds can be predicted on the basis of information on the spatial variation of soil properties. Heisel *et al.* (1999) used details on silt content of a field for co-kriging *Lamium* spp and improve the prediction variance. Dille *et al.* (2002) adequately predicted probabilities of *Setaria* spp. and *Solanum ptycanthum* using site property factors and presence of these weeds in a previous year. The aim of the study at present was to investigate the presence and nature of spatial relations between the most dominant weed species and soil characteristics in a field cropped with continuous maize.

## Materials and methods

Weed spatial patterns were mapped in contiguous quadrats of 0.75m×0.75m in a 12m wide (16 quadrats) by 50.25m (67 quadrats) long plot located near the headlands on a 1.8 ha clay soil arable field in 3 subsequent years (2001, 2002 & 2003). During the study, maize was grown at a row distance of 0.75 m. Number of plants per species was counted per quadrat before herbicide application. On 17<sup>th</sup> of April 2003, soil sampling took place in 12m×48m part of the field, coinciding with 16×64 quadrats of the total of 16×67 quadrats of the observation plot nearest to the headlands. Soil samples (151 in total) were taken in a stratified random manner, meeting geostatistical requirements. An Edelman soil drill was used, till 25 cm depth. Soil samples were air dried (40°C), crushed and sieved to remove particles > 2mm. Soil pH and available nitrogen (as N-NO<sub>3</sub>, N-NH<sub>4</sub> and N-total soluble), phosphorous and potassium content were determined by extraction with 0.01 M CaCl<sub>2</sub>. Available magnesium content was established with 0.01 M CaCl<sub>2</sub> (ICP-MS). Soil organic matter content was determined by loss-on-ignition. Soil texture was characterized by determination of the particle size distribution by sieve and pipette. Initial data analysis revealed that *Echinochloa crus-galli* and *Chenopodium polyspermum* were the most dominant weed species and that they exhibited significant relations with most soil factors, which were consistent throughout the years. Spatial dependence of soil variables was studied by fitting variograms. Relation between the weed species (response variable) and their environment (co-variates) were further investigated using Generalized Linear Models (GLM's). Best performing GLM with Poisson-log link function was found using a stepwise procedure. Performance of all models was measured with Akaike's Information Criterion (AIC). In a next step, models were defined in which the residuals were spatially correlated according to a spherical variogram model and Taylor's Power Law was used as a link function as described by Dalthorp (2004).

## Results and discussion

Spatial patterns of the dominant weed species are shown in Figure 1 & 2. Sill/nugget ratios revealed that part of the variation of the soil variables could be explained spatially (not shown here). Depending upon the details of the procedure, stepwise GLM selected the following covariates for the models of the three year weed-soil relations:

*E. crus-galli* ~ + pH + NO<sub>3</sub><sup>-</sup> - sand fraction - organic matter content

*C. polyspermum* ~ + pH - silt fraction

Selection of NO<sub>3</sub><sup>-</sup> could be doubted as it is known to vary temporally. The level of scale studied here is relatively small compared to other studies. Relations between weed species and soil properties can vary between fields (Walter *et al.*, 2002). To determine the scope of these models for SSWM in general, similar studies in other fields and at various levels of scale should be done.

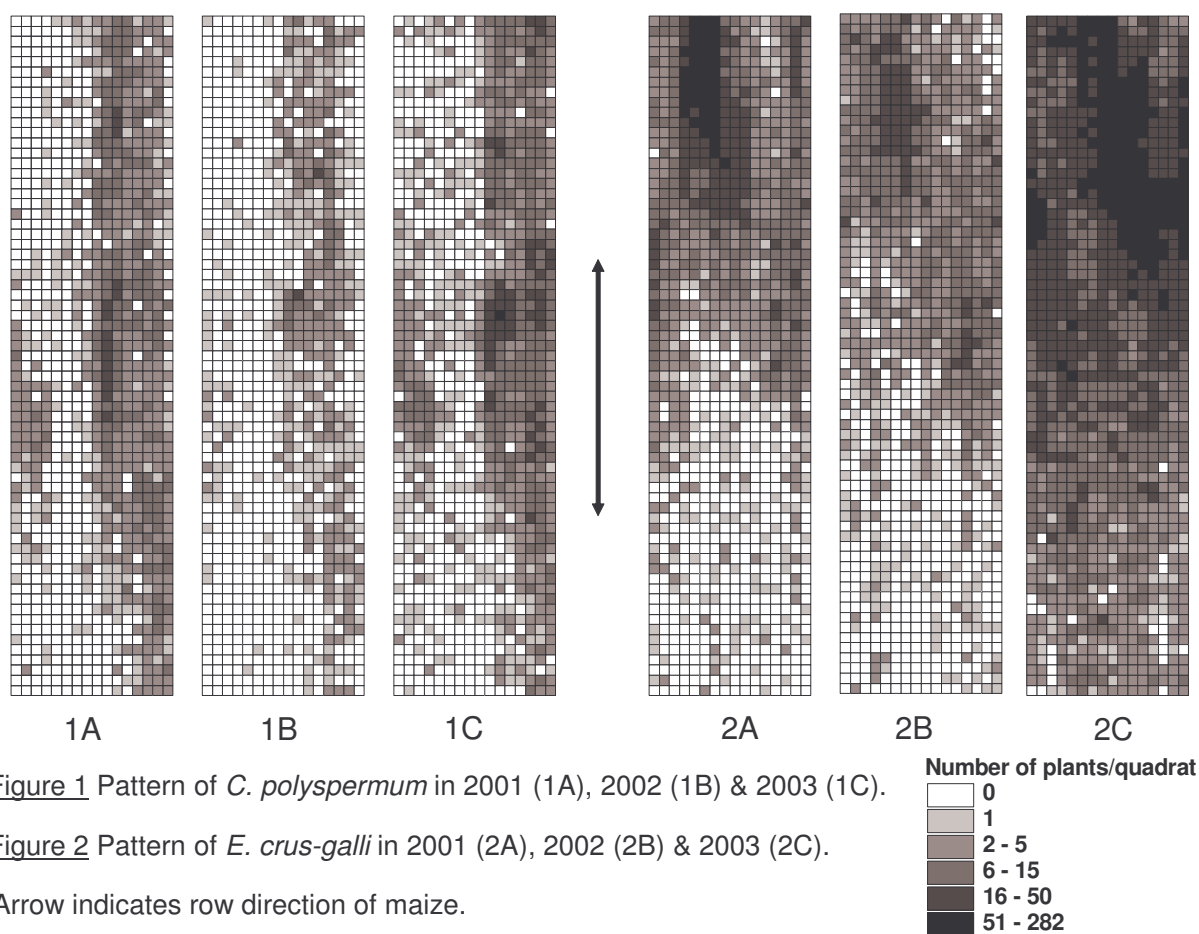


Figure 1 Pattern of *C. polyspermum* in 2001 (1A), 2002 (1B) & 2003 (1C).

Figure 2 Pattern of *E. crus-galli* in 2001 (2A), 2002 (2B) & 2003 (2C).

Arrow indicates row direction of maize.

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# Dosing of potato haulm killing herbicides with sensing techniques

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## Introduction

Prior to harvest, the haulm of most potato crops in the Netherlands is killed by specific herbicides (Bus et al., 2003; Kempenaar et al., 2004). Four herbicides differing in many properties are registered for potato haulm killing in the Netherlands today: glufosinate ammonium (Finale SL 14<sup>®</sup>), metoxuron (Purivel<sup>®</sup>), carfentrazone-ethyl (Spotlight 24 EC<sup>®</sup>) and diquat dibromide (Reglone<sup>®</sup>). Active ingredients are glufosinate ammonium, metoxuron, carfentrazone-ethyl and diquat dibromide, respectively. Diquat dibromide is the mostly used product by far. Concern about side effects of the potato haulm killing herbicides and interest to explore options to increase the net return of potato production initiated the study described in this paper. The aim of the study was to develop an innovative dosing system for potato haulm killing herbicides. Some results of the study are presented.

## Materials and methods

The concept of the innovative dosing system for potato haulm killing herbicides was derived from the MLHD system for chemical weed control (e.g. Kempenaar et al., 2002; ([www.mlhd.nl](http://www.mlhd.nl) contains an MLHD manual in English)). The innovative concept consists of the following elements:

1. Reflection measurements just before application of the herbicide
2. Decision on need to treat and dosing on the basis of reflection parameters
3. Fluorescence and reflection measurements 3 to 6 days after herbicide application
4. Decision on need to additionally treat and dosing on the basis of the post treatment fluorescence and/or reflection measurements.

Reflection measurements were done with Cropscan ([www.cropscan.com](http://www.cropscan.com)) or N-sensor ([www.sensoroffice.com](http://www.sensoroffice.com)). Fluorescence measurements were done with an EARS Plant Photosynthesis (PPM) meter ([www.ears.nl](http://www.ears.nl)). For details on the meters, the measurements and the experiments, see Kempenaar et al. (2004).

## Results and discussion

Figure 1 shows minimum effective doses in 15 different field trials with diquat dibromide in the Netherlands in the period 1999-2002. In each trial a titration of 4 doses (1, 2, 3 and 4 l/ha) was tested. On the x-axis of the Figures, experimental series numbers (trial numbers) are shown. The numbers are ranked in order of increasing value of the reflection parameter assessed with Cropscan. The higher the experimental series number, the higher the reflection parameter. The reflection parameter was higher when the canopy was greener and denser. There was a positive correlation between reflection parameter and minimum effective dose. Consequently, the doses recommended by the system are based on this correlation.

Figure 2 shows a good correlation between PPM value 6 days after treatment and efficacy 3 weeks after treatment. A PPM value of 20 is a threshold value that predicts good efficacy (comparable with MLHD system and photosynthesis inhibitors). Efficacy could also be predicted with reflection sensors (not presented).

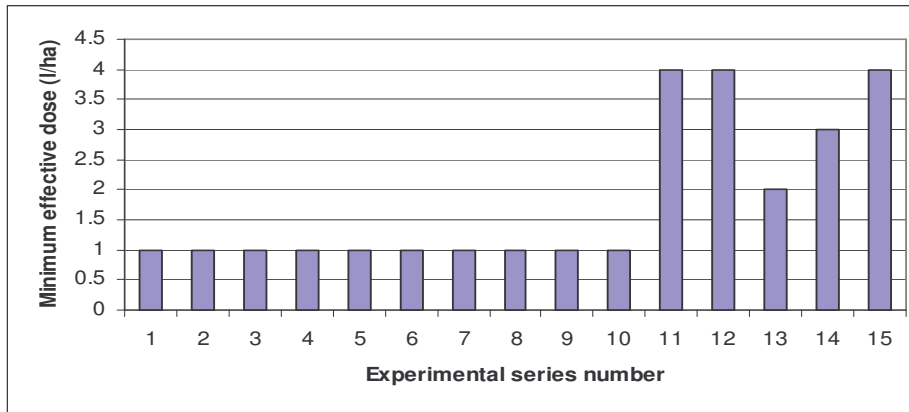


Figure 1. Minimum effective doses of diquat dibromide in trials in 1999 - 2002.

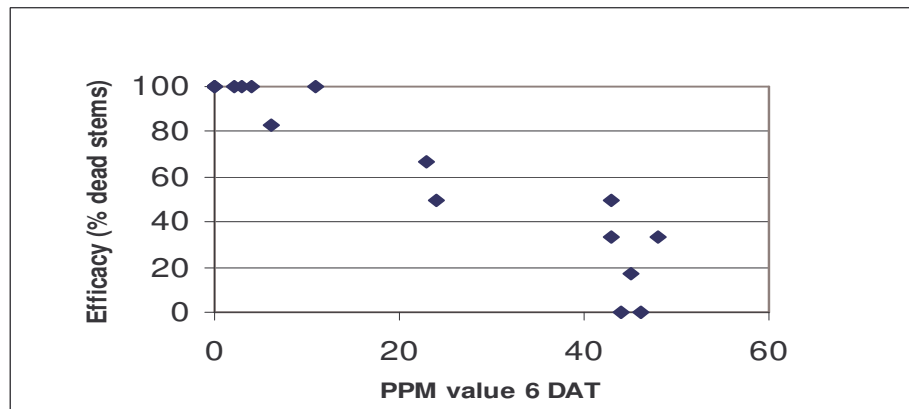


Figure 2 shows results of fluorescence (PPM) data from plots treated with diquat dibromide in 2002.

The innovative system was tested in 2003 and 2004 on commercial fields (Kempenaar, 2004; Kempenaar, in prep.). The tests showed that the sensing techniques supported the farmers to effectively reduce doses of potato haulm killing herbicides. Reductions were circa 30 % when the sensing techniques were applied at the level of spray strips in the crops. The challenge for the near future is to incorporate the sensing techniques at the through level of site specific management (within the spray strips). Farmers were most interested in the site specific management options that reflection measurements offer. Next steps in R&D of this work are:

1. Integration and testing of the reflection measurements on practical sprayers.
2. Assessment of spatial variation in potato crops.
3. Differentiation of dosing system to take into account resolution and specific conditions (e.g. disease pressure of *Phytophthora infestans*, weed infestation of the crop).

Field mapping of potato crops with commercial N-sensors in 20004 showed that there can be site specific variation in reflection parameter of potato crops to allow for site specific dosing of potato haulm killing herbicides.

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# Weed detection using chlorophyll fluorescence imaging and artificial neural network

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## Introduction

In most cases, weeds are not distributed in fields uniformly, but they are aggregated in patches (Wilson & Brain, 1991). A selective application of herbicides to the weed patches represents a considerable potential for reducing total pesticide quantity compared to a uniform application across the entire field. The realization of site-specific weed management in agricultural practice requires the solution of two problems: weed identification and sprayer control with high spatial precision.

At present, two systems are developed for automatic weed detection. The image analysis systems use CCD cameras and image analysis software to detect weed species composition and to discriminate weed from crop plants based on colour, shape and texture features (Gerhards et al., 2002). Optoelectronic sensors measure the reflectance of light in a certain range of wavelengths (Vrinds et al., 1999). Green leaves reflect the light in near-infrared wave band and absorb the light in the red wave band. The reflectance curve for soil is nearly constant. Both methods, however, have their limits that can be hardly overcome by technical progress, and allow only a specific use in practice.

Tyystjarvi et al. (1999) have proposed a new approach to automate weed identification using specific characteristic features in fluorescence kinetics of individual plant species. Chlorophyll fluorescence kinetics gives information enabling to distinguish accurately among individual species or species groups even under conditions of overlapping of crop and weed leaves.

Specific differences in fluorescence kinetics can be analysed using methods of artificial intelligence, such as neural networks.

## Materials and methods

### a) Chlorophyll fluorescence imaging measurement

The fluorescence imaging system FluorCam measures sequences of fluorescence images, which are recorded synchronously with measuring light flashes. Fluorescence emission is induced by two panels of super-bright orange light emitting diodes that provide measuring flashes. The photochemistry is driven by continuous actinic irradiance and by saturation pulses that are generated in a halogen lamp equipped with a shutter. The used measurement procedures (quenching analysis) allows quantifying the relative contribution of photochemical and nonphotochemical energy use together with description of fluorescence kinetics during Kautsky effect.

### b) Plant material

*Apera spica-venti*, *Galium aparine*, *Stellaria media*, *Tripleurospermum inodorum* and *Triticum aestivum* seedlings were grown in a glasshouse after their pregermination in Petri dishes until first leaves or cotyledons were fully developed.

In a second experiment, *Triticum aestivum*, *Brassica napus*, *Beta vulgaris* and *Helianthus annuus* plants were grown in pots together with *Galium aparine* plants under glasshouse conditions. The fluorescence measurements were made on intact plants in pots without dark adaptation under diffuse light conditions. At least 100 plants for each species were analysed.

## Results and discussion



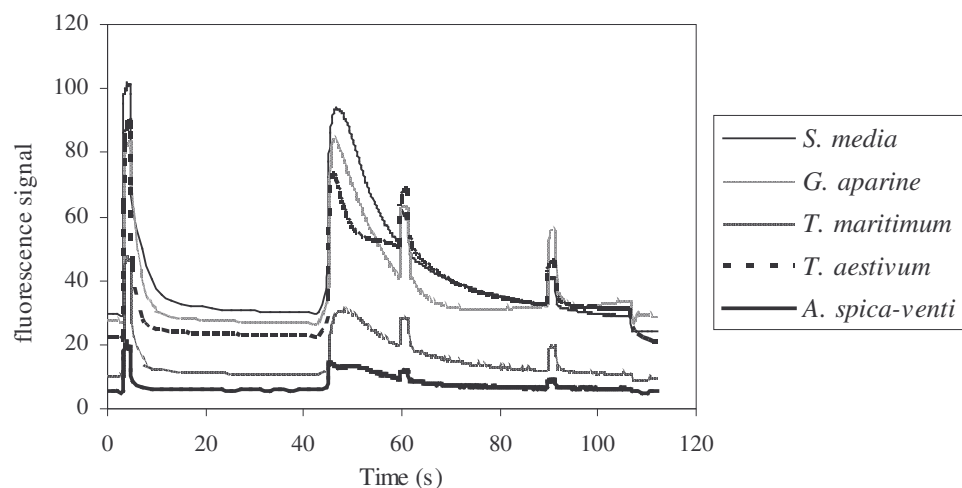
The measurements on *G. aparine* (representing weed species) and *T. aestivum*, *B. napus*, *B. vulgaris* and *H. annus* (as crop representatives) were made at two different growth stages: a) cotyledons (dicots) or first leaf stage (monocots) and b) 2-4 true leaves stage. The measurements shows high discrimination ability of the method at cotyledons (first leaf) stage with decreasing sensitivity by later measurement. The experiment with discrimination between different crop species and *G. aparine* shows comparable results using “Kautsky effect” and “Quenching analysis” measurement protocols. To improve the recognition accuracy, we used the artificial neural network classifier, trained on at least 100 plants. As input variables we used both common fluorescence parameters and newly created parameters extracted from fluorescence kinetics. The classification rate for discrimination between crops and *G. aparine* ranged between 90 and 100% and in almost all cases the correct discrimination rate was higher for short “Kautsky effect” measurement protocol. In the experiment with discrimination between individual weed species, the correct discrimination rate ranged between 85 and 100%. The neural network analysis represents improvement of discrimination accuracy in comparison with discriminant analysis where correct classification rate ranged from 78 to 99%.

Fluorescence imaging combined with artificial neural network represents a feasible alternative in the development of automated weed recognition methods which can be well supplied with plant shape features easily extracted from fluorescence images on the basis of high contrast between green leaves and soil background.

Classification table for neural network (RBF 29:29-258-5:1) recognition

Classification	<i>G. aparine</i>	<i>A. spica-venti</i>	<i>T. inodorum</i>	<i>S. media</i>	<i>T. aestivum</i>
Total	235	199	192	472	250
Correct	235	187	165	451	217
Correct (%)	100.00	93.97	85.94	95.55	86.80

Figure 1. Average fluorescence kinetics for individual species



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# Discrimination of grassweeds in winter cereal crops using hyperspectral data

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## Introduction

From an economical and environmental point of view, the use of herbicides on specific areas within a field rather than on the entire field, as traditionally applied, is an important concern. Maps of weed populations are required to support site-specific treatments. Traditional sampling techniques are not cost-effective when the required intensive sampling has to be applied. Remote sensing techniques could be an alternative to producing spatial data on weed distribution. Various research projects are currently evaluating ground-based and aerial remote sensing techniques for weed detection and mapping (Gibson *et al.*, 2004; Koger *et al.*, 2004). This paper studies the spectral differences between weed and crops at different phenological stages and at different levels of environmental noise (soil, illumination, etc). This analysis is a critical phase prior to assessing the potential use of satellite data in this field of research.

## Materials and methods

Laboratory and field experiments were carried out during 2003 at both the “La Poveda” research farm (SE Madrid) and the Department of Geography (University of Alcalá) in order to analyze the spectral separability between two grass weeds (*Avena sterilis* and *Lolium rigidum*) and two winter cereals (wheat and barley). Reflectance measurements were collected by use of a GER2600 spectroradiometer with 640 bands ranging from 350 to 2500 nm.

The laboratory experiments were conducted to explore the spectral behaviour of weeds and crops under controlled conditions regarding illumination and vegetation ground cover. The aim was to determine the existence of spectral differences between the two plant species by minimizing external factors. Four laboratory measurements were taken (from January to April), according to cereal phenological stages. Measurements were carried out by use of plant and leaf level (Figure 1) requirements. In the first case, four measurements were taken directly over the potted plants, which had been dug from the ground and brought from the same field where the exterior experiments were conducted. A leaf level measurement was made once the plant leaves had been collected and arranged over a flat and black tray. Two spectral measurements per species were taken, ensuring maximum speed in the overall procedures to minimize leaf deterioration.

