

S12

GENETIC RESOURCES: NEW TOOLS FOR THE CONSERVATION AND MANAGEMENT OF GENETIC RESOURCES IN HORTICULTURE

S12.001

How Climate Change Is Already Affecting the Conservation of Fruit Diversity

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From the deserts of Central Asia, through the Middle East, across North Africa and into the Americas, orchard keepers are already reporting the failure of diverse fruit tree crops due to the effects of global warming and urban heat islands. In some fruit-growing regions such as the Central Valley of California, the percent of the landscape suitable to apple production has decreased from 50 percent in 1950, to less than 4 percent today, and will likely be entirely lost by 2050. For grapes, it is projected that 80 percent of the varieties grown today under optimum conditions for wine production will be forced to move to more northerly or higher elevations, with devastating consequences to the wine industry. Even on the Arabian Peninsula, the Pamirs and the Ti'en Shan, where certain heritage fruit land races have been grown in the same places for millennia, a reduction in chill hours will soon force a shift in varieties if fruit production is to continue at all. Although it is certainly possible to breed for low-chill tolerant fruits, chilling hours are not the only factors affected by global climate change. The spread of newly-introduced diseases and pests, the increasing frequency of tropical storms and devastating droughts, are already affecting the conservation and production of diverse fruits. Screening the diversity of extant fruit varieties for low-chill requirements, drought and salinity tolerance and pest or disease resistance may be far more cost effective than breeding alone; both need to be advanced. This work cannot be done by germplasm repositories and plant breeding stations alone; participatory on-farm screening, selection and breeding should be encouraged, and collaborative conservation embraced. A few success stories will hopefully inspire more scientists and farmers to take up this challenge, so that we will not be victims but participants in adaptation.

S12.002

The Genebank of the Institute for the Conservation and Improvement of the Agrobiodiversity (COMAV) of the Polytechnic University of Valencia

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The genebank of the Institute for the Conservation and Improvement of the Agrobiodiversity (COMAV) of the Polytechnic University of Valencia started its activities in the early eighties. Today, the genebank holds more than 10.000 accessions of horticultural crops, mainly those belonging to the *Solanaceae* (nearly 4000 accessions) and *Cucurbitaceae* (nearly 2000 accessions) families. The *Brassicaceae* include more than 500 accessions and more than 800 belong to *Lactuca sativa* and related species. The total number of species is 115. Accessions are stored at 4 °C after previous dissection with silicagel. Cold chambers at -18 °C are also available. Most part of the collection comes from Spain, but many accessions from Latin America, Europe, Asia and Africa are also included in the collection. The COMAV coordinates the regeneration and characterization activities on vegetable genetic resources in Spain. Additionally, the COMAV coordinates the genetic resources of cucurbit crops in the European Cooperative Programme on Genetic Resources, ECPGR. Regeneration, morphological and molecular characterization and evaluation activi-

ties are conducted with the materials stored in the genebank by the research laboratories of the COMAV. Core collections have been developed for tomato, pepper and *Cucurbita* spp. crops. Activities on evaluation are performed mainly in solanaceous and cucurbits crops and deal with search for resistance to biotic stresses, mainly viral and fungi pathogens, breeding for quality, application of genomic tools for the management of genetic resources in melon, exploitation of natural diversity in breeding, EcoTilling and phylogenetic studies on *Solanum* genus. Breeding and exploitation of traditional varieties are also an important research line.

S12.003

Field Collection and Conservation of Wild Edible Species in the Germplasm Bank of Zaragoza (Spain)

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There is a high number of non-cultivated edible plant species. Some of them are still appreciated and consumed, in other cases their consumption is only in the memory of the elderly people and the knowledge about these species is at risk of disappearing. Within the framework of the Spanish project 'The genetic resources of edible underutilized species: field collection, multiplication and conservation in the Germplasm Bank' during 2009 various collecting expeditions across Aragón, a region of 47720 km² in the northeast of Spain, were performed with the following aims: 1) to compile information about wild species used for eating, either nowadays or in the past, 2) to locate these species within their natural habitat, 3) to classify them taxonomically, and 4) to obtain some seeds and maintain them at long-term storage conditions. To compile the information a list of descriptors based on the document 'Descriptors for farmers' knowledge of plants' (Bioversity and The Christensen Fund, 2009) has been developed. A total of 23 localities have been visited and 27 personal interviews have been carried out. Most of the informers were more over than 65. Among the documented species, a total of 89 accessions were collected, belonging to 14 families and 25 genera. All the seeds are held in the Germplasm Bank of Zaragoza at -18 °C and 4% RH. These species include: *Allium ampeloprasum* L., *Anchusa italica* Retz., *Asparagus acutifolius* L., *Bryonia dioica* Jacq., *Chondrilla juncea* L., *Eruca sativa* Mill., *Lactuca serriola* L., *Picris echioides* L., *Rorippa nasturtium-aquaticum* (L.) Hayek, *Rumex acetosa* L., *Scolymus hispanicus* L., *Scorzonera laciniata* L., *Silene vulgaris* (Moench) Garcke, *Silybum marianum* (L.) Gaertn., and *Taraxacum officinale* Weber.