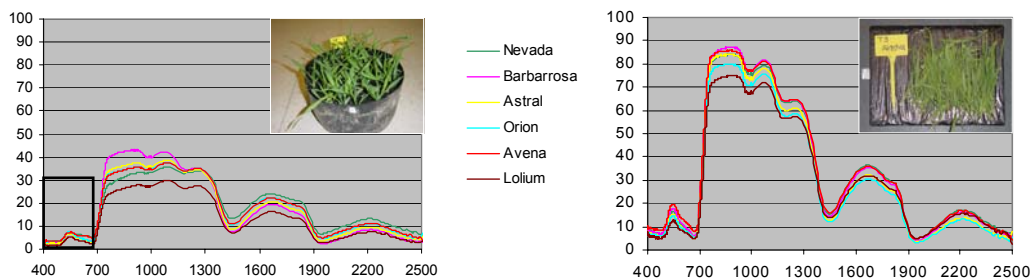


Figure 1. Laboratory measurements. Spectral curves for all the species (13/03/2003). Left: plant level; right: leaf level

The field experiment site consisted of two rows of 6 × 3 m plots, where one plot per species and row (plus two additional plots of bare soil) were randomly distributed. Field measurements were taken at five different dates from January to May. The aim was to analyze the spectral separability between weeds and crops under field conditions (which are more similar to the satellite image data) including the potential noise, mainly illumination and atmospheric variability, from external factors. Within each plot, two measurements were taken on two different FOVs, always between 11 and 2 p.m.

Both in laboratory and in field experiments, a series of biophysical variables were measured simultaneously to investigate their relationship with the spectral behaviour of the species. Data collection included Leaf area index (LAI), plant water content, chlorophyll content (using a Minolta SPAD 512 instrument) and percentage of green cover.

### Results and discussion

Qualitative and quantitative methods were used to analyze the spectral separability between the weeds and crops species. The quantitative analysis was based on statistical methods such as normalized distance and discriminant analysis. Those methods were applied on both reflectance values and spectral indices (TCARI, OSAVI, etc). The aim was to determine the band/s or the band combinations that showed the highest spectral separability between crops and weeds, and which could therefore be considered the most appropriate for weed discrimination. Results from the discriminant analysis by use of field measurements (Table 1) show that the spectral differences between weeds and crops were insufficient during the first phenological stages due to the predominance of background noise from the soil. However, for the third date (end of March), the discriminability improves significantly (Figure 1), and the most frequently selected bands are in the red and NIR.

Table 1. Hyperspectral bands selected in the discriminant analysis

Measurement date	Selected bands	
January	None	
March	None	
March (end)	470-480	Blue
	630-640; 650-660	Red
	710-720; 720-730; 750-760	NIR
	1500-520	SWIR
April	640-650; 690-700	Red
	760-770; 840-850	NIR
June	980-990; 1060-1080	SWIR

Results showed that the best discrimination was obtained a late tillage stage. The percentage of cases that were well classified at a spectral window between 630 to 760 nm, which corresponds to the ‘red edge’ portion of the spectrum, varied from 75 to 100 %. The best index to discriminate grass species based on hyperspectral data was the ratio TCARI/OSAVI (Transformed Chlorophyll Absorption in Reflectance Index/Optimized Soil-Adjusted Vegetation Index). This index has shown to be very sensitive to chlorophyll content variations and very resistant to changes in LAI and illumination. Multispectral data proved satisfactory results in some cases, but they proved less adequate than hyperspectral information. However, *Lolium rigidum* was correctly identified in most cases by multispectral measurements in the jointing-heading stages of growth. This information could be very useful in the development of new and existing dedicated remote sensing systems and in the improvement of methods to discriminate weeds using current airborne and satellite systems

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# Long term investigations in precision weed control

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## Introduction

All weed infestations have an aggregated distribution at one or more scales. Weeds are never distributed uniformly within a field (Nordmeyer *et al.* 2003). Precision farming in weed control takes into account this heterogeneity and offers a powerful set of tools to reduce herbicide use. Additionally, economical and ecological benefits are obvious. Therefore a greater social acceptance of farming practices is to be expected. In general, chemical weed control in agricultural practice does not take into account the spatial distribution of weeds. Normally, farmers make a decision and select a herbicide or tank mixture for the whole field according to the main weeds. Consequently, field areas will be treated with herbicides unnecessarily based on local weed occurrence.

## Materials and methods

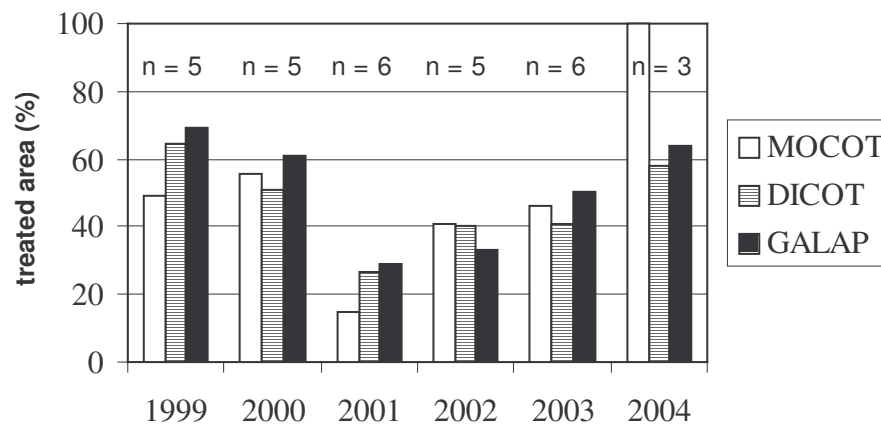
Site specific weed control in winter wheat was carried out for several years in the northern part of Germany (Braunschweig region). Weed densities (2 samples of 0,1 m<sup>2</sup> per grid point) were counted in an irregular grid (spacing 25 x 36 m) by field-walking using a Differential Global Positioning System (DGPS) for point location (Nordmeyer *et al.* 1997).

Weed estimations were done shortly before herbicide applications in spring. Weed distribution maps were created for single weed species (*Galium aparine* = GALAP) and groups of weed species (monocotyledonous weeds = MOCOT, dicotyledonous weeds = DICOT [without *G. aparine*]). Geostatistical methods and kriging interpolation were used to estimate weed densities at unknown locations. Based on the weed maps and threshold levels, herbicide application maps were created and herbicide application was done according to the spray / no spray concept. Areas with weed densities above a threshold level were treated with herbicides. Details on site specific herbicide application are described by Nordmeyer & Häusler (2000). The general threshold levels were 30 plants/m<sup>2</sup> for MOCOT, 40 plants/m<sup>2</sup> for DICOT and 0.2 plants/m<sup>2</sup> for *G. aparine*.

## Results and discussion

A heterogeneous weed distribution with weed patches and weed free areas could be observed for single weed species and groups of species within all fields and in all years. Obviously, site specific weed control reduced herbicide use on agricultural fields. Mean values varied from year to year according to weed biology and development, local environmental conditions and agricultural practices. For a 6-year period the percentage of herbicide treated areas is presented in Figure 1. The field portions to be treated with herbicides for single weed species e.g. *G. aparine*, or groups of species (monocotyledonous and dicotyledonous weeds) varied every year in a wide range. In 1999, an average 60.2% of the field area considered was sprayed against dicotyledonous weeds, 46.2% against monocotyledonous weeds and 66.5% against *G. aparine*. In 2000, the values were very similar: 49.8% of the area was sprayed against dicotyledonous weeds, 51.0% against monocotyledonous weeds and 52.4% against *G. aparine*. In the year 2001 the herbicide treated area for the 3 weed groups was less than 30%.

Figure 1. Herbicide treated areas in winter cereals (1999-2004). Mean values (n = number of fields) of monocotyledonous (MOCOT), dicotyledonous (DICOT) weeds and *Galium aparine* (GALAP).



In general, the site specific weed control resulted in a reduction in herbicide use on all fields. As a consequence of limited field travelling, it may also contribute to a reduction of working hours and machine work for the herbicide application. A problem with site specific weed control using the mapping concept is the time consuming process of weed monitoring by field-walking. The costs of manual identifying and counting the weeds are too expensive for agriculture practice. In future, it will be necessary to have automatic weed detection systems and an appropriate spraying technique.

Site specific weed control represents a reduction strategy for herbicide use. In the recent discussion about consumer protection, quality management in agriculture, environmental tolerance and transparency in the production process this type of weed control will have high future prospects.

### Acknowledgements

This project was funded by the German Volkswagen Foundation.

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# Using digital image analysis for automatic weed identification and weed mapping in the field

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## Introduction

Spatial and temporal heterogeneity of weeds in agricultural fields is taken into account for GPS-controlled patch spraying in Precision Farming systems. For automatic sampling, optoelectronic sensors were used to measure the reflectance in the green, red and near-infrared light wave bands. Plant density information was used for a real-time non-selective herbicide application (Biller 1998). Gerhards et al. (1998) and Sökefeld et al. (2000) used digital image analysis systems to identify plant species based on characteristic shape features. An automatic weed species identification system in arable fields allows site-specific application of selective herbicides.

## Materials and methods

For image acquisition in the field, three digital bi-spectral (BS) cameras (Gerhards *et al.* 2002) were mounted in the front of the spray boom. Differential images (NIR-VIS) with BS cameras were taken in 180 hectares of winter wheat, winter barley, spring barley, sugar beet, winter rape and maize. The cameras, with a resolution of 636 \* 480 Pixel, took well focused images with a strong contrast between green plants and soil, mulch and stones of an area between 0,2 m<sup>2</sup> and 0,02 m<sup>2</sup> per image at a speed of 5-8 km/h. Approximately 3000 images/ha were taken and stored together with their GPS coordinates on an on-board computer.

In addition to that, images from 50 different weed species were taken with an Infrared (IR) camera (1024 \* 1024 Pixel) during different development stages (BBCH 10 up to BBCH 14). Image analysis software (Sökefeld et al. 2000) was used to identify characteristic shape features of crops, weed species (IR images) or classes of several weed species with similar sensitivity to herbicides (BS images) and stored in a database. Those shape feature including area, compactness, the quotient of minimum Ferrets diameter and maximum Ferrets diameter and Fourier descriptors of the transformed contour, were analyzed with the SAS-Statistic Software for normal distribution (Shapiro-Wilk Test with  $p > 0,05$ ) and significant difference (Student-Newman-Keuls Test) for differentiation. The results were used to determine the best decision algorithm for a fuzzy classification procedure. Weed distribution maps were then created from analysed images and compared to maps from visual grid sampling and linear triangulation interpolation.

## Results and discussion

Shape parameters of most crops and weed species differed significantly between species and growth stages. Broad-leaved weeds including *Daucus carota*, *Galium aparine* and *Chenopodium album* showed different shape parameters in all growth stages tested. Invasive species including *Abutilon theophrasti* and competitive species like *Galium aparine* were significantly different from other broad-leaved weeds, grass weeds and crops. Grass weeds including *Alopecurus myosuroides*, *Apera spica-venti* and *Avena fatua* showed similar shape parameters in BBCH 10 and BBCH 11 but were different from wheat and barley.

*Bromus sterilis* and *B. secalinus* could be differentiated from other grass weeds and cereals, but not from each other when they occurred in the same growth stage.



Shape parameters of weeds and crops obtained from the BS images were classified in four classes including crop, grass weeds, broad-leaved weed species and *G. aparine* or *A. theophrasti*. Automatic classification of 14.000 images in a maize field resulted in 72 % correct identification (Figure 1).

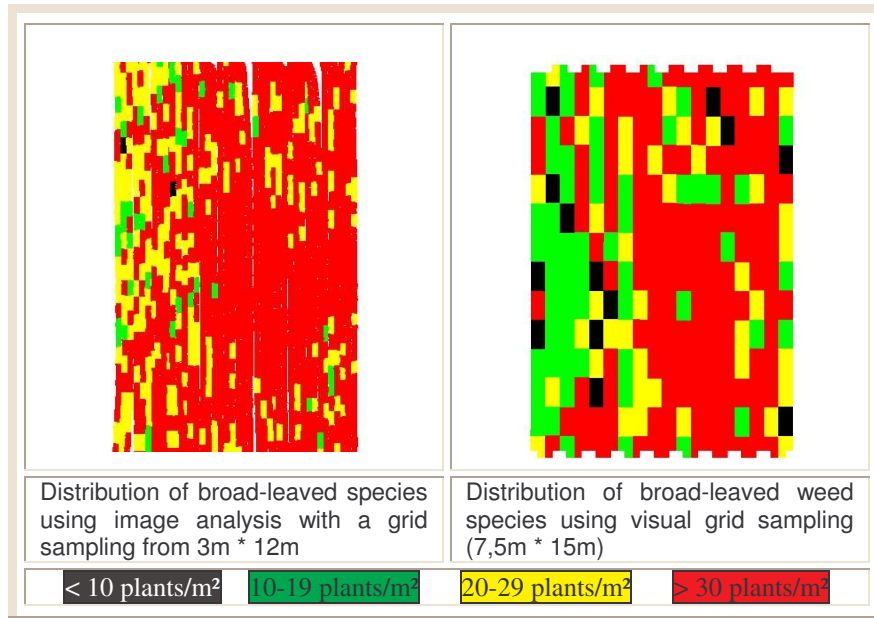


Fig. 1: Distribution maps for broad-leaved weed species on a 2.4 ha maize field at Dikopshof Research Station of the University of Bonn; sampling was performed on 2004-05-21.

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# ***Phalaris brachystachys* biotypes: resistance to fenoxaprop-p-ethyl and study of morphological and phenological characteristics**

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## **Introduction**

The increase in the use of graminicides, which inhibit acetyl coenzyme A carboxylase (ACCase), led to a parallel increase in the evolution of resistant populations to these herbicides. Graminicide resistant grasses are of economic importance globally because of the large acreage infested and the limited number of herbicides available for their control. *Phalaris brachystachys* is a major grass weed infesting wheat in Greece. It has been reported, that in certain areas of Northern and Central Greece, fenoxaprop-p-ethyl fails in effective control of *P. brachystachys* after 8-year repeated use. Development of resistant biotypes may account for reduced fenoxaprop-p-ethyl efficacy.

The objective of this study is first to assess the resistance and second to study several morphological and phenological characteristics of *P. brachystachys* biotypes in order to investigate any possible relation between resistance and these characteristics.

## **Materials and methods**

**Biological material:** *P. brachystachys* seeds were collected in 2000 from seven infested winter wheat fields (biotypes A,B,C,D,E, F,G) of Northern and Central Greece, where fenoxaprop-p-ethyl was repeatedly used for more than 8 years. Seeds from a field near Serres (biotype H) in Northern Greece that had never been treated with fenoxaprop-p-ethyl were also connected. All seeds were kept refrigerated at 4°C in dry storage until used.

**Pot experiments:** *P. brachystachys* seeds pre-germinated in petri-dish were planted in 200 ml plastic pots containing sandy soil and peat mixture (2/1/v/v) and 1g kg<sup>-1</sup> of a slow release fertiliser. The pots were irrigated and kept in a net house. There were 2 plants per pot. When plants were at the three- to four- leaf stage of development they were treated with five different dosages (Table 1) of fenoxaprop-p-ethyl, clodinafop-propargyl and tralkoxydim using a motorised sprayer equipped with a flat- fan nozzle (8001E) calibrated to deliver 300 l ha<sup>-1</sup> at 245 kpa. Treated plants were returned to the net house and arranged in a completely randomised design with four replications. Two weeks after treatment, plants were cut at the soil surface and fresh weight per pot was determined.

In another pot experiment, morphological and phenological characteristics (plant height, number of spikes, length and number of roots, number of heads, date of flowering and maturing) of the aforementioned biotypes were studied, in order to investigate any possible relation between resistance and these characteristics.

Table 1. Dosages of herbicides.

Pot experiments					
Treatment	Rate (g ai/ha)				
fenoxaprop-p-ethyl	0,55	110	220	440	880
clodinafop-propargyl	0,20	40	80	160	320
tralkoxydim	0,150	300	600	1200	2400

## **Results and discussion**

Results of the first experiment showed that all biotypes used apart from the control were found to be resistant to fenoxaprop-p-ethyl (Figure 1); however, all of them were sensitive to

clodinafop-propargyl and tralkoxydim. So practically, control of resistant *P. brachystachys* biotypes can be achieved by alternative selective herbicides.

Results of the second experiment showed, that the sensitive biotype significantly differed from the others to almost all characteristics studied (except number of roots), which could permit its rapid identification in resistance research.

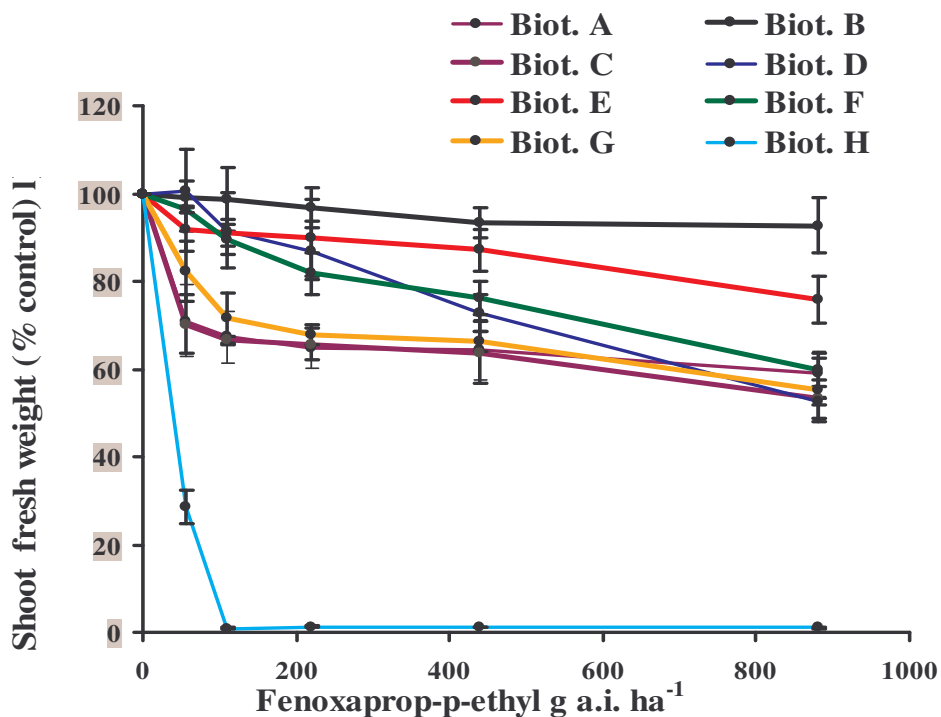


Figure 1. Effect of fenoxaprop-p-ethyl on the shoot fresh weight of susceptible (Biot.H) and resistant *P. brachystachys* biotypes. Vertical bars represent the standard error of the mean.

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# Management of herbicide resistant annual ryegrass, *Lolium rigidum* Gaudin, in cereals

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## Introduction

Cereals monoculture and repeated use of ACCase inhibiting herbicides have caused the appearance of populations of resistant *Lolium rigidum* to the aforementioned herbicide families, in the sub humid zones in the northeast of Spain (Taberner *et al.* 2001). The continued emphasis on herbicide application for weed control will increase this problem. The aim of this study is to evaluate the effectiveness of different primary tillages to control populations of *L. rigidum* through two years in winter barley fields.

## Materials and methods

The experiments were carried out in winter barley fields during 2002-2003 and 2003-2004 in Apies (Huesca). The soil type of the area is a calcisol with low content of organic matter. Previously a seed-bioassay was made, which confirmed the resistance of these populations to an ACCase inhibiting herbicide (diclofop methyl). The field experiments consisted of four treatments (namely A, B, C and D) with three replicates at random, in order to evaluate the effectiveness of different previous tillage prior to the preparation of the sowing bed (Gill & Holmes, 1997). The variables used in this study were several combinations of ploughing at different times of the year and vertical tillage (subsoiler).

Table 1. The main data of tillage systems.

Treatment	Month of ploughing		Month of vertical tillage	
	2002-2003	2003-2004	2002-2003	2003-2004
A	-	-	October	January
B	-	January	October	
C	-	July	October	
D	October	January	-	-

In all cases two evaluations of the *Lolium* density were made. One was made before tillage and the other one before harvesting. *Lolium* plants were counted throwing at random 6 hoops of 0,1 m<sup>2</sup> per plot. In the plot A ploughing tillage was not carried out. In the year 2003-2004 the sowing was delayed due to excessive rain.

## Results and discussion

In Figure 1, the evolution of the infestation throughout two years for each treatment is shown. During the first year, the vertical tillage made in A, B and C did not result in decreasing *Lolium* density, nevertheless a significant decrease occurred in treatment D (ploughing tillage). In treatments A, B and C a high plants density was maintained while the population lowered slightly in the ploughing tillage plots.

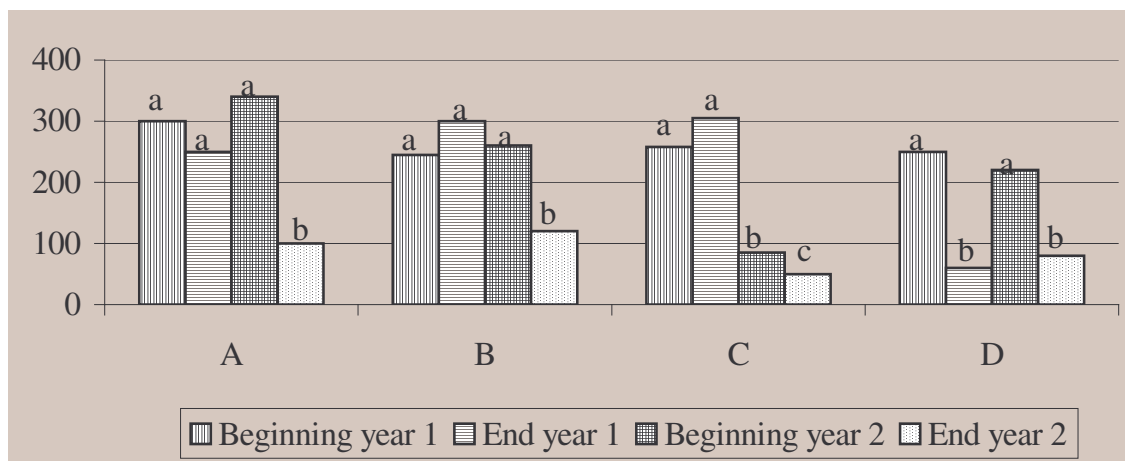


Figure 1. Treatment means of ryegrass plant density (plants/m<sup>2</sup>).

For the second year, a decrease of the *Lolium* density was observed in the plots with vertical tillage, which was due mainly to the delay in seeding. Ploughing tillage before seedtime (treatment B) diminished the initial density, but it was not sufficiently effective. Due to the fact that the seeds were buried just after falling to the ground during the harvesting, a significant reduction of the initial *Lolium* population happened. In plots D, where mouldboard ploughing took place twice, there was a decrease in infestation because of both the tillage and possibly a lower initial bank of seeds, due to the population reduction during the previous year.

Ploughing tillage reduces the *Lolium* population up to a point but its efficiency seems to depend on the infestation level (bank of seeds) and on the timing of the tillage as well. Tillage just after the harvest, in spite of being an obsolete method, can be effective in cases like this one, where resistant populations have a high density. On the contrary, if the tillage is made before seeding the effectiveness diminishes. The ploughing tillage repetition does not increase the efficiency, because of the possibility that some viable seeds, which have been buried deep into the ground, get an adequate depth for their germination.

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# Quick-tests for detection weed resistance to sulfonylurea and triazine herbicides – preliminary results

I. M. Calha, A. Matias, A. C. Alves, L. Mora, F. Rocha

## Introduction

Sulfonylurea and s-triazine resistant weed biotypes were confirmed in Portuguese rice and maize fields, respectively (Calha et al., 1995; Calha & Rocha, 2003). A large survey on herbicide weed resistance on main crops: vineyard, maize, rice tomato and potato was performed (Rocha, 2002). There is a need for quick-tests to detect bensulfuron-methyl and atrazine resistant biotypes for routine purposes in large number of samples. Whole plant bioassays are the most used methods since they best mimic field conditions. However they are labour and time consuming (Moss, 1995). For resistance screening a single dose assay should be preferable to analyse huge number of samples. The choice of the discriminant dose is crucial to avoid misunderstand results. This paper presents preliminary results of two methods to reduce response time on whole plant assays: seed soaking method, and chlorophyll fluorescence for herbicide resistance confirmation to atrazine and bensulfuron-methyl.

## Material and methods

**Plant material:** Resistant biotypes of *Alisma plantago-aquatica* L. and *Chenopodium album* L. came from fields where bensulfuron-methyl and atrazine were sprayed for 6-8 consecutive years, respectively. A susceptible population came from neighbour fields which had never been treated with those herbicides. Pre-chilled seeds were used for all tests: 30 days in KNO<sub>3</sub> solution (0.2%) at 4° C in the darkness for *A. plantago-aquatica* seeds (Munscher, 1936) and 2 days in moist sand at 4°C in the darkness for *C. album*.

**Seed soaking method:** 60 to 130 *C. album* seeds were soaked for 12h in 10 ml atrazine solution (0.3–30 000 ppm) inside a growth chamber with alternating conditions of light and temperature (15°C/25°C). After incubation seeds were transferred to soil: peat (2:1) substrate inside a greenhouse. When plants reached the 6-leaf stage *C. album* were sprayed with 7500 g ha<sup>-1</sup> atrazine. Fresh weight was assessed 21 days after treatment (Hashem et al., 2002).

**Chlorophyll fluorescence assay:** Procedure was identical from previous stated except the step of seed soaking. Plants of *A. plantago-aquatica* at the 6-leaf stage were sprayed with 30 to 120 g ha<sup>-1</sup> bensulfuron-methyl. 72 hours after spraying, detached leaves were dark adapted for 1 h and chlorophyll fluorescence induction curves were measured with a Plant Efficiency Analyser (PEA; Hansatech), with recording time of 5-10 s and an excitation light intensity of 2700 μmol photons m<sup>-2</sup> s<sup>-1</sup>. The area above induction curve was measured (Moss, 1995). The experiments were in a complete randomized design, with six replications and experiment was repeated at least twice

## Results and discussion

**Seed soaking method:** Four weeks after seeding, atrazine reduced *C. album* seedling survivorship in the S biotype to near zero at 30000 ppm. In the R biotype many seedlings survived the seed soaking treatments at the same atrazine dose. Subsequent foliar spray of atrazine (7500 g ha<sup>-1</sup>) did not kill the surviving seedlings of population R indicating that the seed soaking was effective in differentiating the S and R biotype (Figure 1a).

**Chlorophyll fluorescence:** 72 hours after application photosynthesis was inhibited by 120 g ha<sup>-1</sup> bensulfuron-methyl for the S biotype of *A. plantago-aquatica*. R biotype was not affected by any dose studied (Figure 1b). Leaf incubation on bensulfuron-methyl solution (3000 ppm) for 24h also inhibits photosynthesis of S plants (unpublished data). Similar results were obtained in barley and flax, after 24h incubation on imazamethabenz (Percival & Baker, 1991) or after 2h incubation on imazaquin (Judy et al., 1990). With these techniques it was possible to



confirm resistance in a shorter time period compared to conventional whole plant assays which took 7-8 weeks for testing resistance to atrazine in *C. album*, and 11-12 weeks for testing resistance to bensulfuron-methyl in *A. plantago-aquatica*. Chlorophyll fluorescence as an indirect approach to assess the effect of ALS inhibiting herbicides on photosynthesis might be a promising low cost and quick test for evaluation of bensulfuron-methyl resistance.

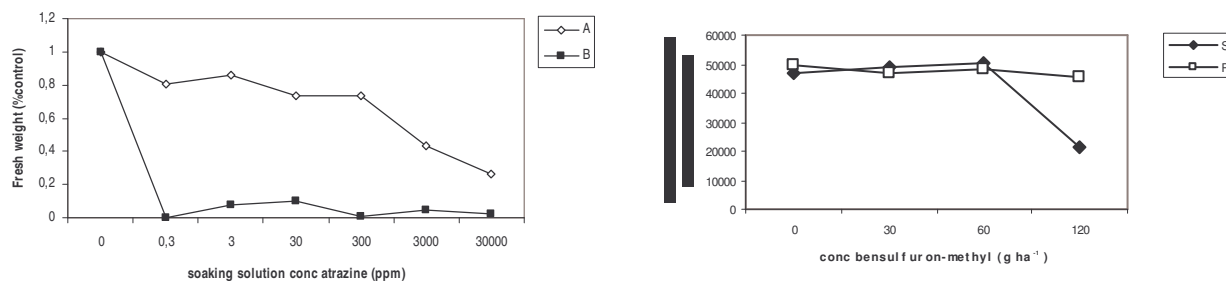


Figure 1. Dose response curve for a) *Chenopodium album* resistant (A) and susceptible (B) biotypes after a 12 h period of seed soaking on atrazine solutions and for b) chlorophyll fluorescence assessed 72h after bensulfuron-methyl application to *Alisma plantago-aquatica* R and S biotypes

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# Long-distance wind dispersal of *Conyza canadensis* and management implications

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## Introduction

The development and rapid adoption of glyphosate-resistant crops by farmers during the last decade has caused North American agriculture to become increasingly dependent on the herbicide glyphosate (most common trade name Roundup). Additionally, glyphosate-resistant crops have promoted the use of no-tillage, leading to the increase in many winter annual weeds, including *Conyza canadensis*. Spread of glyphosate-resistant *C. canadensis*, threatens the sustainability of no-tillage, low-input agricultural systems that utilise glyphosate. Since, glyphosate-resistant *C. canadensis* was first confirmed in 2000 in Delaware (VanGessel, 2001), resistant populations have been confirmed in 11 states and cover greater than 40,000 hectares (Heap, 2004). In order to develop effective resistance management plans, it is necessary to first understand the dynamics underlying spread of this invasive biotype. For a wind dispersed species, this includes both local and long-distance dispersal (LDD). Therefore, the objectives of this research were to quantify 1-dimensional and 2-dimensional *C. canadensis* seed dispersal patterns and the mechanisms that give rise to such patterns.

## Materials and methods

Field experiments were conducted in 2003 in Pennsylvania in two 700 m by 150 m soybean fields. Two glyphosate-susceptible *C. canadensis* source populations were established; each source patch area was 28 m<sup>2</sup> with weed densities of 17 and 45 plants m<sup>-2</sup>. The sampling design consisted of seed traps placed at 10 m intervals along transects oriented at angles of 0°, 22.5°, 45°, 90°, 135°, 180°, 225°, 270°, 315°, and 337.5° aligned in the prevailing wind direction (PWD). Transect length varied from 70 m to 200 m in the PWD. In the PWD, arcs with the same sign interval, covered 22.5° with arcs located every 50 m from 200 m to 500 m. Six, 8 to 10 day long trials were conducted during the entire seed dispersal period of 15 August to 6 October, 2003. A one-dimensional analysis examined dispersal in the PWD and data were restricted to these traps only. Three empirical models (negative exponential, inverse power and mixed (Bullock & Clark 2000) were fitted to the data. One mechanistic model, with parameters collected independently, was constructed and compared to the empirical models. For the 2-dimensional analysis, traps in the arcs and at angles 22.5° and 337.5°, were excluded. Seed direction weighted by distance and wind direction weighted by speed for each trial were correlated using circular statistical methods.

## Results and discussion

Seed production was very high in both sources (approximately 70 and 180 million seeds released). Seeds were collected at distances up to 500 m from the source in both fields, but most seed were concentrated near the source. All three empirical models fit the data well near the source but underestimated LDD events (See Table 1). The inverse power and mixed models fit the data significantly better than the negative exponential ( $p < 0.05$ ). The mechanistic model overestimated seed deposition near the source and provided comparable estimates of LDD events. None of the models were robust enough to capture the second mode located at approximately 450 m from the source population. The second mode is a phenomenon that may be the result of wind updrafts at the time of seed release leading to longer distance movement (Tackenberg et al., 2003). Two-dimensional spatial seed distributions appeared to reflect the PWD. Wind velocity and seed distributions were

approximately equal but statistically different in many trials (10 of 12) in both fields. While these experiments used a shorter time increment than many seed dispersal trials, the scale of wind/seed interaction is still smaller. Evaluation of seed movement over minutes and hours is a more appropriate scale and will lead to a better correlation between wind direction and seed distribution.

The long dispersal distances observed and modelled in this research indicate *C. canadensis* can easily move out of a single field and into adjacent fields and farms. Predicting how seeds move across a complex landscape is crucial to preventing continued spread of this resistant biotype. This work provides foundational data for a spatial spread model to assess movement across a heterogeneous landscape and provide realistic management options for farmers to slow the rate of spread. Management options to slow the spread must include recognition of the dispersal potential and history of resistance of *C. canadensis*. These results call into question the scale of current management practices. While area-wide weed management is not commonly practiced when managing weeds, efforts to slow the spread of glyphosate-resistant *C. canadensis* may now require landscape scale adoption of practices that minimise the reproductive success of this plant.

Table 1. Cumulative seed collected at a subset of distances with the predicted values from three empirical models and one mechanistic model. Values represent predicted seed collected and starred values (\*) represent values of less than 0.1.

Distance (m)	Seed Collected		Negative exponential		Inverse power		Mixed		Mechanistic	
	P1	P2	P1	P2	P1	P2	P1	P2	P1	P2
Source	29756	66452	-	-	-	-	-	-	-	-
10	1061	2067	747	2145	1055	2634	1055	2634	12900	27205
20	195	484	363	857	196	476	209	476	318	893
30	80	179	176	341	75	179	67	179	36	121
40	37	135	84	135	38	90	32	90	7	29
50	19	62	40	53	23	54	20	54	2	9
100	4	7	1	0.5	5	11	5	11	0.1	0.3
200	1	3	*	*	1	2	1	2	*	*
300	0	0	*	*	0.6	1	1	1	*	*
400	3	0	*	*	0.4	0.8	0.6	0.8	*	*
500	2	2	*	*	0.2	0.5	0.5	0.5	*	*
<b>AIC</b>			1025.2	910.0	121.9	141.8	121.8	145.8		

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# Evolution of resistance to herbicides inhibiting acetyl-CoA carboxylase in French black-grass (*Alopecurus myosuroides* Huds.) populations

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## Introduction

Environment-driven, positive selection leads to the fixation of adaptive mutations in populations. In agricultural fields, herbicides represent the most powerful selective pressure exerted upon weed populations. Weeds adapt by developing resistance mechanisms. We considered the selection exerted since the end of the 1980s by herbicides targeting the chloroplastic acetyl-coenzyme A carboxylase (ACCase) in black-grass, a major weed in winter crops. Mutant ACCase alleles with reduced sensitivity to herbicides were identified in black-grass populations (Délye et al. 2002; 2003; in press), suggesting that strong selection pressure can drive fast adaptation of weed populations (Délye et al. 2004a). The purpose of this work was to use molecular genetics to understand how target-based resistance to ACCase inhibitors appeared and spread within and between black-grass populations in France.

## Materials and methods

We analysed 116 black-grass populations collected in 2000, which represented all areas where winter cereals are grown in France. To determine the mode of appearance of ACCase-based resistance, we examined nucleotide variation in ACCase within and between 18 out of the 116 black-grass populations. For each population, the occurrence of resistance to four ACCase-inhibiting herbicides (fenoxaprop, clodinafop, haloxyfop and cycloxydim) was assessed by analysing 100 plants per herbicide using seed bioassay (Letouzé and Gasquez 1999). The frequency of each of the five mutations within ACCase that confer resistance to herbicides (Délye et al. 2002, 2003, 2005) was assessed by genotyping 50 plants per population. Knowing the cross-resistance pattern associated with each ACCase mutation (Table 1) and the frequencies of the mutations within each population, it was possible to compute for each of the four herbicides used the proportion of resistant plants that would occur in a population if the five ACCase mutations explained 100% of the resistance. These theoretical proportions were compared with the real ones, obtained from herbicide bioassays.

Table 1. Cross-resistance patterns associated with five point mutations within the gene encoding black-grass ACCase (R, resistant, S, sensitive).

Mutation	Herbicides			
	Fenoxaprop	Clodinafop	Haloxyfop	Cycloxydim
Ile-1781-Leu	R	S	S	R
Trp-2027-Cys	R	R	R	S
Ile-2041-Asn	R	R	R	S
Asp-2078-Gly	R	R	R	R
Gly-2096-Ala	R	R	R	S

## Results and discussion

*Mode of appearance of ACCase-based resistance: multiple origins.* We analysed a total of 172 ACCase sequences using a population genetics approach (Délye et al. 2004a). They comprised 72 haplotypes, 49 of which did not confer resistance and served for comparison. At least two out of the five mutations were proved to have evolved from multiple, independent origins in the populations investigated (Délye et al., 2004b). Conversely, different mutations occurred in a given population, and even in a given plant (Délye et al. 2004b).

*Distribution of resistant ACCase alleles in France: a mosaic.* Among the 116 populations genotyped for the presence of the five mutations within ACCase, 77 contained at least one mutant ACCase allele. Among these 77 populations, 44 contained one of the five mutations, 24 contained two mutations, five contained three mutations, three contained four mutations and one contained all five mutations. There was no geographical organisation of the distribution of mutations at the level of the area sampled. The distribution of ACCase alleles between black-grass populations was rather like a mosaic. Adjacent populations could display strikingly different contents in ACCase mutations.

*Resistance to some ACCase-inhibiting herbicides is not solely due to ACCase-based resistance.* Comparison of theoretical and real proportions of resistant plants was performed for all four herbicides. While the five ACCase mutations explained most of the resistance to cycloxydim (Figure 1), this was not the case for a significant proportion of resistance to fenoxaprop (Figure 1), clodinafop and haloxyfop. This proved that other mechanisms (other ACCase mutations, metabolism-based resistance...) were also responsible for resistance to these three herbicides.

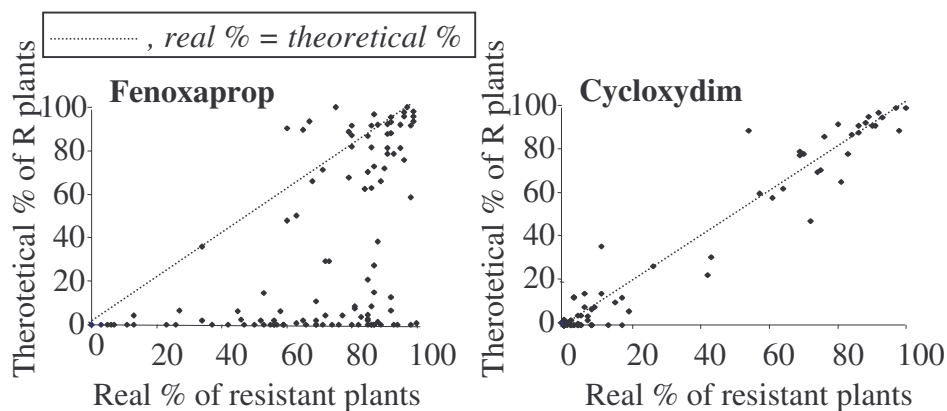


Figure 1. Real vs theoretical proportions of plants resistant to two herbicides in 116 black-grass populations.

*Is there a fitness cost associated with resistant ACCase alleles?* The biochemical properties of some mutant ACCase alleles (Délye et al. 2003; 2005) as well as their respective frequencies between French black-grass populations suggested that reproductive cost may be associated with some of the five mutations. These costs likely differ between mutations.

*Conclusion.* Genetic control of resistance to ACCase-inhibiting herbicides in black-grass is rather complex, and seems to evolve independently in each population. Various resistance mechanism can coexist within a single population, and even within a single plant. The picture emerging from this work is that, under local herbicide selection, each population evolves its own choice of “a la carte” resistance mechanisms, thus developing unpredictable cross-resistance patterns. Hence, a global policy for black-grass management seems unachievable.

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# Physiological and molecular basis of glyphosate resistance in *Conyza canadensis* Cronq.

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## Introduction

At the present, approximately 250 herbicide-resistant weed biotypes distributed among 52 different countries, involving at least 17 different modes of action were identified. Since 1974 glyphosate has been used worldwide. No case of evolved resistance to glyphosate under field conditions had been identified by 1993. The first case of glyphosate-resistant weed biotype has been documented in 1996. In the last eight years the number of new cases has dramatically increased. To date, evolved resistance to glyphosate has been identified in accessions of *Lolium rigidum* from Australia and South-Africa, *Lolium multiflorum* from California (USA) and Chile, *Eleusine indica* from Malaysia, *Plantago lanceolata* from South-Africa, *Conyza bonariensis* from South-Africa and Spain, *Conyza canadensis* from USA. The occurrence and spread of glyphosate-resistant *C. canadensis* in the eastern US likely represents the greatest immediate concern and impact to corn and soybean production in the upper Midwest. The rapid increase in occurrence of glyphosate-resistant *C. canadensis* biotypes is due in part to the widespread adoption of no-tillage Roundup Ready soybean production systems, but it is also due to the horseweed biology. The aim of the present study was to investigate the physiological and molecular basis of glyphosate resistance in two *C. canadensis* biotypes (sampled in RR soybean fields from Ohio and Delaware, USA).