S12.004

Conservation, Characterization, Evaluation and Utilization of Horticultural Genetic Resources Carried out by the Italian Agricultural Research Council

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Following Italy's ratification of the FAO International Treaty on PGV/FA in 2004, the Ministry of Agriculture (MiPAAF), in 2005, launched an open-ended project targeted at the implementation of this Treaty, giving priority to the safeguard, characterization, evaluation, utilization and documentation of plant species of agricultural interest. This project is particularly devoted to the research institutions of the Agriculture Research Council (CRA) belonging to the same Ministry. Fifteen of these institutions are involved in horticultural crops of current and potential interest for the Italian economy. Thirty-three of these crops are fruit species (including Grapevine, Olive and Citrus), 14 are vegetables, 130 medicinal and aromatic plants and 3 ornamental species. Currently, 16.497 accessions are maintained, of which 15.526 are fruits, 520 vegetables, 163 medicinal and aromatic plants and 288 are ornamental species. Vegetative and generative conservation methods (*in vivo* and as seeds) are most common, but steady research on specific protocols for other techniques, such as *in vitro* conservation and cryopreservation allow for a constantly increasing rationalization of the collections. Accurate characterization and documentation of the conserved material as well as the free access to this information is crucial to the promotion of global exchange of germoplasm for food safety and

sustainable agriculture. Characterization comprises not only morphological and agronomical traits, but also biochemical and molecular analysis of the accessions. According to the policy of the Ministry, the economical valorization of autochthonous varieties is one of the main objectives and several positive results have been achieved so far. As far as possible, the descriptors used are those adopted at international level (FAO/IPGRI). The collected data and research results are available through the National Inventory on plant genetic resources conserved *ex situ* in Italy.

S12.005

A Window to Europe's Plant Diversity: An Example of a European Network EURISCO - The European Plant Genetic Resources Search Catalogue

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EURISCO provides information on European agricultural biodiversity via an on-line catalogue, allowing users to access key data, primarily for the identification and localization of germplasm samples (accessions). To answer questions such as "Who holds what?" and "Where can I find materials?", the European region established National Inventories (NIs) of *ex situ* germplasm collections and launched a web-based window (<http://eurisco.ecpgr.org>), making accession-level data universally available and readily accessible for users. EURISCO is designed to serve as the European plant genetic resources (PGR) information hub, with the NIs providing the catalogue's backbone and granting users access to passport data, and soon, as well as accession-related phenotypic (characterization and evaluation) data. EURISCO uses international standards for information registration, access and exchange, enabling users to query and locate data on crops, forages, wild species and crop wild relatives, landraces and breeding lines for a wide range of crops. It is based on a European network of 42 National Focal Points and ensures that biodiversity data on over one million accessions from 40 European countries is globally available. These accessions, covering 5,403 genera and 34,465 plant species, represent more than half of the *ex situ* accessions maintained in Europe and roughly 18% of total worldwide holdings. EURISCO supports policy makers in meeting country commitments and national, regional and international obligations; especially those relating to the FAO Global Plan of Action (GPA) and the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA). EURISCO also contributes to the Multilateral System (MLS) and the Standard Material Transfer Agreement (SMTA) of the ITPGRFA and the Convention on Biological Diversity (CBD), with regards to the conservation and utilization of PGR materials. EURISCO is hosted at and maintained by Bioversity International on behalf of the Secretariat of the European Cooperative Programme for Plant Genetic Resources (ECPGR).