## Materials and methods

The experiments were carried out on two glyphosate-resistant (HO, OH) and one susceptible (WA) *C. canadensis* biotypes. The R and S biotypes, collected in Delaware and Ohio (USA), were kindly provided by Prof. M. VanGessel and Prof. M. Loux. The EPSPS enzyme extraction and activity assay were performed according to the method proposed by Yuan et al. (2002). For uptake, metabolism and translocation trials  $^{14}\text{C}$ -glyphosate solution ( $2.5 \text{ KBq } \mu\text{l}^{-1}$ ) was applied on leaves of *C. canadensis* plants at the rosette stage. Absorbed and unabsorbed radioactivity was quantified by liquid scintillation spectroscopy (LLS) (2500 TR Liquid Scintillation Analyser, Packard). Plant extracts were spotted on 250- $\mu\text{m}$  silica gel TLC plates (SG60 with fluorescent marker; Merck, Germany) in order to separate glyphosate and potential metabolites. Electronic auto-radiography and image analysis of TLC plates were performed using a Molecular Imager (Bio-Rad, USA). The distribution of radioactivity was determined in the following three parts of the plant (rosette stage): leaves, culm and roots. Total extractable and unextracted  $^{14}\text{C}$  was determined in the three plant parts by LLS. The overexpression of the target site (EPSPS enzyme) was determined using multiplex PCR technique. For each experiment (target site assay and overexpression, herbicide uptake, metabolism and translocation) determinations were carried out on at least 20 plants. Each experiment was conducted at least twice.

## Results and discussion

The resistance of the investigated R biotypes was dependant on growth stage. At two leaf stage approximately the same ED50 values were observed for the S and R biotypes, while at the rosette stage the R biotypes were approximately three times more resistant than the S biotypes. After the treatment different morphological responses in resistant and susceptible biotypes were observed. In susceptible biotypes the first phytotoxic effects were found in the



meristematic tissues, while in the resistant biotypes the first phytotoxic effects were observed at the leaf level (irreversible dehydration of leaves). From four to six weeks after the treatment the resistant plants recovered by emitting from the center of the rosette new leaves and/or new branches. As regards the physiological mechanism of resistance, the main difference between R and S biotypes was the different mobility of the glyphosate. In the R biotypes the herbicide was less translocated in the downward direction (from the leaves to the roots) and more translocated in the upward direction (from the culm to the leaves) with respect to the S biotypes. In particular eight days after the application of [<sup>14</sup>C]-glyphosate on the top of the leaves, the amount of radiolabel found in R culms was approximately half with respect to that observed in the S culms (Table 1). In contrast eight days after the application of [<sup>14</sup>C]-glyphosate on the centre of the rosette, approximately 20% of absorbed glyphosate was found in the R culms, while approximately 40% of absorbed glyphosate was observed in the S culms (Table 1). Finally the target site (EPSP synthase) is overexpressed in the R biotypes: four weeks after the treatment the EPSP synthase is approximately two times more expressed in the R than in S biotypes (Figure 1). On the basis of results obtained the resistance in the investigated biotypes seems due to three different factors. The first one is the translocation of the herbicide. After the uptake the resistant biotypes accumulate the active ingredient in the leaf tissues. However this mechanism does not protect completely the culm and the growing points. As a consequence the overexpression of the EPSP synthase plays also an important role contributing to the surviving strategy. Finally the third component is the aptitude in emitting new branches. Probably in some cases the culm of R plants is too intoxicated by the herbicide and the plants survive by emitting new branches.

Table 1. Mean distribution of radioactivity (% of absorbed <sup>14</sup>C) 8 DAT in different parts of glyphosate-resistant (HO,OH) and susceptible (WA) plants.

	Leaves	Culm	Root
Basipetal translocation			
R (HO,OH)	61 ± 6%	36 ± 4%	3 ± 1%
S (WA)	30 ± 4%	68 ± 3%	2 ± 1%
Acropetal translocation			
R (HO,OH)	78 ± 4%	19 ± 2%	3 ± 1%
S (WA)	57 ± 3%	39 ± 3%	4 ± 1%

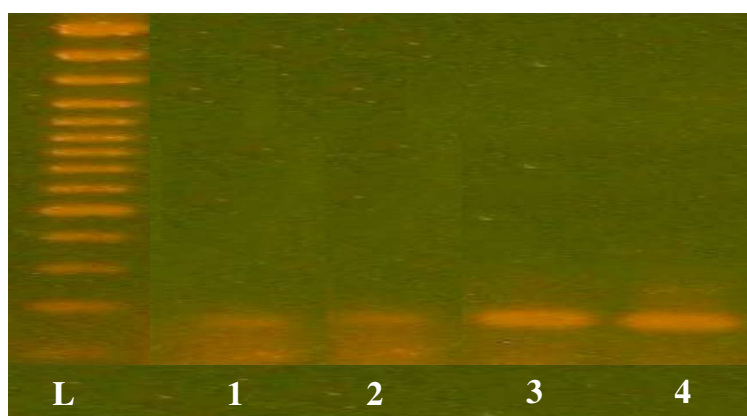


Figure 1. The expression of EPSPS enzyme in glyphosate-susceptible (WA) and resistant (HO) *C. canadensis* biotypes. L = ladder; Lane 1-2 = RNA extracted 4 WAT from WA (S) biotype; Lane 3-4 = RNA extracted 4 WAT from HO (R) biotype.

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# **An aspartate to glycine change in the carboxyl transferase domain of ACCase is correlated to ACCase inhibitors resistance in *Lolium spp.***

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## **Introduction**

Acetyl CoA carboxylase (ACCase) is a key enzyme in *de novo* fatty acid biosynthesis in all organisms. Chloroplastic ACCase is multi-domain in gramineae and is the target of two dissimilar classes of herbicides, the aryloxyphenoxypropionates (FOPs) and the cyclohexanediones (DIMs).

Resistance to these ACCase inhibitors in monocot weeds has developed over the years and is mainly caused by detoxification via enhanced metabolism and/or insensitivity of the target enzyme. In the latter case a single point mutation in the carboxyl transferase binding domain is sufficient to cause broad resistance to most ACCase herbicides in several monocot weeds of agronomic importance, including *Lolium spp.* Two such mutations have been documented, namely the I1781L change conferring strong resistance to all FOPs and most DIMs, and the I2041N mutation affecting FOPs significantly and DIMs marginally<sup>1</sup>.

Integrated field/glasshouse biological and molecular studies, however, indicate that other forms of herbicide insensitive ACCase exist in monocot weeds. In this context we have determined the molecular basis of an additional target site resistance mechanism in a UK *Lolium spp.* biotype and have subsequently designed a PCR method for the simple detection of this ACCase resistance mutation. Understanding the basis of resistance remains very important for development of a rational weed resistance management strategy.

## **Materials and methods**

Seed source and initial whole plant pot assays: ACCase inhibitor resistant *Lolium spp.* seeds (biotype 2001UK24) were collected from a wheat field in Leicestershire, UK in 2001. Initial herbicide resistance profiles were obtained based on 2.5 leaf stage seedlings of the resistant and a standard sensitive biotype treated with diclofop-methyl (500 g ha<sup>-1</sup>) and sethoxydim (200 g ha<sup>-1</sup>) in a whole plant pot assay format.

Selection of sensitive and resistant plants: Seeds from the resistant biotype were sown, 34 individual plants were tiller propagated and two separate tillers from each the plants were sprayed with 200 g ha<sup>-1</sup> and 400 g ha<sup>-1</sup> sethoxydim for selection of resistant and sensitive plants from biotype 2001UK24.

ACCase gene sequencing and comparison: Invitrogen kits (USA) were used to (i) extract RNA from 6 sensitive and 6 resistant biotype 2001UK24 plants (ii) amplify, following a RT-PCR procedure, a 2274 bp ACCase coding fragment comprising the whole carboxyl transferase (CT) binding domain. Seqman software (Lasergene, USA) was used for sequence alignment and comparison.

Routine molecular identification of the novel ACCase resistance mutation: A dCAPS<sup>2</sup> procedure using genomic DNA was designed for routine detection of the novel critical herbicide resistance nucleotide change at position 6233. This comprises amplification of a 181 bp PCR fragment with an altered reverse primer allowing differentiation between resistant and sensitive ACCase alleles upon digestion with restriction enzyme *Rsa I* and separation using simple horizontal agarose gel electrophoresis.

## **Results and discussion**

Whole plant herbicide pot assays on resistant *Lolium spp.* biotype 2001UK24 showed 5% control with diclofop-methyl and around 50% control with sethoxydim whilst the standard

sensitive biotype PP1 was totally controlled. Molecular characterisation (via PCR) of resistant plants revealed the absence of both I1781L and I2041N mutations previously shown to confer resistance to ACCase inhibitors. Since sethoxydim has never been found to be affected by metabolic resistance in *Lolium spp.*, early indications were therefore that this biotype was a mixed population of sethoxydim sensitive and resistant plants with a new insensitive target site mutation. Some metabolism was also suspected due to higher levels of resistance to diclofop-methyl.

Sethoxydim treatments at 200 and 400 g ha<sup>-1</sup> on 2 separate tillers of each of 34 individual plants gave identical results with the same 18 plants being killed and 16 surviving the 2 herbicide treatments. Six each of these two sensitive and resistant plant groups were sequence characterised using an RT-PCR based procedure. The 2274 gene fragment generated showed high homologies (>90 % amino acid identity) with the published black grass sequence, confirming the identity of the gene, and revealed 18 SNPs within the CT binding domain among the plants. Four were non-synonymous, of which 3 were equally distributed in both sensitive and resistant plants. By contrast, a change from adenine to guanine at position 6233 (black grass equivalent), corresponding to an aspartate to glycine (D2078G) mutation in the ACCase, was always present in 1 or 2 copies in resistant plants but absent in sensitive ones (Figure. 1). This latter finding was upheld in the other 10 resistant (DG or GG2078) and 12 sensitive (exclusively DD2078) plant lines, implying dominance of G2078 over D2078 allele at the sethoxydim rates used. This D2078G resistance mutation can be conveniently identified using a dCAPS procedure (see Figure 2).

In conclusion this study has identified a novel point mutation closely correlated with and very probably responsible for sethoxydim resistance in *Lolium spp.* Its' easy detection in *Lolium spp.* populations will facilitate management of ACCase inhibitor resistance in these important agronomic weed species.

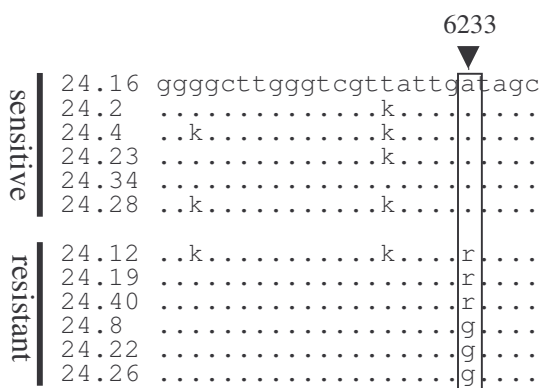


Figure 1. ACCase gene alignment around the critical 6233 nucleotide position among 6 each sethoxydim sensitive (aa) and resistant (r = ag or gg) plants. Variability at nucleotide positions 6216 and 6228 amounts to silent mutations.

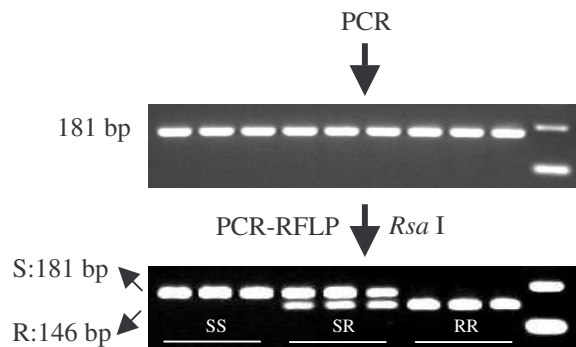


Figure 2: dCAPS procedure for the detection of the adenine to guanine change (D2078G mutation) at nucleotide position 6233 in *Lolium spp.*: *Rsa* I undigested (181 bp) and digested (146 bp) fragments correspond to sethoxydim sensitive D2078 and resistant G2078 ACCase alleles respectively. Heterozygous plants display both S & R fragments.

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# Studies of weed species *Solanum nigrum* L. resistance to ALS inhibitors

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## Introduction

According to data obtained by HRAC (2004) to-date resistance of 172 weed species has been determined in over 2000 fields. In 1988 in Serbia, *Amaranthus retroflexus* resistance to triazines was established for the first time. ALS inhibitor resistance first determined in our country was of species *A. retroflexus* and *Echinochloa crus-gally* (Konstantinovic & Meseldzija, 2002). Due to the occurrence of a huge *Solanum nigrum* L. population in 2003 research began on it's resistance to ALS inhibitors.

## Materials and methods

During 2003 and 2004 studies were performed with the aim of determining resistance of *S. nigrum* L. biotypes to ALS inhibitors. Studied biotypes from localities Becej, Zabalj and Kikinda in the Vojvodina region had long history of ALS inhibitors use. Plant populations from ruderal sites that were free of herbicide treatment were used as reference. Studies were performed by Petri dish essays (Clay & Underwood, 1990) and whole plant studies (Moss, 1995). Plants were sprayed with a range of imazethapyr rates of 0.04; 0.08; 0.15 and 0.2 kg a.i./ha<sup>-1</sup>

## Results and discussion

The highest germination capability of the species *S. nigrum* L. was established at the locality Zabalj (Figure 1.), whereas somewhat later (Figure 2.) high germination capability was also determined at the locality Becej. Results of the stem height measurement suggest that plants originating from localities Kikinda and Becej T=7 had lower values in regard to the remaining localities. Stem height values from ruderal sites exhibited satisfactory development only without treatment, whereas imazethapyr concentrations of 0.04; 0.04 and 0.08 kg a.i./ha<sup>-1</sup> caused extreme susceptibility and plant decay.

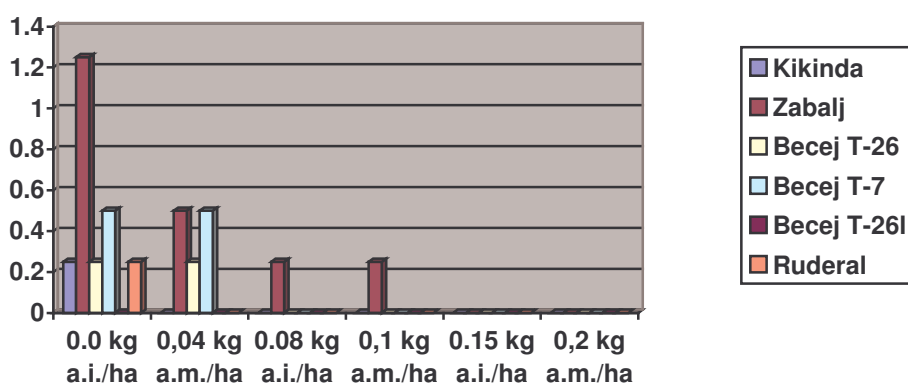


Figure 1. *Solanum nigrum* L. seed germination capability 5 days after setting up of the trial

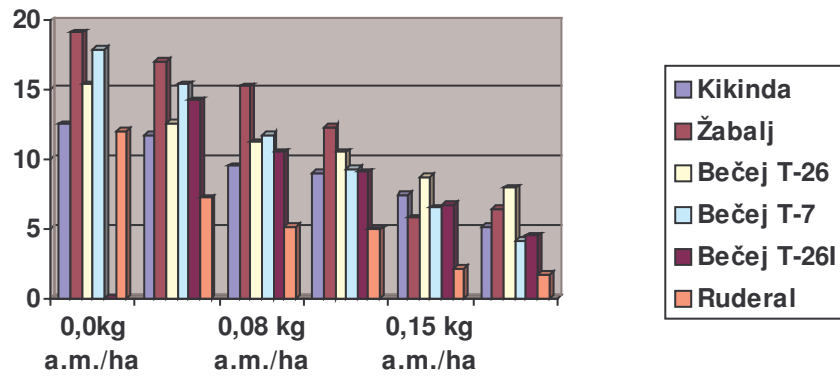


Figure 2. *Solanum nigrum* L. seed germination capability 15 days upon setting up of the trial

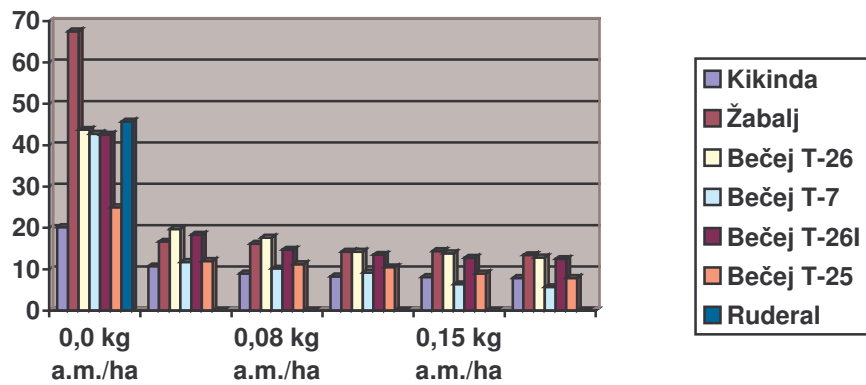


Figure 3. *Solanum nigrum* L. mean stem height values

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# Phenomenon of weed resistance in Poland – identification and prevention

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## Introduction

The origination of resistant weed populations and their spread is one of the most serious problems that complicate the system of weed control in many advanced countries throughout the world.

In the U.S.A., the first reported case of a weed resistant to the triazine herbicides was reported in the mid-1960's (Ryan, 1970). Selection pressures put on weeds by herbicides have actually resulted in 291 herbicide-resistant biotypes (Heap, 2004).

In Poland, the first cases of resistance origination were recorded in the mid 1980's, in the areas with intensive weed killing. Persistent herbicides with the same control mechanism (inhibition of photosystem II) were applied for many consecutive years to maize monocultures and orchards (Gadomski et al., 1996; Lipecki, 1988; Rola et al., 1989). On these fields, weed biotypes as: *Chenopodium album*, *Amaranthus retroflexus* and *Echinochloa crus-galli* exhibited resistance to atrazine, simazine, cyanazine and prometryne.

The aim of research was identification of resistant weeds to herbicides as: derivative of triazines (atrazine, simazine, cyanazine and metamitron), ureas (chlortoluron, linuron and isoproturon), uracils (lenacil), pyridazinones (chloridazon), benzothiadiazoles (bentazone) and phenyl-carbamates (phenmedipham and desmedipham) and develop prevention chemical methods for limited this phenomenon.

## Materials and methods

The research was conducted during five years (2000-2004). Plants (leaves) and seeds of weeds were collected from over 400 fields in South-West Poland (Lower Silesia). On these fields farmers cultivated through 3-8 years, usually in monoculture, maize, sugar beet and cereals (mainly winter wheat) and intensive chemical weed control by herbicides was used. All information was obtained from discussion with farmers.

Identification of herbicide resistant weeds by biological test: The seeds of weed were planted in a plastic pots (5 seeds per pots) containing 1:1 (by volume) mixture of peat and sand. Seedlings were grown in a greenhouse. Herbicides were applied to the plants at the four-leaf stage of development. Rates of herbicide corresponded to 0.5, 1, 2, 4 and 8 time the recommended dose applied in field. Treatments were evaluated for fresh weight measurement and phytotoxicity to plants 14 days after treatment. Visual observations were recorded using a scale of 0-100, with 100 indicating no injury and 0 equal to plant death.

Fluorescence measurement: For chlorophyll fluorescence analysis, the method of Ducruet and Gasquez (1978) was used with the following modifications (based on works De Prado et al. (1988) and Fraga & Tasende (2003)). The slow fluorescence induction curves of leaves were recorded.

Some resistant samples were tested by molecular method - PCR technique (Gadomski et al., 1996).

## Results and discussion

Based on the methods resistant biotypes of tested weeds collected from fields in South-West Poland were identified. Results from researches are shown in Table 1.

Moreover, also phenomenon of cross-resistance to all of triazines and other tested herbicides were observed.



Table 1. Identification of herbicide resistant biotypes of weed.