S12.006

The Development of the UK National Fruit Collections

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The UK National Fruit Collections at Brogdale contain over 2200 accessions of apples as well as over 500 accessions of pear, along with collections of plums, cherries, bush fruit and other related species, including ornamental material. The collections have developed over a number of years in line with the variety trialling and PVR testing that was previously carried out at Brogdale. The University of Reading is now responsible for the scientific curation and maintenance of the collections. The overall aims of the scientific curation are to increase the security and improve the utility of the collections as a genetic resource. Toward this development we have carried out a detailed molecular genetic screen of the apple collection and used molecular markers to verify the recently repropagated pear collection. We are also using cryopreservation techniques to produce a safety back up to the collections. The molecular analysis of the apple collection has so far been based around carrying out DArT analysis across the whole collection. Verification of the pear collection has involved screening the newly repropagated trees using SSR markers to check

for errors in the repropagation process. The findings of these studies have allowed us to begin an assessment of the collections and look further at proposed duplicate and mislabelled accessions. Data from these studies will be made available through publication and via our web-based germplasm database and will be of value to researchers and breeders. Material from the collections is available for research, breeding and training purposes in line with the workings of the International Treaty on Plant Genetic Resources for Food and Agriculture.

S12.007

Domestication and Varietal Diversification in Fig: Insights for Conservation and Local Genetic Resources Management

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Previous genetic study on Mediterranean fig, *Ficus carica* L., showed the existence of multiple domestication centers. The objective of this study is to understand how do selection and varietal diversification process occur at a fine geographic scale. Based on population genetic approach using nuclear and chloroplast microsatellites, we analyzed spontaneous populations and varieties issued from six Moroccan eco-geographical areas. Fig varieties are very diversified and cultivated locally: the variety does not exceed a village scale. They are characterized by a pattern of isolation by distance similar to that observed in spontaneous figs indicating a signature of a genetic equilibrium situation, with no trace of a recent colonization process. We noted a genetic differentiation among the six zones ($F_{st} = 4\%$) and particularly between Rif (northern Morocco) and Oasis (South). Within each of these zones, varieties are genetically close to spontaneous fig. We showed that domestication occurs at a geographical area as the Rif and the Oasis. Furthermore the varieties are mainly originated from seedlings involving spontaneous caprifigs (male figs). Our results suggest that cultivated and spontaneous fig form a single evolutionary unit, and fig domestication is a continuous process. We discuss on the relationships between *ex situ* and *in situ* conservation and on determinants of *in situ* conservation related to traditional agro-ecosystems evolution.

S12.008

Assessment of Genetic Diversity of Five Coconut Population Bangladesh Locating at Different Geographical Position of the Country

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Genetic diversity of five coconut populations of Bangladesh was estimated followed by morph metric characterization during 2000-2002. Sample populations were selected by course grid sampling method strategy. A suitable sized map of Bangladesh was obtained and grids of approximately 40x40 km were marked following latitude and longitude divisions. Each 40 km was marked as A, B, C and so on along the longitude and 1, 2, 3 and so on along the latitude and population in each grid were designated as grid number and sample number. Thus five populations were as T1= D7, T2= G6, T3= G13, T4= D11 and T5= G11 were identified. Three sampling sites in a grid at least 2 km away from each other, represented replications of the grid. Thirty normal palms were chosen for data collection. Stem, leaf, inflorescence and nut characters were measured during data collection. Shannon weaver diversity index was used to estimate diversity of both qualitative and quantitative characters within the population. Significant variation in respect of length of 11 leaf scare, spike without female flower in the inflorescence and fruit characters were noticed between the populations. Geographical location of the populations did not influence the pattern of diversity between the populations. The results were validated by the high diversity indices of qualitative and quantitative of the population. High Shannon Weaver Diversity Indices 73-89% and 75-85% in flower and fruit characters respectively indicated existing of high variability within the population each location.

S12.009

Juglans regia in Uzbekistan Potential Resource of Biodiversity, Trees and Fruits Description

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The natural area of walnut in Central Asia includes the territory of Kyrgyzstan, Tajikistan and Uzbekistan, there the walnut was grown since Oligocene times. It is considered that the walnut forests saved in these countries are the center of distribution of walnut around the world by people. Forest stands with walnut are widespread in Uzbekistan up to the area of about 4 thousand hectares. They occupy areas with optimum forest growing conditions on slopes of mountains of Western Tien-Shan and Pamir-Alai at height from 800 up to 2300 m above sea level. The Bostanlyk District of Tashkent Region is related to the zone of the densest concentration of walnut plantations. Surveys carried out in 2005-2009 in Uzbekistan walnut forests in a part of Western Tien-Shan territory showed that *Juglans regia* is widespread on ranges as separate small populations on small river inflow and slopes of Ugam, Pskem, Chatkal, as well as Angren. From the complex of investigated biomorphological attributes of trees it is possible to search and classify forms as related to breeding-valuable category. From area considered three selected trees in Charvak and Chatkal (trees coded KRK-29, CHVK-200 and CHVK-76) deserve the special attention. Fruits of the surveyed trees has also been evaluated. Variability in size and shape of nuts as well as the contents of basic spare nutrients in the nut kernels has been done. Fats/lipids, fatty acids composition, proteins, soluble sugars, vitamin E were analysed. To the selected forms based on in forest trees survey, from nuts evaluation some more forms (KRK-23, SDJ-9, SDJ-10) can be added to the attention in the perspective for reproduction and establishment of collection-mother production plantations and to be candidates to become varieties.

S12.010

Management of Temperate Fruits Genetic Resources in India

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India is enormously rich in temperate fruit genetic resources. In the North, the temperate Himalayas stretch from Jammu and Kashmir to the North-eastern hill region and in the South, the Nilgiri hills, also harbour a vast genetic diversity of temperate fruits, including apple, pear, peach, apricot, plum, walnut, almond, pecan nut, hazelnut, chestnut, berries, and several of their wild relatives. The National Bureau of Plant Genetic Resources is the nodal organization for exchange, quarantine, collection, conservation, evaluation and the documentation of plant genetic resources. Numerous germplasm comprising broad genetic diversity in temperate fruits were introduced from other countries. Many introduced varieties have been used directly for large scale cultivation, for example, the Red Delicious group of cultivars in apple, etc. Diverse temperate fruit crops have been collected, characterized and evaluated; promising germplasm have been identified and utilized in crop improvement programmes. The germplasm is conserved through *in situ* and *ex situ* complementary strategies. The wild temperate fruit species are conserved in protected areas and national reserves. The National Genebank at NBPGR has a large *ex situ* conservation facility including field genebanks, seed genebanks, *in vitro* multi-crop repository, and cryobank. Important temperate fruit accessions are being conserved in the field genebanks; however, germplasm remains under the threat of loss due to pest-pathogen attack, natural calamities and climate change. A back up-*in vitro* collection includes more than 300 exotic and indigenous accessions of *Actinidia*, *Fragaria*, *Malus*, *Morus*, *Prunus*, *Pyrus*, *Rubus* and *Vaccinium*. Future strategies must include monitoring of *in situ* genetic diversity loss due to climate change, identification of trait-specific germplasm such as low-chilling, biotic and abiotic stress resistant, systematic conservation and efficient utilization. This review focuses on the present status of temperate fruits genetic resources, their management and future perspectives.

S12.011

Seed Conservation - An Effective Tool for Long Term Conservation of Genetic Resources in Amaranth

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Seed conservation is a valuable tool for preservation of genetic diversity. It is popular amongst conservationist in view of easy handling, economical and genetically stable of the stored material even after long period of storage. Maintenance of high seed quality in terms of viability and vigour is important parameter in seed conservation, which was adversely affected by the high storage temperature and high relative humidity or seed moisture. Lowering these parameters help in extending the storability. Thus an experiment was conducted with the view to optimize conditions for longer storage of amaranth seed where information is inadequate. Storage temperature significantly improved the seed longevity and able to maintained high seed viability and vigour for 20 years. In this per cent recovery of viable seeds were >90% and seedlings emerged from stored seeds were normal, vigorous and free from morphological variations. Thus seed conservation at sub-zero temperature (-20 °C) is effective, efficient, beneficial and economical for preservation of genetic resources of amaranths an underutilized and nutritious vegetable. The paper describes details of variability in amaranth and suggests effective means for longer maintenance of amaranth germplasm.