Weeds	Resistant to:					
	Triazines	Ureas	Benzothia- diazoles	Pyridazi- nones	Uracils	Phenyl- carbamates
<i>Anthemis arvensis</i> L.	Yes	No	No	Yes	NT	O
<i>Amaranthus retroflexus</i> L.	Yes	NT	Yes	O	O	No
<i>Capsella bursa-pastoris</i> L.	Yes	No	Yes	Yes	No	No
<i>Chenopodium album</i> L.	Yes	Yes	Yes	Yes	Yes	No
<i>Echinochloa crus-galli</i> L.	Yes	O	O	O	O	O
<i>Lamium amplexicaule</i> L.	Yes	NT	Yes	Yes	Yes	No
<i>Lamium purpureum</i> L.	Yes	NT	Yes	Yes	Yes	No
<i>Papaver rhoeas</i> L.	Yes	NT	Yes	Yes	No	No
<i>Polygonum aviculare</i> L.	Yes	Yes	No	Yes	Yes	No
<i>Polygonum convolvulus</i> L.	Yes	Yes	No	Yes	Yes	No
<i>Polygonum persicaria</i> L.	Yes	Yes	No	Yes	Yes	No
<i>Sinapis arvensis</i> L.	Yes	No	Yes	No	No	No
<i>Solanum nigrum</i> L.	Yes	Yes	No	Yes	Yes	NT
<i>Stellaria media</i> (L.) Vill.	Yes	Yes	Yes	No	NT	No
<i>Viola arvensis</i> Murr.	Yes	O	No	NT	NT	No

NT – not tested

O - natural tolerance

Similar results of resistance and cross-resistance to triazines and other substances inhibitors of photosystem II occurred in weeds described De Prado (1995), Furest et al. (1986), Mikulka & Chodova (2000), Gadomski et al. (1996).

Moreover, in field and green house conditions, chemical methods to resistant weed control were tested. Herbicides from other groups than inhibitors of photosynthesis very good controlled resistant biotypes of weeds (85-100% of efficacy), e.g. in maize crop – nicosulfuron, rimsulfuron, mesotrione, iodosulfuron, isoxadifen; in sugar beet crop – ethofumesate, clopyralid, triflurosulfuron; in cereals – diflufenican, triasulfuron, 2,4-D, MCPA, mesosulfuron, iodosulfuron, pendimethalin, carfentrazone-ethyl.

### Acknowledgements

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# Biotypes of *Apera spica-venti* and *Centaurea cyanus* resistant to chlorsulfuron in Poland

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## Introduction

One of the negative aspects of the intensive use of herbicides is related to the selection of resistant biotypes (Gasquez, 2001). Of all biotypes resistant to herbicides, 70 biotypes do not respond to sulfonylurea herbicides (ALS-inhibiting herbicides). The enzyme acetolactate synthase (ALS) takes part in biosynthesis of a branched chain amino acids (valine, leucine, isoleucine).

In Poland the problem of resistance to sulfonylurea herbicides has been discussed since 2001 (Rola & Marczewska, 2002).

The aim of the experiments was to investigate the level of resistance of different weed biotypes in fields of winter wheat.

## Materials and methods

Resistance tests of *Apera spica-venti* and *Centaurea cyanus* biotypes to chlorsulfuron were conducted in greenhouse by biological tests. The experimental material was collected from fields in south–west Poland, where winter wheat monoculture and intensive chemical weed control with chlorsulfuron had been used for long time. The weed seeds were sown in plastic pots containing 2:1 mixture of peat and sand.

Chlorsulfuron was applied at four–leaf stage of development at rates ranging from 1 to 32 times of the recommended field dose (11.25–360 g a.i./ha). The herbicide was applied using a chamber sprayer with nozzle type Teejet XR11003 - VS and calibrated to deliver 250 l/ha at a pressure of 200 kPa.

The experiments were conducted using a completely randomized design with three replications. ED<sub>50</sub> was used to evaluate herbicidal efficacy.

## Results and discussion

For *C. cyanus*, the application of chlorsulfuron at rates of 11.25 to 45 g a.i./ha caused a fresh weight reduction from 0 to 25%, and a dry weight reduction from 5 to 40%. The application of chlorsulfuron at rates of 90 to 180 g a.i./ha caused a reduction of the aboveground part of *C. cyanus* plants of about 50%, but the plants still showed good vigour. Only when the highest dose of herbicide (360 g a.i./ha) was applied leaves started to wilt and fresh and dry weight were reduced by more than 50% (Figure 1). This result confirms that the tested biotype is resistant to chlorsulfuron.

As far as *A. spica-venti* is concerned, the application of chlorsulfuron at doses ranging from 11.25 to 45 g a.i./ha, caused a reduction of fresh and dry weight from 0 to 10%. When chlorsulfuron was applied at the highest dose (360 g a.i./ha) the reduction of aboveground part of *A. spica-venti* was about 25%. The plants were in good conditions and leaf wilting did not appear. The biotypes of this species proved to be highly resistant to the herbicide —more so than the *C. cyanus* biotype.

Bioassays showed that the tested biotypes of *A. spica-venti* and *C. cyanus* are resistant to chlorsulfuron. The high level of resistance of *A. spica-venti* may indicate the occurrence of a target site.

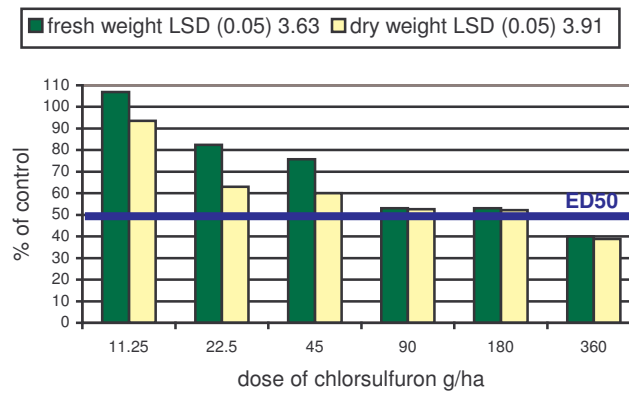


Figure 1. Influence of different doses of chlorsulfuron on fresh and dry weight of *Centaurea cyanus*.

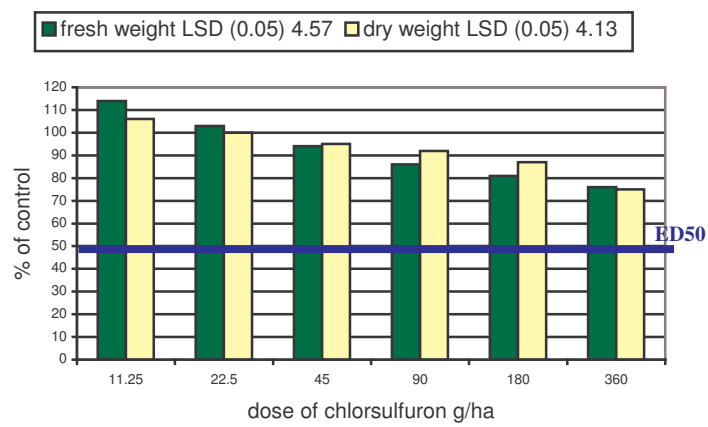


Figure 2. Influence of different doses of chlorsulfuron on fresh and dry weight of *Apera spica-venti*.

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## Ploidy level assessment of the Czech kochia (*Kochia scoparia* (L.) Schrad.) biotypes resistant to acetolactate synthase inhibitors and atrazine

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### Introduction

Resistance of kochia (*Kochia scoparia* (L.) Schrad.) to acetolactate synthase (ALS) inhibiting herbicides, and atrazine was found in the proximity of railway stations in the Czech Republic (Chodová & Mikulka 2000). Diploid number of chromosomes ( $2n = 18$ ) was previously proved in the radicles of both resistant and susceptible populations of kochia (e.g. Měsíček & Jarolímová 1992). As polyploidy may rapidly enhance the adaptation of species to environmental changes (Rauber 1977), ploidy level estimation using flow cytometry was performed on vegetative parts of both biotypes. Flow cytometry has become very popular technique in plant sciences particularly beneficial for convenient cytotype screening in large population samples.

### Material and methods

Eight kochia biotypes originating from different sites in the Czech Republic were analysed (Table 1). They differed in the susceptibility/resistance to herbicides inhibiting ALS and atrazine. Germinating seeds with radicles of 5-15 mm in length were the most suitable material for chromosome counting (stained with Schiff's reagent). The number of chromosomes was determined in meristematic cells of root tips for 50 plants per biotype. Various vegetative parts of both susceptible and resistant biotypes of *Kochia scoparia* were screened for their ploidy levels using Partec PA II flow cytometer. Nuclei isolation and DAPI staining (4 µg/ml) followed the modified two-step procedure originally developed by Otto (1990). The *Glycine max* cv. Polanka (2C = 2.5 pg) was selected as an internal standard. The histograms were evaluated using Flomax software.

### Results and discussion

Diploids with 18 chromosomes clearly prevailed in the analysed plant set. Low proportion of tetraploids ( $2n = 4x = 36$ ) was found in the resistant population from Bubny. Besides diploidy, a high frequency of mixoploidy (2x, 4x) was observed in the root tips of plants from both herbicide susceptible and resistant populations (Table 1).

Table 1. Results of karyological analyses of the root tips of *Kochia scoparia*

Sites	Susceptibility / resistance	% of plants with number of chromosomes		
		2 n	Mixoploidy	4 n
Bubny (railway)	R <sub>atr</sub> R <sub>ALS</sub>	22	68	10
Karlín (parking-site)	S <sub>atr</sub> S <sub>ALS</sub>	62	38	0
Žižkov (roadside)	S <sub>atr</sub> S <sub>ALS</sub>	48	52	0
Karlín (roadside)	S <sub>atr</sub> S <sub>ALS</sub>	50	50	0
Libeň (railway)	S <sub>atr</sub> R <sub>ALS</sub>	18	82	0
Vršovice (railway)	R <sub>atr</sub> R <sub>ALS</sub>	36	64	0
Olbramovice (field)	S <sub>atr</sub> S <sub>ALS</sub>	80	20	0
Jihlava (railway)	S <sub>atr</sub> R <sub>ALS</sub>	36	64	0
Commercial seed *	S <sub>atr</sub> S <sub>ALS</sub>	78	22	0

R<sub>atr</sub> = resistant to atrazine, S<sub>atr</sub> = susceptible to atrazine, R<sub>ALS</sub> = resistant to ALS inhibitors, S<sub>ALS</sub> = susceptible to ALS inhibitors, \* *Kochia scoparia* var. *trichophylla* (Seva-Valtice, CR)

None of the karyologically-examined populations consisted exclusively of plants with diploid number of chromosomes in their radicles as it had been reported for kochia biotypes from Kansas and North Dakota (Thompson et al. 1994).

Only DNA-diploid individuals were detected using flow cytometry. Nevertheless, endopolyploidy occurred in all vegetative parts analysed (nuclei with 2C, 4C, and 8C DNA contents) (Figure 1). Mean proportions ( $\pm$  SD) of particular nuclei classes in leaf tissues were  $55.2 \pm 11.0$  % (2C),  $37.6 \pm 7.4$  % (4C), and  $7.2 \pm 4.7$  % (8C). No significant difference in endopolyploidy profile between susceptible and resistant biotypes was observed. Generally, older tissues showed higher level of endopolyploid nuclei indicating certain age-related correlation. Progressive endopolyploidy was previously found in several other angiosperms, e.g. in various *Brassica* species (Kudo & Kimura 2001). Nuclei isolated from stems and both non-flowering and flowering branches yielded histograms comparable to those obtained from leaf tissues. High stability of genome size in investigated *Kochia* plants was proved, owing to the very low variation (< 2 %) in nuclei fluorescence intensity during simultaneous analyses with internal standard *Glycine max*.

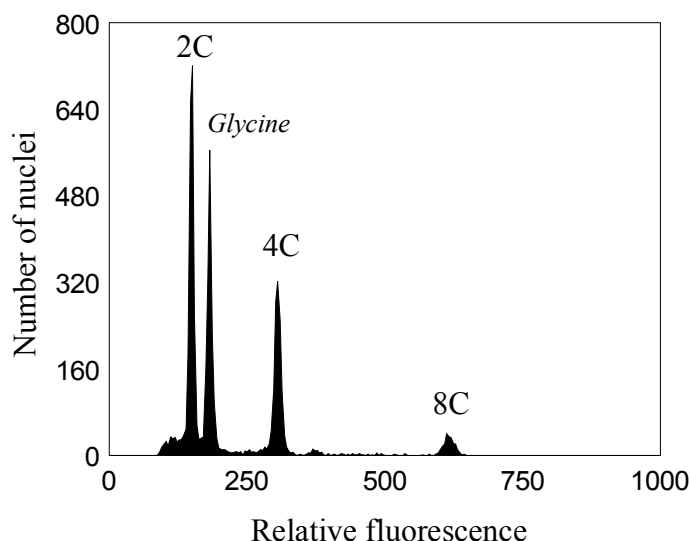


Figure 1. Flow-cytometric profile of leaf tissue of *Kochia scoparia* showing nuclei with 2C, 4C, and 8C DNA amounts. *Glycine max* was used as an internal standard.

### Acknowledgement

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## **Herbicide resistance in weedy plant species: From evolution to management.**

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It is approaching 40 years since the landmark paper (Ryan, 1968) first describing the evolution of herbicide resistance. Since then, herbicide resistance has evolved many times in many weedy plant species in many parts of the world (see the Heap WSSA weedscience.com website for a full list of resistant biotypes). Herbicide resistance is now a global phenomenon causing significant production challenges in some agro-ecosystems. Herbicide resistance is a classic example of Darwinian evolution. When very large, initially herbicide susceptible weedy plant populations are exposed to a strong herbicide selection pressure there is very high mortality. However, not all individuals succumb to the herbicide. In large populations with sufficient genetic diversity there are some individuals that survive and reproduce because they possess a gene or genes (resistance genes) which endow survival at the rate of herbicide used. These surviving individuals reproduce and if the herbicide use persists, these resistance genes are enriched in the population to the point where the majority of the individuals in the population contain the resistance genes and commercial herbicide failure becomes evident. There is now a wealth of understanding about herbicide resistance at levels from the factors that lead to resistance, the molecular mutations endowing resistance and through to ways in which to best minimise resistance evolution. This presentation will review some major issues in herbicide resistance.

**a) Resistance evolves where there is insufficient diversity.** Due to economic and other realities there is often insufficient diversity in agro-ecosystems. Too often the same herbicide is applied too frequently to the same area. Too often, there is no or very limited diversity in the crops under cultivation. Too often the herbicide is the sole weed control tool used on the area of land. This lack of diversity is a fertile breeding ground for herbicide resistance. Resistance will evolve because the insufficient diversity provides the environment for resistance genes to be enriched in the weed population to the point that herbicide failure occurs. Conversely, where land managers achieve diversity in herbicide and crop choice and use non-herbicide weed control methods then, often, resistance genes and/or weed numbers remain at relatively low levels. Therefore, diversity is a key to sustainable herbicide usage. Of course, the challenge is to balance crop, herbicide, weed control choices, with prevailing economic realities. Wise decision makers will recognise both the economic realities as well as the biological reality that insufficient diversity will lead to resistance. There are major needs and opportunities for biologists and economists to tackle sustainability issues with our precious herbicide resources.

**b) Herbicide rate is important.** It must be emphasised that a large, genetically diverse weed population is NOT comprised of individuals that are fully genetically susceptible to a herbicide, plus a very few individuals that are fully genetically resistant. Rather, there is great genetic diversity in the population, manifest as a continuum of degree of herbicide susceptibility. This is why an herbicide dose response is always evident. Treat any large weed population with an herbicide and a continuum is evident from these few individuals which die at a very low rate of herbicide with mortality asymptotically increasing at progressively higher herbicide rate. Thus, there is continuous genetic variation in herbicide susceptibility in the population. The great majority of individuals cannot survive high herbicide dose but can survive moderate herbicide rate because they contain “weak” resistance genes. The practical reality flowing from this realization is that when an herbicide is applied it should be used at



rates that give very high weed mortality. This will ensure that individuals with “weak” resistance genes will be killed and therefore these “weak” resistance genes will not be enriched. Conversely, when herbicide rates are lower then individuals with “weak” resistance genes survive and these “weak” resistance genes are enriched in the population, leading to resistant populations. Recent direct evidence of the influence of herbicide rate on resistance evolution will be presented.

**c) All possible resistance mechanisms are selected and enriched.** Thirty years of research on the biochemical mechanisms endowing herbicide resistance reveal that a range of resistance mechanisms are selected. Target site based resistance can be due to single nucleotide mutations causing changes in herbicide target site enzymes that endow high level resistance. Less appreciated is that single nucleotide mutations that endow low level target site resistance are also selected, especially where herbicide selection rates are lower. Both target site and non-target site resistance mechanisms/genes are selected. Non target site based mechanisms such as enhanced rates of herbicide metabolism or reduced rates of herbicide translocation are common but not always recognised. It must be realised, especially but not exclusively in cross-pollinated weed species, that a diversity of target site and non-target site resistance mechanisms (genes) are enriched and accumulated in resistant weed populations. This results in individuals possessing multiple herbicide resistance genes/mechanisms. The diversity of resistance mechanisms/genes that are responsible for resistance and the threat of multiple resistance is not as widely appreciated as it should be.

**d) Resistance minimisation and management requires diversity.** Given the factors discussed above, it is evident that to slow the rate of resistance evolution and/or to manage resistance when it has evolved requires that there be sufficient diversity in herbicide usage and the agro-ecosystem in which selection is occurring. While solutions to resistance problems will always be on a case-by-case basis they will all require diversity. Substantial challenges exist for the herbicide industry and for researchers and practitioners to devise systems which minimise and manage herbicide resistance within economic realities. Recent research on the above factors will be summarised.

# Fallow management and its implication on a herbicide-resistant *Papaver rhoeas* L. population

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## Introduction

The appearance of herbicide resistant biotypes of *Papaver rhoeas* in NE of Spain, is related to the reduction of cultural methods and a greater dependence of herbicides (Taberner et al., 2001). Several non chemical strategies have been proposed to control these populations by soil ploughing or by alternative mechanical control (Cirujeda et al., 2003, Recasens et al., 2004). Soil ploughing efficiency is variable during different seasons due to difficulty in obtaining a correct soil turn. Mechanical control by harrowing offers a good alternative control of the populations, but the efficacy depends on different factors as the phenological stage of plants, speed of harrowing, and soil conditions (Rasmussen & Svenning 1995). If the establishment of a program to manage herbicide resistant populations is required, other strategies should be included. For other weeds of cereal crops, the incorporation of one fallow season (Daugovish et al, 1999) or delaying the crop sowing date (Gill & Holmes, 1997) can promote a depletion of seed bank and better weed control during the next crop season. In this work, these two management strategies are analyzed on a herbicide resistant *P. rhoeas* population.

## Materials and methods

The work was carried out during the cropping season 2002/03 and 2003/04 in a commercial wheat crop field in Cubells (Lleida, Spain) in which the presence of a herbicide resistant *P. rhoeas* population to 2,4-D had previously been confirmed. Six different management systems were applied in a randomized block design with three replicates. Each block contained six plots of 8-m width and 10-m length. The six systems carried out were: 1) Wheat monocrop with herbicide application each season. 2) Wheat in 02/03 and barley in 03/04 with weed control by herbicides both seasons. 3) Fallow in 02/03 with an application of one persistent herbicide, and wheat in 03/04 with weed control by herbicides. 4) Fallow in 02/03 with an application of one non persistent herbicide and wheat in 03/04 with weed control by herbicides. 5) Fallow in 02/03 and wheat in 03/04 with weed control by mechanical harrowing both seasons, and 6) Fallow in 02/03 with soil ploughing in spring and wheat in 03/04 with soil ploughing before sowing and weed control by mechanical harrowing. In 02/03 season the herbicide applied was Image (bromoxinil 12% + ioxynil 12% + mecoprop 36%) in both cropping plots. The persistent herbicide for the third system was Image plus Gadisan (linuron 12% + trifluralin 24%) and the non persistent herbicide for the fourth system was Roundup (glyphosate isopropylamine salt 36%). In 03/04 season the herbicide was Granstar (tribenuron-methyl) for plots with chemical control. All herbicides were applied as commercial doses. The weed density was estimated periodically in five fixed frames of 33 x 33 cm installed in each plot. The wheat plots were sown on the 12 November 02 and on the 28 November 03, and the barley plots were sown on the 21 January 04.

## Results and discussion

A high initial plant density was observed in November 2002 in all plots (from 1800 to 2700 seedlings/m<sup>2</sup>) just after crop sowing (Table 1). During the following weeks a significant natural mortality was detected (from 21 to 43% of the initial seedling density). In the plots with wheat the plant density at treatment was 863 plants/m<sup>2</sup> and the efficiency of herbicide was higher than 98%. In all other plots the different control strategies applied on fallow were

also effective and practically with no plants during the rest of crop season. Only in the fallow plots with control by harrowing and in the fallow plots with application of a persistence herbicide, a very low density of 2 pl/m<sup>2</sup> and 1 pl/m<sup>2</sup> was observed respectively.

The initial seedling density just after sowing in December '03 (from 4 to 96 seedlings/m<sup>2</sup>) was significantly lower than that in the previous season and represented an average reduction of 98% from one year to another. These lower values of initial seedling density this second season, seem to be a consequence not only of the reduction of soil recruitment of seeds the previous summer, but also of the delay in sowing date (16 days in comparison with autumn 2002), due to the frequent autumnal rainfall. This delay allows a significant reduction of soil seed bank due to the emergence of seedlings before sowing (with mean values of 48 seedlings/m<sup>2</sup>). During the season 03/04, in plots with wheat monocrop, an initial seedling density of 96 seedlings/m<sup>2</sup> was observed, whereas in plots in fallow the previous season, these initial weed densities were lower and approximately 50 seedlings/m<sup>2</sup> in all cases. In the plots where barley was sown in January '04 a lower and petty initial plant density was observed (4 seedlings/m<sup>2</sup>). The different managements carried out during the crop season in the different plots were equally efficient in the reduction of weed density. Only the mechanical control by harrowing showed efficacy less than 100% and a final plant density of 5 plants/m<sup>2</sup> at harvest time.

Table 1. Seedling density

Crop system	02/03			03/04		
	Initial density (pl/m <sup>2</sup> )	Final density (pl/m <sup>2</sup> )	Reduction of plant density (%)	Initial density (pl/m <sup>2</sup> )	Final density (pl/m <sup>2</sup> )	Reduction of plant density (%)
1	2273 ± 383	33 ± 17	98.7	96 ± 47	0	100
2	1867 ± 47	32 ± 19	98.3	4 ± 1	0	100
3	2716 ± 25	1 ± 1	100	48 ± 4	0	100
4	2701 ± 482	0	100	50 ± 7	0	100
5	2000 ± 228	2 ± 2	99.9	49 ± 2	5 ± 4	89.7
6	1865 ± 179	0	100	40 ± 3	0	100

Values are means and S.E

In the management of a resistant herbicide *P. rhoeas* population, different strategies should be incorporated. The incorporation of fallow within a crop system during one season allows a significant reduction of plant density. The management of this fallow is possible without dependence of herbicides (i.e. ploughing or harrowing) and with the same results. However, the delay of crop sowing date seems to be the most significant factor in reducing the soil seed bank. The use of cereal varieties of short cycle or adapted to a sowing delay can favour this soil seed depletion even more.

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## The impact of transgenic herbicide-resistant crops on herbicide-resistant weeds and the environment: a review

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Genetically modified (GM) crops are cultivated commercially in 2004 on more than 80 million hectares in 17 countries in the developed and developing world (James, 2005). The most important GM crops in terms of acreage are soybean, corn, oilseed rape (canola), and cotton, while the most important trait is herbicide resistance comprising almost 80% of the total GM crops acreage. Herbicide-resistance traits have been introduced and tested in numerous other crops, but for various reasons their introduction to the market has been terminated or postponed (Devine, 2005). The steady decrease in the number of workers employed in agriculture ensures the dominance of herbicide-resistant crops (HRC) over other GM crops.

Genetically-modified herbicide-resistant crops were first introduced in 1995 with bromoxynil-resistant cotton followed by glufosinate-resistant ('Liberty') and glyphosate-resistant (Roundup-Ready<sup>®</sup> – RR) crops. Plants confer herbicide-resistant by two major mechanisms, *i.e.* by expression of an insensitive form of the target site of the herbicide in the plant, such as the modified enolpyruvyl-shikimate-phosphate synthase (EPSPS) that is insensitive to glyphosate or by expression of detoxification enzymes that convert the herbicide to non-toxic compounds, such as the phosphinothricin acetyl transferase (PAT) enzyme that acetylates the herbicide glufosinate to the non-toxic N-acetylglufosinate.

A novel glyphosate-resistance mechanism was recently introduced in which plants became resistant by expressing a microbial enzyme that detoxifies glyphosate via N-acetylation (Castle et al., 2004). The original enzyme isolated from *Bacillus licheniformis*, was modified through DNA shuffling and its glyphosate N-acetylation activity enhanced 7000-fold before expressed in several crops endowing high resistance to field rates of glyphosate.

The risks associated with the adoption of GM in general and HRC in particular, were and are under public dispute and disagreement. Part of the public is concern with moral and environmental consequences of the adoption of HRC (and GM crops in general) such as introgression of the resistance trait to weeds or other non-transgenic crops, harm to the biodiversity and effects on non-target organisms. In fact, the adoption of HRC resulted in significant changes in agronomic and weed management practices, along with changes in the spectrum and amount of herbicide use. The application of a powerful and simple-to-use herbicides such as glyphosate or glufosinate that effectively control annual and perennial weeds, offer the farmer a cost-effective way to manage weeds in reduced- or zero-tillage fields. The use of a non-selective herbicide such as glyphosate provides a new and effective tool to improve the control of troublesome weeds such as purple nutsedge (*Cyperus rotundus*) in glyphosate-resistant crops (Table 1).

The dramatic worldwide increase in use of glyphosate is attributed to its excellent performance as a non-residual 'total vegetation control' herbicide, low cost, suitability to reduced or zero-tillage conditions, and the massive adoption of RR transgenic crops. This long-term and repeated use of glyphosate in plantations, orchards and minimum tillage fields, resulted in a clear shift of weed population toward more 'naturally-tolerant' weeds such as *Malva* spp., *Commelina* spp. and others (Owen & Zelaya, 2005).

Table 1. Response of purple nutsedge (*Cyperus rotundus*) to herbicides applied over the top<sup>§</sup> or as a directed spray\* in DP5415RR cotton (After Yasuor et al., 2002).

Herbicide	Application time (DAP)	Rate (kg ae/ha)	Nutsedge infestation (%) <sup>a</sup>
Untreated control	--	--	80
MSMA	18 <sup>§</sup> +40 <sup>§</sup>	1.0 + 1.0	43
Glyphosate	18 <sup>§</sup> +40 <sup>§</sup>	0.7 + 1.4	5
Glyphosate	18 <sup>§</sup> +40 <sup>§</sup> +67*	0.7+0.7+1.1	5
Glyphosate	30 <sup>§</sup> +50*	1.1 + 1.4	10
Glyphosate	30 <sup>§</sup>	1.1	10

<sup>a</sup> Infestation level was visually estimated 100 DAP.

In addition, after more than 30 years of selection pressure, large number of weed populations evolved resistance to glyphosate, that spreads rapidly worldwide. Globally, glyphosate-resistant weed populations were confirmed in Australia, Asia, The Americas and Africa in grass weeds such as *Elusine indica* and *Lolium* spp., and the broadleaved weeds *Conyza canadensis*, *C. bonariensis*, *Plantago lanceolata* and *Ambrosia artimisiifolia*. In general, the level of resistance is relatively low (X2 to X10) and is attributed to limited translocation of glyphosate within the treated plant. In several cases (*E. indica* and *L. rigidum*) a less sensitive EPSPS has been found due to a single change in amino acid. Multiple-resistance was detected in certain glyphosate-resistant populations, which possess several resistance mechanisms. No reports are available so far, on evolution of weed populations to glufosinate.

The expected increase in the adoption of HRC, and particularly of glyphosate-resistant crops throughout the world calls for caution and requires the development and adoption of preventive management practices.

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# An Italian population of *Amaranthus retroflexus* resistant to ALS-inhibiting herbicides: resistance pattern and molecular basis

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## Introduction

The evolution of herbicide resistant weeds is of increasing concern to growers around the world and poses serious problems where there are no alternative selective herbicides to control resistant populations. The extensive use of ALS-inhibiting herbicides has led to the widespread development of resistant weeds which threaten several crop production systems. In the last ten years, resistance to ALS inhibitors has been evolving rapidly in Italy. Three species, *Alisma plantago-aquatica*, *Scirpus mucronatus* and more recently *Cyperus difformis*, have evolved resistant populations in rice crops. Meanwhile, ALS-resistant populations of *Papaver rhoeas* infesting durum wheat crops have been documented in central and southern Italy (Scarabel *et al.*, 2004). So far, in Italian soyabean and maize crops, resistance has only been found to PSII-inhibiting herbicides. However, in 2003, seeds of an *Amaranthus retroflexus* L. population poorly controlled by imazethapyr were collected from a soyabean crop in north-eastern Italy. Field-selected resistance to ALS-inhibitors is mostly due to an altered target site, namely, point mutations that occur within discrete conserved domains of the *als* gene (Tranel & Wright, 2002).

The aim of this study is to characterise the resistance status of the imidazolinone-selected *A. retroflexus* biotype and elucidate the molecular basis of resistance in this population. Alternative post-emergence herbicides for controlling this population were also tested.

## Materials and methods

Seeds from plants of *A. retroflexus* that had escaped a treatment with imazethapyr were collected during summer 2003 from a soyabean crop in north-eastern Italy (population 03-07). The field had been cropped with soyabean treated with imazethapyr for the past 5 years. Seeds from a population of *A. retroflexus* again susceptible to ALS-inhibitors (population 99-01) were collected from a location that had never been treated with this group of herbicides. Pre-germinated seeds were planted in plastic boxes filled with a standard potting mix (60% silty loam soil, 15% sand, 15% perlite, 10% peat) and placed in a greenhouse. On reaching the three to four-leaf stage, herbicides were applied as commercial formulations with recommended surfactants using a precision bench sprayer. Herbicides were: imazethapyr (1x: 35 g a.i. ha<sup>-1</sup>), imazamox (1x: 40 g a.i. ha<sup>-1</sup>), oxasulfuron (1x: 75 g a.i. ha<sup>-1</sup>), nicosulfuron (1x: 40 g a.i. ha<sup>-1</sup>), thifensulfuron-methyl (1x: 7.5 g a.i. ha<sup>-1</sup>), mesotrione (1x: 50 g a.i. ha<sup>-1</sup>), metribuzin (1x: 350 g a.i. ha<sup>-1</sup>) and terbuthylazine (1x: 1100 g a.i. ha<sup>-1</sup>). The herbicide dose ranges were chosen after preliminary screenings and ranged from 1x (i.e. recommended field dose) to 32x, and from 1/32x to 1x for resistant and susceptible populations, respectively. The number of surviving plants and shoot fresh weight were recorded 3 weeks after treatment. The experimental design was completely randomised with 3 replicates, each of 15 plants. The ED<sub>50</sub> and GR<sub>50</sub> values were estimated using the log-logistic model (Seefeldt *et al.*, 1995).

Genomic DNA was extracted from the leaf tissue of five plants. Specific primers were designed (from aligned sequences of *Amaranthus* species available in GenBank) to amplify the 430-bp fragment of ALS gene containing domains A, C and D, while another set of primers was used to amplify the 340-bp fragment containing domains B and E. The PCR-purified products were sequenced in both strands and the sequences of susceptible and resistant biotypes were aligned using Lasergene software DNASTAR.



## Results and discussion

Dose-response experiments at whole plant level showed that population 03-07 was highly resistant to both imidazolinones tested, with the selecting herbicide showing a much higher resistant ratio (>1898) (Table 1). This population was also cross-resistant to all sulfonylureas tested, with resistant ratios varying from 34 with nicosulfuron to more than 429 with thifensulfuron-methyl. The high level of resistance to ALS-inhibitor herbicides indicates that the resistance mechanism is likely to be site of action mediated. Conversely, the GR<sub>50</sub> of susceptible and resistant populations, calculated for metribuzin, terbuthylazine and mesotrione, were similar, determining resistant ratios close to 1 (Table 1). This proves that the ALS-resistant population can still be adequately controlled by PSII inhibitors and bleaching herbicides.

Although the values of the resistant ratios based on plant survival (data not shown) were generally lower than those calculated for shoot fresh weight, they gave the same indications.

Table 1. GR<sub>50</sub> (and relative standard error - S.E.) and resistant ratio of the resistant population of *Amaranthus retroflexus*. Resistant ratio = GR<sub>50</sub> resistant / GR<sub>50</sub> susceptible.

Chemical family	Herbicide	HRAC group	Shoot fresh weight		
			GR <sub>50</sub> g a.i. ha <sup>-1</sup>	S.E.	Resistant ratio
Imidazolinones	imazethapyr	B	>1120	-	>1898
	imazamox	B	167	50.6	293
Sulfonylureas	nicosulfuron	B	139	8.49	33.9
	oxasulfuron	B	>1200	-	>215
	thifensulfuron-methyl	B	>60	-	>429
Triazinones	metribuzin	C1	26.8	3.59	1.73
Triazines	terbuthylazine	C1	59.8	5.61	1.00
Triketones	mesotrione	F2	5.31	1.86	0.57

Nucleotide sequences of the ALS-amplified regions comprising A to E Domains between resistant and susceptible populations differed in one nucleotide only. The substitution had occurred in Domain B at position 574 (referred to *Arabidopsis thaliana* sequence) that determines the substitution of Trp to Leu in resistant population 03-07. The same substitution has been identified in the five plants analysed, which all resulted as being homozygous for the mutation at the ALS-locus. In other species of *Amaranthus*, the same substitution of Trp<sub>574</sub> with Leu has been shown to confer a comparable level and similar pattern of resistance to ALS inhibitors (Heap, 2004).

Although similar European cases from Serbia (*A. retroflexus* and *E. crus-galli*), Poland (*Conyza canadensis* from railways) and the Czech Republic (*Kochia scoparia* from railways) are reported in the world herbicide resistance database (Heap, 2004), this is the first documented case of resistance selected by imidazolinones used on European crops.

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# Screening for biotypes of *Conyza bonariensis* resistant to glyphosate in Southern Spain

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## Introduction

There are 3 weed species of genus *Conyza* in Southern Spain: *C. albida* Wild ex Sprengel, *C. bonariensis* (L.) Cronq. and *C. canadensis* (L.) Cronq. (Valdes *et al*, 1987).

The most problematic infestations take place in no-till permanent crops such as, peaches, citrus and olives. Difficulties in controlling *Conyza* spp with glyphosate has been reported in most regions of Iberian peninsula (Saavedra & Pastor, 2002).

Genus *Conyza* shows great potential for resistance with reported resistance to 6 modes of action in *C. canadensis* and 4 modes of action in *C. bonariensis* (Heap, 2005). A number of cases of resistance to glyphosate have been reported in *C. canadensis* and one case has been cited in *C. bonariensis* in South Africa. No case of glyphosate resistance has been confirmed in Europe to date.

A preliminary study showed that *C. bonariensis* is the predominant species within the genus in Southern Spain.

## Materials and methods

Seed was collected in the field from single plants in 43 locations of southern Spain (provinces of Sevilla, Huelva and Cordoba). Seeds from each plant were sown in separate trays. Fertilized peat substrate was used. Seeds were gently placed on the surface of the substrate, watered, and covered with cheesecloth-type material. Trays were maintained under controlled conditions with a photoperiod of 16h/8h (day/night) and temperature of 28°C/21°C (day/night). Seedlings with 2-4 true leaves were transplanted to individual pots (9 cm of diameter and 6 cm depth). The same substrate was used.

Two plant progenies (10 individuals by progeny) were transplanted for each location. After transplanting, 4 plants from each progeny were kept in the previously mentioned controlled conditions and the 6 remaining were moved to a glasshouse (without light and temperature control).

When plants reached the rosette stage (8-9 cm diameter) they were sprayed with 238 g ai.ha<sup>-1</sup> of glyphosate (Roundup Plus, a formulation of 360 g ai.L<sup>-1</sup> IPA salt of glyphosate, Monsanto). Of the 4 plants grown in the growth chamber, 3 were sprayed and 1 kept as control; of the 6 plants maintained in the glasshouse, 3 were sprayed and the other 3 kept as control.

Plants were sprayed using a Potter tower, which precise application to individual plants.

Biomass was scored 21 days after treatment (DAT).

A second experiment was conducted with seed from 6 locations to determine the GR<sub>50</sub> value. Preliminary results indicated that 1 of the populations (04-033) was “potentially sensitive” and the 5 remaining (04-013, 04-015, 04-019, 04-038 and 04-050) were “potentially resistant”. Sowing and transplanting was made in the same manner as described for the previous experiment. This experiment was developed in a net house and repeated under controlled conditions (16h/8h, 28°C/21°C day/night) with populations 04-033 and 04-038. In both environments a randomized block design was used with 5 replications and 10 glyphosate rates (0, 0.04, 0.07, 0.14, 0.29, 0.58, 1.12, 2.23, 4.46, and 9.00 kg ai.ha<sup>-1</sup>). The same herbicide (Roundup Plus, Monsanto) was used, with distilled water (200 L.ha<sup>-1</sup>).

Plants were sprayed with a compressed-air experimental sprayer at constant pressure of 2,5 kg m<sup>-2</sup> with flat nozzles. Biomass and %control was measured at 23 DAT.

Regression analyses following the model proposed by Seefeldt et al (1995) were used to calculate the GR<sub>50</sub> values.

## Results and discussion

The ANOVA results indicated that sampling location was a significant source of variation in both experimental environments (growth chamber and glasshouse).

There were populations in which the biomass of treated plants was about 25% of the control (those populations were called “potentially sensitive”) and there were other populations in which the biomass of the treated plants was only 75% of the control (and were called “potentially resistant”). In addition, there were a number of intermediate responses revealing a gradual transition in tolerance between the “sensitive” and the “resistant” populations.

Location was the most important variance factor; however, significant differences between progenies of the same location were also found

The regression curves (Figure 1) clearly indicate differences in the response of the sensitive (04-033) when compared with the other 5 “potentially resistant” populations. The most resistant population was 04-038 with a GR<sub>50</sub> value of about 10 fold the value of the sensitive population (04-033) (Figure 1).

The results of the repeated experiment under controlled conditions confirm glyphosate resistance of population 04-038, with a resistant factor of 15.2 (Figure 2).

At 23 DAT, glyphosate rates of 1.12 and 2.23 kg ai.ha<sup>-1</sup> provided 100% control of the sensitive population (04-033) and 0% control of the resistant.

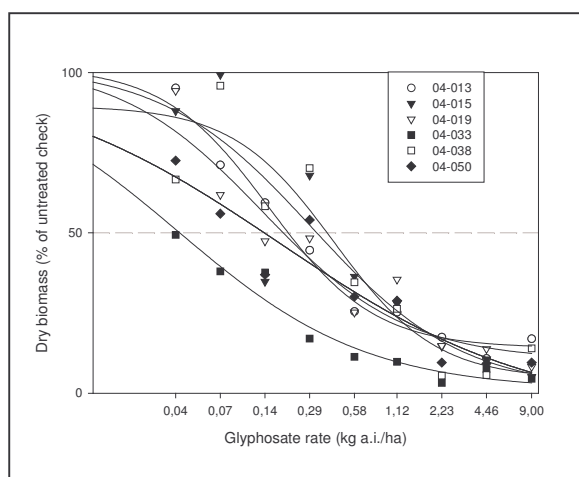


Figure 1. Dose-response experiment under net house conditions

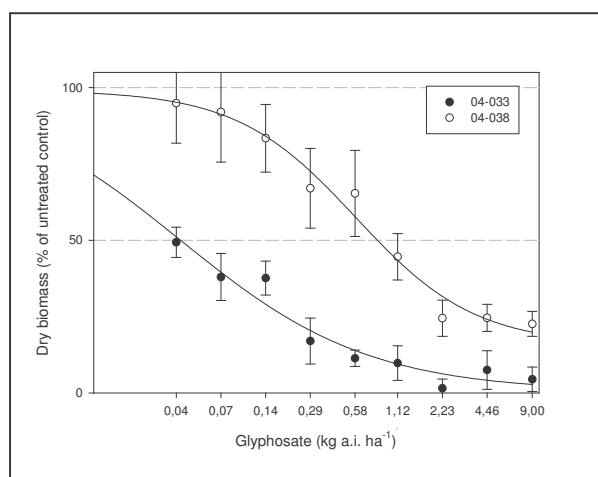


Figure 2. Dose-response experiment under controlled conditions

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# Effect of glyphosate on the reproductive biology of glyphosate-resistant cotton and corn

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## Introduction

Glyphosate-resistant (GR) crops, such as cotton and corn, allow growers greater flexibility in the timing of herbicide applications, as well as a broader spectrum of weed control than that offered by other herbicide systems. Since the commercialisation of GR cotton in 1997, however, concern over the reproductive tolerance of GR cotton to glyphosate has been raised. Numerous reports of increased boll abscission and pollination problems in response to glyphosate applications have occurred in cotton, occasionally leading to yield loss and modified fruiting pattern.

## Materials and methods

A number of experimental methodologies were used to conduct the field, greenhouse, and laboratory investigations. Details on these methodologies are provided in the following papers (Pline *et al.* 2002, 2003; Thomas *et al.* 2004).