S12.012

Modified Atmosphere Storage - A Useful Tool for Germplasm Conservation

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Seeds are widely used for raising different vegetable crops and also form an important means for germplasm conservation. In modified atmosphere storage seeds are packed in laminated aluminium foil pouches along with nitrogen, carbon dioxide or in vacuum. Of late, in view of rapid development and greater commercialization of vegetable industry a large number of newer genotypes are available for growing which has caused greater damage to diversity and resulted in disappearance of several old and primitive cultivars. It is essential to conserve large variability for survival of plant under heterogeneous environment. It may possess valuable genes required for future crop breeding. The storage life of seeds generally dependent on storage conditions such as temperature, relative humidity and oxygen content. The latter causes more damage especially in un-dried seeds. Seeds deteriorate faster in presence of excess of atmospheric oxygen. Seed conservation by replacing air with nitrogen or carbon dioxide reduces the deterioration process and helps in extending storage life in several vegetable crops. The present study was conducted in view to harness the benefits of modified atmospheric storage for germplasm conservation. The study revealed that seed conservation with nitrogen preserved high viability in onion (5 years), muskmelon (7 years), bottle gourd (5 years), amaranth (5 years), and cucumber (5 years) and with carbon dioxide in cabbage (3 years), bell pepper (4 years), cowpea (4 years), kohlrabi (5 years), garden peas (5 years), cluster bean (5 years), fenugreek (2 years), spinach beet (5 years), ridge gourd (8 years), and bitter gourd (5 years), under ambient conditions. The seedling emerged were uniform, healthy and free from variations. The modified atmospheric storage especially in seed germplasm conservation. This will be beneficial for short term preservation of germplasm especially in absence of cold storage facilities. The details will be discussed during presentation.

S12.013

Apple Key - An Internet Tool for Genebank Data Including Cultivar Identification

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Cultivar descriptions and correct identification are central for the work with

genetic material, especially in Genebanks. The data collection of many descriptive parameters is very laborious in itself, but the handling of the data is also a challenge. However, when it comes to the level of use and communicating the information to breeders, researchers and especially the public the challenge appears to be almost impossible. If one tries to visit a range of genebank homepages around the world it is very difficult to extract the information even for specialists. At Pometet a collection of around 800 cultivars of apples from 22 countries is maintained. Most are historic local varieties from Denmark and the neighboring countries. Generally we experience a large public interest in information about the collection. In order to communicate the information collected over the years we initiated a project to develop an internet platform. A dual aim was; firstly to present the diversity of apples in terms of appearance and qualities and also allow a possibility to include unique historical and cultural information including photos, pomological drawings and paintings. Secondly, the aim was to develop a tool for identification of varieties and easy possibilities for targeted search for cultivars with specific characteristics. In September 2009 the 'Apple Key' was published on the internet www.agri.life.ku.dk/Pometet.aspx which includes 319 apple cultivars. The user interface and all data are available in both Danish and English. 37 different groups of descriptive parameters are used with a major emphasis on external and internal characters of the fruit. 15 parameters are used to describe the external appearance such as color, shape, eye and stalk while 7 internal parameters are focused on structures like tube and eye structures and stamen position developed by Robert Hogg in 1880'ies. Other parameters focus on quality for use (table fruit, cooking, juice), flowering characteristics and tree growth and health (disease sensitivity). The development and use of the Apple Key is demonstrated and the stability of different characters in cultivar identification is discussed.

S12.014

Conservation Strategy of Genetic Resources in Strawberry in Germany

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The 'National Program for Genetic Resources of Agricultural and Horticultural Plants' in Germany is designed to provide long-term preservation, utilization, research and development for these species. The conservation strategy requires the application of different methods for safety duplication. One way is the German Fruit Genebank established as a network of collections held at several locations. Based on an assessment performed in different institutions, 369 strawberry cultivars were selected to be included into the National German Strawberry Genebank. In addition to the cultivar collection the Gene bank of the Institute for Breeding Research on Horticultural and Fruit Crops in Dresden-Pillnitz comprises with 318 *Fragaria* wild species the biggest collection in Europe. High operating expenses and budget limitations for field collections do not allow further duplication. First, *in vitro* cold storage was elaborated and adapted. The average storage duration at 4 °C for strawberry cultivars was 22 months and for wild species accessions 9 months using five chamber bags as a storage container and a combined medium without plant growth regulators or vitamins. A calculation required to establish and maintain a safety duplication of the collection demonstrated that the application of the *in vitro* cold storage is too labor intensive to be maintained. The cryopreservation should be established as the cost effective long-term storage. Preliminary experiments with PVS2 vitrification using 22 cultivars and 7 wild species accessions demonstrated recovery data varied from 37 to 86 % for the cultivars and from 0 to 62 % for the wild species. Further experiments included the comparison of different cryopreservation methods (1) PVS2 vitrification, (2) encapsulation and (3) controlled slow cooling. Results of the regeneration rate after cryopreservation will be presented and conclusion for the further routine work to managing the cryopreservation for the *Fragaria* gene bank will be given.