## Results and discussion

Cotton: Anatomical studies were conducted to characterise the effect of glyphosate treatments on the development of male and female reproductive organs of cotton flowers at anthesis. In comparison with non-treated plants, glyphosate applied at both the four-leaf stage post-emergence (POST) and at the eight-leaf stage POST directed inhibited the elongation of the staminal column and filament, which increased the distance from the anthers to the receptive stigma tip by 4.9 to 5.7 mm during the first week of flowering. The increased distance from the anthers to the stigma resulted in 42% less pollen deposited on stigmas of glyphosate-treated plants than in non-treated plants. Moreover, pollen from glyphosate-treated plants showed numerous morphological abnormalities. Transmission electron microscopy showed the presence of large vacuoles, numerous starch grains, and less organized pockets of the endoplasmic reticulum containing fewer ribosomes in pollen from glyphosate-treated plants than from non-treated plants. Pollen development in glyphosate-treated plants is likely inhibited or aborted at the vacuolated microspore and vacuolated microgamete stages of microgametogenesis, resulting in immature pollen at anthesis. Although stigmas from glyphosate-treated plants were 1.2 to 1.4 mm longer than those from non-treated plants, no other anatomical differences in stigmas were visibly evident. The presence of the GR 5-enolpyruvylshikimate-3-phosphate synthase (CP4-EPSPS) enzyme from *Agrobacterium* sp. strain CP4 was quantified in reproductive and vegetative tissues using enzyme-linked immunosorbent assay. The content of CP4-EPSPS in the stigma, anther, preanthesis floral bud (square), and flower petals was significantly less than that in the vegetative leaf tissue. Glyphosate effects on the male reproductive development resulting in poor pollen deposition on the stigma, as well as the production of aborted pollen with reduced pollen viability provide a likely explanation for reports of increased boll abortion and pollination problems in glyphosate-treated GR cotton (Pline *et al.* 2002).

Additional studies were conducted to determine whether both stamen and pistil are affected by glyphosate treatments by measuring seed set from reciprocal reproductive crosses made between GR treated, untreated GR, and conventional non-transgenic cotton. Pollen viability was 51 and 38% lower for the first and second week of flowering, respectively, in GR plants

treated with a four-leaf post-emergence (POST) and an eight-leaf POST-directed treatment of glyphosate than in GR plants that were not treated. Seed set per boll was significantly reduced when the pollen donor parent was glyphosate treated vs. non-treated for the first 2 wk of flowering. There were no significant differences between treatments applied to male parents as measured by seed set at weeks 3 and 4 of flowering. Seed set was not influenced by glyphosate treatments applied to female parents at any time. Retention of bolls resulting from crosses was reduced by glyphosate treatment of male parents during the first and third week of flowering, but was not affected by glyphosate treatment of female parents (Pline et al. 2003).

Corn: Experiments were conducted in the North Carolina State University Phytotron greenhouse and field locations in Clayton, Rocky Mount, and Lewiston-Woodville, NC, in 2002 to determine the effect of glyphosate on pollen viability and seed set in GR corn. Varieties representing both currently commercial corn events, GA21 and NK603, were used in phytotron and field studies. All glyphosate treatments were applied at 1.12 kg ai.ha<sup>-1</sup> at various growth stages. Regardless of hybrid, pollen viability was reduced in phytotron and field studies with glyphosate treatments applied at the V6 stage or later. Scanning electron microscopy of pollen from affected treatments showed distinct morphological alternations correlating with reduced pollen viability as determined by Alexander stain. Transmission electron microscopy showed pollen anatomy alterations including large vacuoles and lower starch accumulation with these same glyphosate treatments. Although pollen viability and pollen production were reduced in glyphosate treatments after V6, no effect on kernel set or yield was found among any of the reciprocal crosses in the phytotron or field studies. There were also no yield differences among any of the hand self-pollinated (non-treated male X non-treated female, etc). crosses. Using enzyme-linked immunosorbent assay to examine CP4-5-enolpyruvylshikimate-3-phosphate synthase expression in DKC 64-10RR (NK03) at anthesis, we found the highest expression in pollen with progressively less in brace roots, ear leaf, anthers, roots, ovaries, silks, stem, flag leaf, and husk (Thomas et al. 2004).

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# Glyphosate-induced male sterility in RR cotton is associated with changes in auxin production in anthers

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## Introduction

Glyphosate applied over the top to transgenic, glyphosate-resistant (Roundup Ready<sup>®</sup>, RR) cotton, at the recommended rate and timing, is safe to crop and provides excellent annual and perennial weed control. However, off-label "late" treatment with glyphosate (1.44 kg ae ha<sup>-1</sup>) applied beyond the 5<sup>th</sup> leaf stage to RR-cotton (DP5415RR), may result in yield reduction due to male sterility (Yasuor and Rubin 2001). Although no vegetative damage has been observed when glyphosate applied "late", flowers exhibited various levels of male sterility. Injured flowers exhibited various levels of deformed anthers and degenerated pollen grains, while female organs were functional. Bolls developed from injured flowers were deformed, beaked or moon shaped. Glyphosate treatment resulted in a temperature-dependent male sterility; anthers developed at high temperature (34/28°C day/night) or under summer field conditions did not dehisce and did not release any pollen whereas normal anther dehiscence was observed in anthers developed under moderate temperature (28/22°C day/night) (Yasuor *et al.*, 2003).

## Materials and methods

The orientation of the microtubules in the anther endothecium cells was examined using immuno-fluorescent staining with specific anti- $\alpha$ -tubulin antibodies (DM1A). *Arabidopsis thaliana* seedlings expressing GFP-tubulin (TUA6) were dipped in glyphosate (10mM) for 5 sec and the cortical microtubules of the hypocotyl epidermis were examined using a confocal microscope. Auxin was extracted and isolate from cotton anthers according to Thompson *et al.*, 1981, and quantitatively determined 1 day before anthesis by a radioimmunoassay (RIA) and GC-MS (Roessner-Tunali *et al.*, 2003).

## Results and discussion

Glyphosate applied at the 8<sup>th</sup> leaf stage resulted in male sterile flowers with indehiscent anthers and nonviable pollen grain. Microscopic observations have shown that anther dehiscence was apparently prevented due to changes in the orientation of the cell wall thickening (CWT) of the endothecium from a longitudinal to a transverse position. The observed change in CWT orientation was associated with a parallel modification in the orientation of the microtubules (Fig. 1). Similar symptoms were observed in non-transgenic cotton plants treated with low rate of glyphosate (0.29 kg ae ha<sup>-1</sup>) applied at the 8<sup>th</sup> leaf stage (Fig. 1).

In order to better understand the phenomenon, glyphosate was applied to *Arabidopsis thaliana* seedlings expressing GFP-tubulin (TUA6). The orientation of microtubules in untreated *Arabidopsis* plants was mostly longitudinal (normal), whereas in glyphosate-treated plants the orientation was rapidly (9 h after treatment) changed to either oblique or transverse position (Fig. 1). In addition, *Arabidopsis* treated plants accumulated high concentrations of shikimic acid (>2mg g<sup>-1</sup> fresh weight), indicating a specific effect of glyphosate.

In spite of the severe damage caused by glyphosate treatments in cotton anthers and *Arabidopsis* seedlings, no effect was observed in the of ethylene level. On the other hand, glyphosate-treated cotton anthers one day before anthesis accumulated ten times more auxin as compared to the



untreated anthers (Fig. 2). These results might suggest that male sterility induced by glyphosate in RR cotton is associated with changes in auxin production leading to modifications in CWT and microtubules orientation.

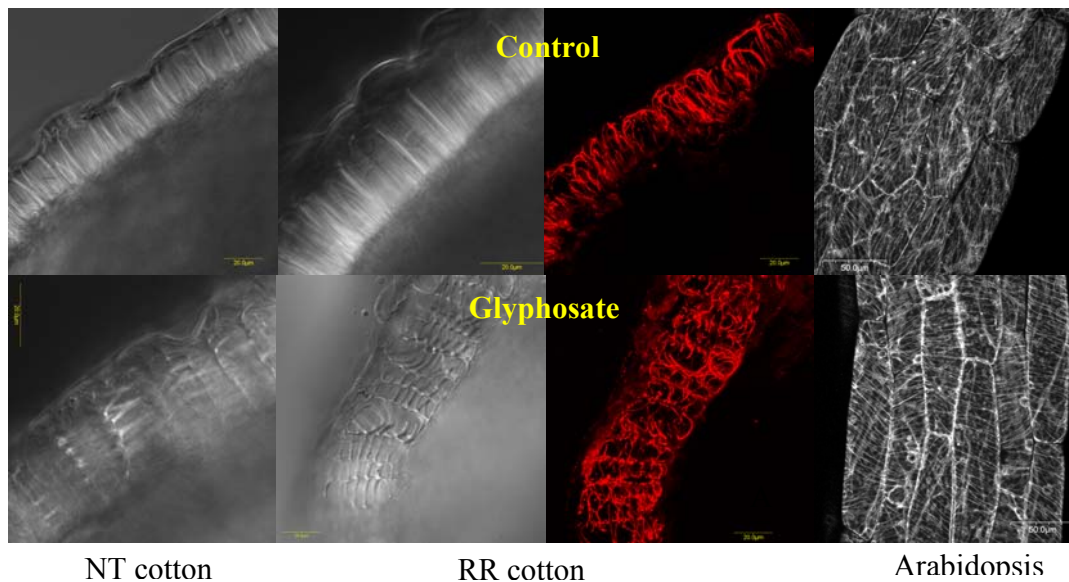


Figure 1. Immunofluorescent staining of  $\alpha$ -tubulin in RR cotton endothecium cells and GFP-tubulin (TUA6) in epidermal cells of *Arabidopsis thaliana* hypocotyl. Cotton anthers were collected before pollen dispersal. Cortical microtubules of 10 days *Arabidopsis* hypocotyls were observed 24 h after glyphosate (10mM) treatment. NT=Non-transgenic cotton treated with glyphosate (0.29 kg ae ha<sup>-1</sup>). RR=glyphosate resistant cotton treated with glyphosate (1.44 kg ae ah<sup>-1</sup>).

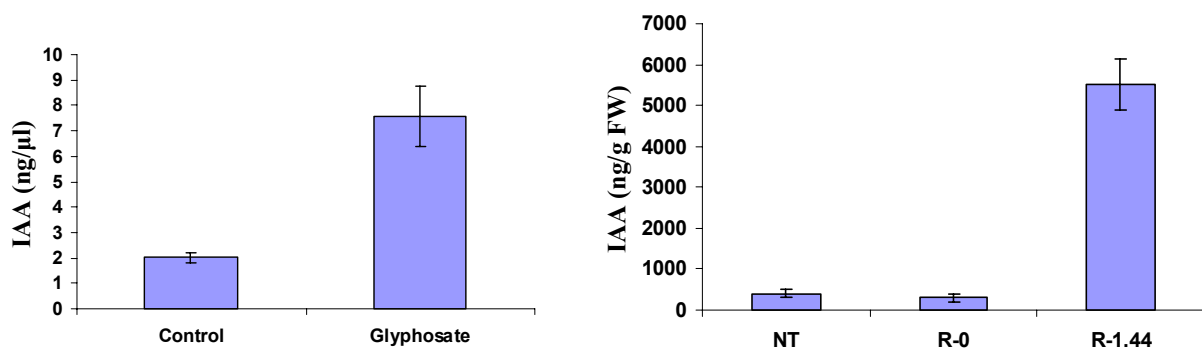


Figure 2. IAA accumulation in cotton anthers as determined by radioimmunoassay (A), and GC-MS analysis (B). Glyphosate (1.44 kg ae ha<sup>-1</sup>) was applied at the 8<sup>th</sup> leaf growth stage; anthers were harvested and extracted 1 day before anthesis. NT=Non-transgenic, R-0= untreated RR cotton; R-1.44 = RR cotton treated with glyphosate (1.44 kg ae ha<sup>-1</sup>).

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## **Evaluation of weed dynamics in different cropping systems**

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### **ABSTRACT**

Manipulation of cropping systems in order to improve weed management requires a better understanding of the effect of crop and soil related factors on weed life cycles and weed population dynamic. Weed population and composition in the field and also vertical distribution of weed seed bank in winter wheat in monoculture and in rotation with sugar-beet and maize were studied. Weed seed densities in organic and integrated cropping systems were higher than conventional and high-input cropping systems. In low-input systems seeds of annual and perennial grasses were dominant. In high-input systems perennial weeds were less frequent. Seed density in continuous winter wheat was higher than when winter wheat was planted in rotation with other crops. The results showed that crop species and crop management practices influenced on weed seed bank and weed population.

A multi-method approach to the study of the host specificity of *Ceratapion basicorne* Illiger (Coleoptera: Curculionidae), candidate biocontrol agent of yellow starthistle, *Centaurea solstitialis* L. (Asteraceae).

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*Centaurea solstitialis* L. (Asteraceae) is an invasive Eurasian weed introduced in the USA at the end of eighteenth century. According to recent reports, the weed infests an area of more than 3,7 million hectares and is considered to be one of the most economically important weeds in California and in other Western United States

A biological control program on yellow starthistle was started in mid 19<sup>th</sup> century; as a result 5 species of seed feeding insects have been released in the US in the last 40 years. Despite the annual biology of the plant and the high infestation rates recorded in the flower heads of the target weed for most of the species released, only recently occasional decline of the weed population density was observed. For this reason, starting from 2000 more effort has been put in the search, selection and evaluation of biocontrol candidates attacking *C. solstitialis* at the root and/or stem level at early phenological stages. Among them, high priority has been given to Turkish populations of the crown boring weevil *Ceratapion basicorne*.

The weevil is univoltine, attacks the weed at the rosette or early bolting stage and showed relevant impact when several larvae are found in a single plant. The presence of closely related, polyphagous species was recorded in sympatric conditions in the areas of study.

This work describes the importance to combine the classical approach (biological observations and host range laboratory and field tests) with genetic analyses as an important tool for the successful discrimination of the target insect species at the population level.

## **Evaluation of fungi for biological control of Canada thistle (*Cirsium arvense* L. (Scop.))**

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*Cirsium arvense* (L.) Scop. is an economically important weed that causes serious losses to agriculture in the temperate areas of North America. Commonly known as Canada thistle, this weed is difficult to control because of its biology, competitive nature, and persistence. Foliar bioherbicides appear to only provide temporary suppression of shoots, so a strategy was developed to seek fungi that could attack the rhizomes and prevent shoot emergence. Surveys were conducted from 1994-2003 to collect diseased Canada thistle plants. Fungal pathogens were isolated and screened using the inoculum mat bioassay under greenhouse conditions, which applies the fungi to soil in pots containing root pieces of Canada thistle prior to shoot emergence. A total of 368 isolates were evaluated and 34 were selected because they significantly reduced root growth, shoot emergence, or foliar biomass relative to untreated controls. Some isolates demonstrated strong inhibition of root growth, shoot emergence, and foliar development. Other isolates were less effective at reducing root growth, but reduced shoot emergence and foliar biomass. Eleven other isolates were found to enhance root growth and foliar biomass relative to untreated controls. Further evaluation and identification of these isolates is underway.

**Release of the ragweed leaf beetle *Zygogramma suturalis* F. (Coleoptera, Chrysomelidae) into Russia for biological control of common ragweed, *Ambrosia artemisiifolia* L. (Asteraceae)**

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The ragweed leaf beetle, *Zygogramma suturalis* F. has been introduced to Russia in 1978 against the common ragweed, *Ambrosia artemisiifolia* L. This project was the pioneering for classical biocontrol of weeds in crop rotation agroecosystems. The ragweed leaf beetle successfully acclimated and in 1983-85 it was able to suppress ragweed in release site (old field heavily infested with ragweed) and several neighboring fields, where the population density of the beneficial herbivore was sufficiently high. In certain cases, local overpopulation up to 5000 adults per m<sup>2</sup> was recorded. This allows one to expect highly efficient biological control of the weed.

By 1988 the ragweed leaf beetle has spread over hundreds of surrounding fields. However, visual estimations periodically conducted during 1988-2004 over a large area (up to 25000 hectares) around the first release site showed relatively low mean herbivore density, despite few patches with the insect overcrowding and local weed extermination.

Although ragweed leaf beetle does not significantly influence ragweed density in most of crop rotation agroecosystems, it still can cause serious damage to the weed under certain conditions, particularly in stable and less disturbed locations, e.g. in fields of perennial fodder legumes or within patches of high *Z. suturalis* density, which are usually located in field margins. Having regard to the spectacular success achieved at the release site and neighboring fields, this suggests that field nurseries, i.e. uncultivated sites grown with ragweed, could be a promising method of *Z. suturalis* propagation.

## Development of High Density Energy Techniques in Robotic Weeding

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Nowadays the agricultural sector requires non-chemical weed control that ensures food safety. Consumers demand high quality food products and pay special attention to food safety. Through the technical development of high-density energy techniques for weed control, such as thermal treatment of weeds, it might be possible to control weeds in a way that meets consumer and environmental demands. The plants used were white mustard (*sinapsis alba L*) and they were grown in a glasshouse. They exposed to our treatments in the early two-leaf stage. The size of the leaf area was almost equal with 3 cm<sup>2</sup> per plant. Two application and ignition methods were used drop and then fire (leaf applied) and liquid at the bottom of the plant (soil applied) with acetone and gasoline. The effects were assessed 10 days after exposure, as dead, alive, and suffering plants according to the colour of the leaf tissue. The dose necessary to achieve 50% reduction for the leaf applied method was 0.013958 gr/plant for acetone and 0.01168 gr/plant for gasoline. For 90% reduction the required dose was 0.034 gr/plant for acetone and 0.030 gr/plant for gasoline. So, the dose necessary to achieve 95% reduction was 0.046 gr/plant for acetone and 0.041 gr/plant for gasoline. The dose necessary to achieve 50% reduction for the soil applied method was 0.01268 gr/plant for acetone and 0.00446 gr/plant for gasoline. For 90% reduction the required dose was 0.039 gr/plant for acetone and 0.012 gr/plant for gasoline. Hence, the dose necessary to achieve 95% reduction was 0.055 gr/plant for acetone and 0.017 gr/plant for gasoline. Together these results suggest that with the utilization of high-density energy techniques it might be feasible for precision weed control without the use of chemicals, especially for selective application.

*Key words: Precision, weeds, non-chemical, selectivity*



## **Precision mechanical weed control**

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Current weed control problems in organic farming show that alternatives for selective herbicides need further development. Mechanical control methods such as weed hoeing are attractive because of the high capacity, wide applicability and low cost. However, the variable effectiveness and limited selectivity at early crop growth stages are major limitations for reliable weed control. The aim of this study is to investigate the factors that influence the design of soil engaging systems to mechanically control weeds between plants within the crop row in widely spaced field vegetables. A mass flow soil dynamics model based on particle dynamics is anticipated to be developed to aid designers in determining the lateral and forward displacement of soil as it is undercut by shallow working implements. To maximize the treated area through soil displacement, laboratory experiments will be carried out to identify and quantify the factors influencing forward and lateral displacement. Investigations into the effect of the implement's geometry over a range of velocities from 1 to 10 km/h in sandy loam soil at densities of 1300 to 1500 kg/m<sup>3</sup> will be carried out at the soil bin facility of Cranfield University, Silsoe.

*Key words: Weeds, Precision farming, Organic, Vegetables, Hoeing*

## The effect of nozzle size, water pressure and nozzle height on dispersal of *Marchantia polymorpha* gemmae, using an overhead sprinkler system

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Liverworts (*Marchantia polymorpha*) growing on the surface of container plants are a major problem in nurseries, estimated to cost the UK horticultural industry £13 million each year, (4% of total production costs). Many aspects of the lifecycle of liverwort are known; the aim of this experiment is to provide information on the epidemiology of infestation. *Marchantia polymorpha* reproduces by gemmae, vegetative propagules produced in circular structures (gemma cups) found on the dorsal surface of the thallus. Gemmae are released when water splashes into the cup, transporting them away from the parent plant; dispersal distances of 0.6m by small raindrops were previously recorded. The dispersal of liverwort gemmae were investigated using a glasshouse overhead sprinkler system with three different nozzle sizes controlling flow rate, four water pressures (1.5, 2, 2.5 and 3 bar) and two nozzle heights (1 and 2 metres). A preliminary experiment using red dye in place of plant material indicated that gemmae may be dispersed within splash droplets and they may be propelled by incident water drops. At the extreme water pressures (1.5 and 3 bar), generally fewer gemmae were dispersed for all nozzles. Using the nozzle with a flow rate of 160 lh<sup>-1</sup>, more gemmae were dispersed with increased nozzle heights; for the other two nozzles there was no clear effect of nozzle height on the number of gemmae dispersed. However, with the 60 lh<sup>-1</sup> flow rate nozzle, the distance travelled by gemmae increased with increased nozzle height. The maximum dispersal distance was 1.6m during this experiment.

# **Experiences and problems of weed management in organic farming in the New EU- Member States and Associated Countries**

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## **Abstract**

After the accession of the New EU-Member States in May 2004 it became obvious, that information about state of the art and frame conditions for organic farming of these countries is very insufficient. The CHANNEL project, which started in November 2004, aims to improve the knowledge about the specific situation in these countries. As one key issue within the CHANNEL project, weed management experiences and problems are covered. Data collection and interpretation will be realized by standardized thematic questionnaires and expert meetings with representatives of the involved 15 countries. Some of the first results will be presented with this paper. The project scope covers four main areas: i.) general, legal, land use and natural conditions, ii.) administrative frame conditions for weed management, iii.) expert assessments on state of weed infestation, state of management practices, experiences and limitations in weed management and iv.) scientific and educational background, overview on recent research.

It is a widespread presumption, that due to a former intensive conventional use and the short period of conversion to organic farming in the New EU-Member States, weed flora and experiences in the management are dominated by the historical conventional use. This picture needs a stronger differentiation. Even the economy of scarcity, which was more or less typical for the past in these countries, led to an enormous variation in management techniques, availability of machinery and knowledge, scientific background and support as well as legal obligations. These factors together resulted in a graduated intensity of weed regulation.

## Predicting weed seed persistence in the soil: towards rapid and reliable assessment

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Management of weed species is often complicated by the unknown status of their soil seed banks. Although a limited number of laboratory-based life expectancy tests exist, few simulate the environmental complexity of the field. Field trials, although more accurate, are time-consuming and expensive to conduct for individual species. Thus, the persistence of weed seeds can be difficult to quickly and accurately predict. This study aims to investigate some of the environmental factors affecting seed persistence in soil and relate them to seed ageing under accelerated ageing conditions simulated in the laboratory. Seeds from four weed species with contrasting physical and anatomical characteristics – broadleaf privet (*Ligustrum lucidum* W.T.Ait), balloon cotton bush (*Gomphocarpus physocarpus* E.Mey), stinking Roger (*Tagetes minuta* L.) and wild oats (*Avena sterilis* L ssp. *ludoviciana*) – were hand-collected from sites in southern Queensland. The vigour of these seeds will be tested via an accelerated ageing test (60% RH @ 45°C for 125 days) to produce an initial seed survival curve. A field plot containing three contrasting soil types has been established and seeds have been buried within tubes at 10-15 cm depth so that they experience natural ageing processes. At 3, 6, 9, 12 and 24 months following set up of the field plot, seeds will be exhumed and tested using a variety of vigour tests in the laboratory. Ultimately, our objective is to elucidate the relationship between accelerated ageing and other seed tests, and the reality of field trials. Such data will be used as a starting point for ecological models that aim to predict weed seed persistence in various environments.

## Effect of red root pigweed density and germination time on yield and yield components of maize

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The main objectives of this research were studying the effect of red root pigweed density and emergence time on yield and yield components of corn. This study was conducted in 2003 in the Research Station Farm of Azad University of Karaj, Iran. Maize cv Ksc 704 was planted at a density of 70000 plants/ha. The seeds of pigweed were planted on farrows by 10cm near two sides of corn rows. Using a completely randomized block design with 4 replicates the combinations of 4 red root pigweed density (0, 2, 3 and 5 plants/m<sup>2</sup>) and 3 emergence times (A; at the maize emergence time, B; one week after maize emergence, C; two weeks after maize emergence) were compared. At three sampling stages (T1; 2 month after crop planting, T2; 50% of silking stage, T3; physiological maturity) effects on crop growth and yield were determined. Crop biomass, grain yield, ear length, leaf and stem dry weight, kernel numbers on ear rows, row numbers on ear, unfertilized part of ear, harvest index (HI), plant height, light extinction coefficient, and light absorption were measured. Results showed that pigweed emergence times had no significant difference on maize growth and yield except in the number of seed rows per ear. Comparison of density treatments had indicated a significant difference about 1% and 5% levels on corn seed and biomass yield reduction percentage respectively. Thus existence or not existence of pigweed had the most effects on corn grain yield reduction, but 2, 3 and 5 plants/m<sup>2</sup> densities of pigweed had no considerable grain yield reduction. Pigweed density reduced corn leaf area index. Results showed a significant differences between contemporary germination and two another germination time treatments. Also grain yield decreased (1 to 3 percentage) in the treatments of Pigweed germination time. Effect of Pigweed density on the length of unfertilized part of ear, ear dry yield, biomass, numbers of kernels on row, HI, seed diameter and seed dry weight were significant (19 to 31 percent). Results showed that density treatment had highest effect on yield and yield components. Seed diameter reduction decreased 1000 kernel weight. The number of seed rows on ear were affected by germination time and density of pigweed as well.

## Control of broomrape on sunflower and tobacco crops in România

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Broomrape (*Orobanche spp.*) is one of the most dangerous parasitic plants for sunflower and tobacco crops in România.

In the sunflower fields the populations of weed broomrapes consist of plants of *Orobanche cumana* Wallr, and in the tobacco field *Orobanche ramosa* L. , *Orobanche cumana* Wallr were found and sporadically other species.

The methods of control and management for this parasite included: crop rotation, clean seeds, sowing dates, biological control, genetic resistance and herbicides.

During the last few years, on sunflower crops, this attack of parasitic *Orobanche cumana*, showed a marked extension throughout the sunflower fields, especially in the southeast area. Under artificial inoculation conditions and natural contamination behaviour of some forms of sunflower cultivars was studied during the attack of *Orobanche cumana* as well as the influence of specific herbicides on parasite fanerogame.

In tobacco, this parasitic plant causes losses both in yield and quality. In order to diminish the negative effect of broomrape on tobacco yield the chemical method was tested. In 2 experimental field of tobacco crops the efficacy of 5 variants of herbicides was tested (Treflan 48 EC + Glyforom RV, Stomp 330 EC + Glyfogan 480 SL, Treflan 48 EC + Basta EC, Stomp 330 EC + Assert 250 EC, Stomp 350 + Basta EC).

The best result was with Treflan 48 EC (trifluralin 480 g/l) preemergent 2 l/ha + Glyforom RV (glyphosate 360 g/l) post emergent 0,2 l/ha + 0,3 l/ha applied at 40 and 60 days from planting.



## Study of phytotoxic activity of cultural filtrate of *Stagonospora cirsii* J.J. Davis

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The cultural filtrate of the fungus, *Stagonospora cirsii*, a foliar pathogen of *Cirsium arvense*, demonstrated the phytotoxicity on detached leaves and seedling roots of the weed. This research work was focused on study of cultural conditions to obtain highly toxic culture filtrates of *Stagonospora cirsii*, isolation and characterization of phytotoxic substances. The nutrient media for toxic production of *S. cirsii* was optimized on the base of modified Czapek media. Concentration of carbon and nitrogen affected on production of toxic metabolites of *Stagonospora cirsii* at higher extend than C/N ratio. Two peaks of toxic activity were observed during cultivation of the fungus on the optimized liquid medium: in the beginning of cultivation and at the stationary phase of growth. The highest level of toxic activity of culture filtrate was observed on 10-12 days at static culture. The low fungicidal activity and moderate zootoxic activity (test on *Paramecium caudatum*) was found for the culture filtrate. High phytotoxic effect of the filtrate was restricted to the host plant. The method of purification of a phytotoxin from cultural filtrate was proposed. It includes extraction with diethyl ether, gradient chromatography (system: hexane/diethyl ether) in column with silica gel 60, and assessment of the purity of a phytotoxic substance by HPLC. Presumable chemical structure of the phytotoxin of *S. cirsii* is very similar to putaminoxin of *Phoma putaminum* (Evidente et al., 1995).

## **Influence of nitrogen and herbicide application in corn on weed community**

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To study the effect of nitrogen rate and herbicide application in corn on the weed community, an experiment was conducted in 2001 at Lods Agronomy Research Center, McGill University, on a clay soil. A split-plot design in 6 replications was used. Herbicides (4 levels) and nitrogen (3 levels) were allocated to main and sub plots, respectively. Corn seeds were planted on May 10. The size of the plots was 10 × 10 m. Row spacing was 75 cm and corn population was based on 76,000 plants ha<sup>-1</sup>. Nicosulfuron (Ultim, 9 g ha<sup>-1</sup>) and mineral oil (Agral 90, 25% v/v) were used to control grassy weeds. Dicamba (Banvel II, 1.2 L ha<sup>-1</sup>) was used for broadleaf control. A mixture of Ultim plus Agral used at above rates, and Banvel II at a rate of 0.6 L ha<sup>-1</sup> was used to control both broad leaved and grassy weeds. Weedy check with no herbicide application was also present. Nitrogen (ammonium nitrate) was split and applied twice (at planting time and on July 3) at rates of 60, 120 and 250 kg ha<sup>-1</sup>, respectively. Our results indicated that nitrogen rate had no significant effect on number, percent coverage or dry weight of both broad leaved and grassy weed species. Herbicides did have a significant effect on both types of weed species, especially at the final evaluation (July 27). Grassy weed species were not completely controlled, but broad leaved species were controlled effectively. It was concluded that nicosulfuron and the mixture of nicosulfuron and dicamba were the best treatments for control of grassy and broad leaved weeds, respectively.

## **Effects of salinity, wheat density and wild oat density on yield and yield components of wheat**

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In 2003, a field experiment was conducted to study effects of salinity, wheat density and wild oat density on yield and yield components of wheat. The experimental design was a factorial split plot in which salinity was used as main plot (represented by three levels; 1.5, 5.5 and 9.5 dS m<sup>-1</sup>) and combinations of wheat density (two levels; 400 and 600 plants m<sup>-2</sup>) and wild oat density (three levels; 0, 80 and 160 plants m<sup>-2</sup>) were used as sub plot. Results showed that an increased wheat density caused an increased wheat yield mainly through an increased number of spikes per square meter. Wild oat caused reductions in wheat yield and this reduction increased with increasing densities of wild oat. Grain yield reduction at the highest salinity level was lower than at the other salinity levels. Interaction effects indicated that by increasing the wheat density up to 600 plants m<sup>-2</sup>, effects of salinity and wild oat competition on grain yield reduced.



**BARI, Italy, June 2005**

## Programme

### Saturday 18 June

09:00 - 18:00 EWRS Scientific Committee Meeting

### Sunday 19 June

09:00 - 18:00 EWRS Executive Committee Meeting

09:00 - 17:00 Joint IBG-EWRS Workshop

17:00 - 20:00 *Registration & posters put up*

18:00 - 23:00 Welcome drink – Puglia wine and food tasting

### Monday 20 June

08:00 - 09:00 *Registration & posters put up*

#### Opening (Chair: Bob Froud-Williams, UK)

09:00 - 09:10 Welcome address 1 - Rector/Dean University of Bari

09:10 - 09:20 Welcome address 2 - Chairman of LoCom

09:20 - 09:30 Welcome address 3- EWRS President

09:30 - 10:00 Andrée CARTER (UK) - Allan Walker's plenary lecture

10:00 - 10:10 Discussion AW's plenary lecture

10:10 - 10:40 *Coffee break*

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### Session 1 Biodiversity, invasive species and biological weed control

(Chair: Paul Hatcher, UK & Jon Marshall, UK)

10:40 - 11:10 **Andy SHEPPARD (FR)** - Keynote paper: Classical biological control of European exotic environmental weeds: opportunities and constraints

11:10 - 11:30 **Heinz MUELLER-SCHÄRER (CH)** - Evolution in invasive plants and implications for biological control

11:30 - 11:50 **Emmanuel YAMOAH (NZ)** - Potential vectoring of *Fusarium tumidum* spores by four insect species for biological control of gorse

11:50 - 12:10 **Giorgio RAGAGLINI (IT)** - Field-scale interactions between weeds and *Aphis fabae* Scop. (homoptera: aphididae) in conventional and low-input sugarbeet

12:10 - 12:30 **Santiago Luis POGGIO (AR)** - Spatial patterns of plant species richness in relation to distance from crop field margins in the Rolling Pampa (Argentina)

12:30 - 12:50 **Richard Hull (UK)** - The potential for managing weeds to minimise impacts on yields and enhance the ecological diversity of arable fields – a winter wheat case study

12:50 - 13:10 Discussion  
13:10 - 14:30 Lunch  
14:30 - 15:30 Poster session

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## Session 2 Weed ecology and community dynamics

(Chair: Andrea Grundy, UK & Bob Froud-Williams, UK)

15:30 - 16:00 **Niels HOLST (DK)** - Keynote paper: Field weed population models: a review of approaches and application domains

16:00 - 16:20 **Arnd VERSCHWELE (DE)** - Effects of organic farming on population dynamics of weeds - long-term results from a site in Northern Germany

16:20 - 16:40 **Pavel SASKA (CZ)** - Seed predation by carabids in organic wheat fields in the Netherlands

16:40 - 17:10 *Coffee break*

17:10 - 17:30 **Peter LUTMAN (UK)** - Studies on the seed production and germination characteristics of *Poa annua*, *Senecio vulgaris*, *Stellaria media* and *Tripleurospermum inodorum*

17:30 - 17:50 **Andrea GRUNDY (UK)** - Long-term changes in the weed populations resulting from strategies for restricted herbicide use in field vegetable systems

17:50 - 18:10 **Nathalie Colbach (France)** - **ALOMYSYS: a model of the effect of cropping systems on weed demography. Example of blackgrass (*Alopecurus myosuroides* Huds.)**

18:10 - 18:30 Discussion

18:30 - 20:00 **Working Groups meetings:**

- Crop-Weed Interactions
- Physical and Cultural Weed Control

Meeting of Weed Research Editorial Board

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## Tuesday 21 June

08:00 - 09:00 *Registration & poster exhibition*

## Session 3 Weed biology and crop/weed interactions

(Chair: Lammert Bastiaans, NL & Francesco Tei, IT)

09:00 - 09:30 **Nicolas DELABAYS (CH)** - Keynote paper: Allelopathy: from field evidences to agronomic utilizations

09:30 - 09:50 **Laila KARLSSON (SE)** - Stepwise rather than cyclic dormancy changes explain emergence of the summer annuals *Fumaria officinalis* and *Galeopsis speciosa*

09:50 - 10:10 **Jack DEKKER (US)** - Patterns in *Setaria* seedling emergence

10:10 - 10:40 *Coffee break*

10:40 - 11:00 **Bärbel GEROWITT (DE)** - Driving forces for the development of *Cirsium arvense* in arable farming systems

11:00 - 11:20 **Jens PETERSEN (DK)** - Inter- and intra-species competition for applied nitrogen

11:20 - 11:40 **Lynn COLLINGS (UK)** - Testing a simple approach to estimating the seed production of arable weeds within contrasting crops

11:40 - 12:00 Discussion

12:00 - 13:00 **EWRS General Assembly** (Chair: Bob Froud-Williams)

13:15 *Buses leave to excursion (lunch at a winery)*

13:15 - 19:30 *Excursion*  
21:00 *Symposium dinner*  
24:00 *Back to hotels*

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## Wednesday 22 June

08:00 - 09:00 *Registration & poster exhibition*

### Session 4 Chemical weed management and optimisation of herbicide technology

(Chair: Per Kudsk, DK & Marco Quadranti, CH)

09:00 - 09:30 **Willy T. RUEEGG (CH)** - Keynote paper: Herbicide development - challenges and opportunities  
09:30 - 09:50 **Nicolas MUNIER-JOLAIN (FR)** - Decid'herb, a decision support system on the WEB, designed for sustainable weed management in cultivated fields  
09:50 - 10:10 **Jan NETLAND (NO)** - Evaluation of the weed model in the Danish decision support system "Plant Protection Online" adapted to Norwegian conditions  
10:10 - 10:40 *Coffee break*  
10:40 - 11:00 **Christian RITZ (DK)** - Analysis of Bioassay  
11:00 - 11:20 **Zenon WOZNICA (PL)** - Extremely reduced herbicide rates applied with adjuvants for weed control in sugar beets  
11:20 - 11:40 **Urs HOFER (CH)** - Challenges and solutions for grass weed management in small grain cereals in Europe  
11:40 - 12:00 Discussion  
12:00 - 13:00 Poster session  
13:00 - 14:30 *Lunch*

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### Session 5 Non-chemical approaches to weed management

(Chair: Bo Melander, DK & Paolo Bàrberi, IT)

14:30 - 15:00 **Rommie VAN DER WEIDE (NL)** - Keynote paper: Innovation in mechanical weed control in crop rows  
15:00 - 15:20 **Jerry A. IVANY (CA)** - More effective weed control methods in potatoes (*Solanum tuberosum* L.)  
15:20 - 15:40 **Andrea PERUZZI (IT)** - The rotating rolls harrow: a new operative machine for physical weed control in pre-sowing and post-emergence  
15:40 - 16:10 *Coffee break*  
16:10 - 16:30 **Fabio FAUSTINI (IT)** - Organically grown durum wheat (*Triticum durum* Desf.) varieties under different intensity of mechanical weed control  
16:30 - 16:50 **Milton McGIFFEN (US)** - Competitiveness of erect, semi-erect, and prostrate cowpea (*Vigna unguiculata*) genotypes with sunflower and purslane  
16:50 - 17:10 **Kico DHIMA (GR)** - Annual grass weed control in cotton by cereal cover crop mulches  
17:10 - 17:30 Discussion  
17:30 - 19:00 **Working Groups meetings:**

- Biological control of weeds
- Germination and early growth
- Optimisation of herbicide dose



- Weed management systems in vegetables
- Weeds and Biodiversity

20:00 Concert

## Thursday 23 June

08:00 - 09:00 *Registration & poster exhibition*

### Session 6 Cutting-edge methodologies and site-specific weed management (Chair: Svend Christensen, DK & Cesar Fernandez-Quintanilla, ES)

- 09:00 - 09:30 **Roland GERHARDS (DE)** - Keynote paper: Practical experiences with a system for site-specific weed control using real-time image analysis and GPS-controlled patch spraying (TURBO)
- 09:30 - 09:50 **Louis ASSEMAT (FR)** - A practical software to compute early weed competition from cover images analysis
- 09:50 - 10:10 **Sanne HEIJTING (NL)** - Species specific spatial relations between weed patterns and soil characteristics; a three year study
- 10:10 - 10:40 *Coffee break*
- 10:40 - 11:00 **Corné KEMPENAAR (NL)** - Adjustment of doses of potato haulm killing herbicides by using reflection and fluorescence sensing techniques (MLHD applied on leaf desiccants)
- 11:00 - 11:20 **Martin PILAR (ES)** - Discrimination of grassweeds in winter cereal crops by radiospectrometry
- 11:20 - 11:40 **Karel KLEM (CZ)** - Weed detection using chlorophyll fluorescence imaging and artificial neural network
- 11:40 - 12:00 Discussion

### Session 7 Herbicide resistant weeds and crops (Chair: Jean-Pierre Claude, FR and Anne Thompson, UK)

- 12:00 - 12:30 **Baruch RUBIN (IL)** - Keynote paper: The impact of transgenic herbicide-resistant crops on herbicide-resistant weeds and the environment: a review
- 12:30 - 13:00 Steve POWLES (AU) - Keynote paper: Herbicide resistance from detection to management: the experience in Australia
- 13:00 - 14:30 *Lunch*
- 14:30 - 14:50 **José Maria URBANO (ES)** - Screening for biotypes of *Conyza bonariensis* resistant to glyphosate in southern Spain
- 14:50 - 15:10 **Giovanni DINELLI (IT)** - Physiological and molecular mechanisms of glyphosate resistance in *Conyza canadensis*
- 15:10 - 15:30 **Maurizio SATTIN (IT)** - An Italian population of *Amaranthus retroflexus* resistant to ALS-inhibiting herbicides: molecular basis and resistance pattern
- 15:30 - 15:50 **Christophe DELYE (FR)** - To better understand how target-site based resistance is selected and spreads within and between weed populations
- 15:50 - 16:10 **Jordi RECASENS (ES)** - The effect of different cultural methods on a herbicide-resistant corn poppy
- 16:10 - 16:30 Discussion
- 16:30 - 17:00 *Coffee break*

## **Final Session** (Chair: Bob Froud-Williams, UK)

17:00 - 17:20 Summary of the scientific programme (Paolo Bàrberi, EWRS Scientific Secretary)

17:20 - 17:30 Concluding remarks (Bob Froud-Williams, EWRS President)

17:30 ***End of Symposium***

17:30 - 20:00 **Working Group meeting:**

- Herbicide resistance Working Group (continues on Friday 24 June, 09:00 to 13:00)



# European Weed Research Society



## 13<sup>TH</sup> EWRS SYMPOSIUM

<http://www.ewrs-symposium.com>



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