S12.015

Japan - US Expedition to Hokkaido and Northern Honshu to Collect Fruit and Nut Genetic Resources

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From 7 to 25 September 2009, Japanese and U.S. scientists collaborated on an expedition to collect genetic resources of temperate fruit genera throughout Hokkaido, and Northern Honshu, Japan. A non-funded cooperative agreement was prepared, according to the rules of the International Treaty on Plant Genetic Resources for Food and Agriculture. This expedition was a collaborative effort between, the Japanese Ministry of Forestry and Fisheries, the United States Department of Agriculture and Akita Prefectural University, Japan. Additional assistance was provided by the Hokkaido Governmental Plant Genetic Resources Center, Forest and Forest Product Research Institute of Hokkaido, Hokkaido Agricultural Research Center, and private botanists. Plant materials were shared between the scientists of both countries. The expedition toured many localities in Hokkaido, Aomori, Iwate, and Akita Prefectures. Different ecosystems were visited including upland forest, coastal marshes, upland bogs, and dense forests. The expedition obtained 147 seed and plant samples representing 21 genera and 53 species. The temperate fruit, nut and specialty crop genera collected include: *Actinidia*, *Chaenomeles*, *Corylus*, *Crataegus*, *Empetrum*, *Gaultheria*, *Humulus*, *Fragaria*, *Lonicera*, *Lycium*, *Malus*, *Mentha*, *Humulus*, *Prunella*, *Potentilla*, *Pyrus*, *Ribes*, *Rubus*, *Sorbus*, *Vaccinium* and *Vitis*. Accessions from this trip are preserved at the USDA ARS National Clonal Germplasm Repository in Corvallis, Oregon. Morphological and molecular evaluation of this germplasm will be conducted at the USDA ARS Corvallis Repository and the Japanese Ministry of Forestry and Fisheries, National Institute of Fruit Tree Science, Tsukuba.

S12.016

Cryopreservation and *in vitro* Culture - State of the Art as Conservation Strategy for Genebanks

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Cryopreservation - storage of genetic material in or above liquid nitrogen is the safest and most cost-effective method to store vegetatively propagated plant germplasm and such forming recalcitrant seeds. Experience has been accumulated in many species. Along the main line consisting in explant trimming, dehydration, cryoprotection and temperature reduction several parameters can be variegated. Active reactions of the tissue prior to cryopreservation treatments may be used, such as cold adaptation. Dehydration conditions and cooling speed are other important factors. Finally repair of cryo-injury and regeneration must be investigated. Using these basic components several methods were developed, such as dormant bud method, two-step freezing, vitrification, encapsulation and various droplet methods, also in combination. Although being suitable for most of targeted species, considerable differences were found in success rates depending on genotype and presence of endophytes. *In vitro* storage uses slow growth methods for medium-term preservation. These methodical surroundings to cryopreservation can also be used in parallel to it, especially when material is more often requested. Genebanks need to maintain highly diverse collections and cannot focus on well-investigated model organisms. Therefore, logistic questions are important and considerations concerning regeneration probability, storage safety and economy. Various scenarios can be followed depending on given situations like highly safe storage of important material or emergency rescue in case of acute danger of loss. It is also necessary to assess relations of storage and request

frequency. Finally, cryopreservation and *in vitro* culture methods can also be potential contributions to store seed-producing populations in case of limitations in true-seed reproduction. Criterion for success is safe maintenance of healthy and genetically unchanged germplasm. At present, the genebank of IPK Gatersleben maintains 1250 accessions in cryopreservation, mainly of potato, garlic and mint. The characteristics of *in vitro* storage and cryopreservation will be exemplified using the experience with these crops.

S12.017

Development of *in vitro* Propagation, Slow Growth Storage and Cryopreservation Protocols for Short-, Medium- and Long-Term Conservation of Young and Mature Explants of Thyme (*Thymus vulgaris* and *T. longicaulis*)

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Thyme is one of the most important genus of the *Lamiaceae* family, covering more than 400 perennial species of aromatic and medicinal shrubs/subshrubs. It serves as a natural source of phenolic oils, oleoresins, fresh and dried herbs. Non-medicinal use of the plant (e.g., food additives, cosmetic and alimentary industries, landscape plants) is also of great importance. However, natural thyme populations are unfortunately far from being adequate to support such a great and even growing demand for its products. In this sense, recently developed *in vitro* propagation techniques provide a unique alternative for the high-rate multiplication of plants of horticultural, economic and medicinal importance, and pave the way to medium- and long-term conservation of such valuable germplasm by means of slow growth storage and cryopreservation. Present study was designated to develop effective protocols for *in vitro* propagation, slow growth storage and cryopreservation of young (i.e., shoot tips originating from young seedlings) and mature explants (i.e., shoot tips originated from mature plants) of two distant *Thymus* species, i.e., *T. vulgaris* and *T. longicaulis*, in order to ensure its applicability to a wide range of thyme germplasm. Optimizations for *in vitro* shoot proliferation included use of cytokinins (6-benzyladenine, kinetin and thidiazuron) alone or in combination with auxins, gibberellic acid and/or silver nitrate. Shoot rooting was easily obtained on semi-solid MS medium, either hormone-free or supplemented with auxins. Rooted plantlets were transferred to 250-ml plastic pots and acclimatized successfully by gradually reducing the relative humidity. Slow growth storage was achieved by keeping the *in vitro* shoot cultures at 4 °C and in darkness up to 12 months. Three different vitrification-based one-step freezing techniques (PVS2 vitrification, encapsulation-vitrification and droplet-freezing) were tested for cryopreservation, however initially all the preliminary steps of cryoprotection (i.e., cold-hardening, sucrose preculture) were also optimized.

S12.018

In vitro Conservation of Economically-Important Fruit and Ornamental Species by Slow Growth Storage

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Slow growth storage refers to the techniques enabling the maintenance *in vitro* of shoot cultures in aseptic conditions by reducing markedly the frequency of periodic subculturing (from few months to one year or more) and the consequent risks of contamination, without affecting regrowth potential of shoots. The most used approach couples the storage of shoot cultures at low temperature (generally, 4 °C) with dark or low-light conditions. However, other factors (such as the medium formulation and the characteristics of containers) have important effects on the time of conservation. As for fruit species, the present study focused the attention

on slow growth storage (at 4 °C and in the dark) of shoot cultures from three fruit rootstocks (GF677, Gisela5 and Mirabolano 29C) and the kiwifruit (cv. Hayward). Different sucrose concentrations (30, 45 and 60 g/L) and a sucrose/mannitol combination (30 and 15 g/L, respectively) in the conservation medium were tested, with the aim to improve the quality of the stored plant material and to prolong the conservation time. Kiwifruit, Mirabolano 29C and Gisela 5 greatly benefited when the sucrose concentration in the conservation medium was increased to 45 or 60 g/L, providing the conditions for a 18-month preservation of shoot cultures which maintained a good quality and the potential of recovering fastly the proliferation activity when moved back to standard culture conditions. GF677 shoot cultures could be preserved up to 21 months, with no marked differences among the 3 sucrose concentrations in terms of shoot quality. Mannitol, used in combination with sucrose, had detrimental effects on the conservation of GF677 and Mirabolano 29C shoot cultures. As for ornamental species, a study on slow growth storage of shoot cultures of *Ranunculus* and *Anthurium* is currently in progress, with promising preliminary results as for the possibility of *in vitro* conservation for at least 6 months.

S12.019

Application of Low Temperature, High Osmotic Pressure, Growth Retardant to Extent Subculture Interval during Long-Term Conservation of Apple *in vitro*

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Germplasm conservation via tissue culture has many advantages, such as pest and disease free, no weather effect, space saving, etc. But the cost of subculture and variation due to frequent subculture limit its application. Retarding plantlets growth could be a method to overcome this problem since it may extend the interval of subculture. Plantlets of Jonagold and Fuji apple that subcultured in medium of MS + BA1.0mg/L + NAA 0.05mg/L + Sucrose 30g/L + Agar 6g/L, pH 6.0, 25 °C were studied under the treatments of: 1) incubated at 3 °C; 2) Sucrose 60g/L, 25 °C; 3) Daminozide 80 mg/L, 25 °C; 4) Sucrose 60g/L, 3 °C; and 5) Daminozide 80 mg/L, 3 °C. The number of bud differentiation and plantlet growth were recorded every 10 days for 100 days. The maximum optimal interval between subcultures for all treatments was evaluated. The results showed that all treatments could significant retard the bud differentiation and plantlet growth. The plantlets under the combination of low incubating temperature with high content of Sucrose or adding Daminozide had less amount growth than that under any single factor treatment after 30-40 days incubation. In doing this, the subculture interval, which was three months for apple plantlet's conservation normally, could be extended to fifteen months, and the surviving rate was more than 87% and the surviving plantlets remained good growth status. The plantlets of Fuji apple showed better adaptability to this strategy than that of Jonagold apple.

S12.020

Photoautotrophic *in vitro* Conservation and Multiplication of Rare *Dianthus* Species

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A lot of endemic or rare species of *Dianthus* genus have also ornamental values. Nowadays *in vitro* culture biotechnology represents a valuable method for their conservation and multiplication. The classic *in vitro* culture develop in culture vessel that are isolated from external atmosphere, the CO₂ is consumed in the first ours after inoculation and therefore, they need a culture media suplimented with 2-3% sucrose. The photoautotrophic (PA) *in vitro* culture develop on culture media without sucrose and in a CO₂ enriched (0.03% to 5%) atmosphere. In this way the PA neoplants grow photosynthetically and their acclimatization is thus facili-

tated. *Dianthus callizonus* Schott et Kotschy is strictly endemic for Piatra Craiului Mountains (Southern Romanian Carpaths) and it is a rare species. *D. nardiformis* Janka is vulnerable/rare and *D. dobrogensis* Prodan is rare/endangered, both being endemic for Romania and Bulgaria. These species were collected from their natural habitats – Nature 2000 sites. *In vitro* cultures were induced starting from nodal or apical explants. Experimental variants were: (i) Control 1 (2% sucrose, no CO₂ supplement), (ii) PA1 (no sucrose, atmospheric CO₂ concentration = 0.03%), (iii) PA2 (no sucrose, 2% CO₂), (iv) Control 2 (no sucrose, no CO₂ supplement). The number and length of neoplants and roots were recorded, at 10, 20 and 40 days after inoculation, while chlorophyll (a+b) and carotenoids contents was measured after 40 days. For *D. nardiformis*, with PA1 variant was obtained better results compared to Control 1 while *D. callizonus* and *D. dobrogensis* on PA1 variant presented similar results compared with Control 1 variant, with a satisfactory multiplication and rhizogenesis. For all three species, the PA1 variant gives better results compared to PA2 variant.

S12.021

The Rooting Characteristics of Apple Plantlets Repeatedly Subcultured *in vitro*

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Tissue culture has been widely used in micropropagation and germplasm conservation in apple due to its advantage of no seasonal limitation and high propagation efficiency. Meristem of the plantlets keeps in active status all time under the stimulation of plant growth regulators supplied in media, and then the plantlets can be multiplied every forty days. Whether the rooting ability changes over a long period repeatedly subculture is a question. The plantlet clones of Jonagold and Golden Delicious Spur apple were set up by sampling explants from some mother plants in May of 1991, 1996, 2005, 2006, and 2007, respectively; and the clone of apple rootstock M26 was set up in 1986 and 2008. Subculture medium was MS+BA 1.0 mg/L+NAA 0.05 mg/L+Sucrose 35.0 g/L+Agar 6.0 g/L. The rooting experiment was conducted in 2009. The subcultured times of the plantlets used for rooting were calculated as eight times each year in average, that was 147, 107, 37, 27, and 17 times for Jonagold and Golden Delicious Spur apple, and 187 and 11 times for rootstock M26. Rooting medium was 1/2MS+IAA 1.0 mg/L+IBA 0.3mg/L+Sucrose 25.0 g/L+Agar 6.0g/L. The rooting status was recorded every ten days. The rooting ability of M26 decreased after a long period repeatedly subculture. The rooting percentage of M26 plantlets subcultured for 187 times was only 32%, which was significant lower than that of plantlets for 11 times. No significant difference was found between Jonagold and Golden Delicious Spur apple for all given subculture times in terms of the rooting percentage, but the root growth status during the late stage of root development was different. The roots from the plantlets with less subculture times, such as 37, 27, and 19 times, were shorter and thicker than that with more subculture times, such as 107 and 147 times.

S12.022

Alternative Storage for Germplasm of Native Hawaiian Berries

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Ohelo berry (*Vaccinium reticulatum* Smith) is a small, native shrub commonly found in disturbed, open sites at 640 to 3700 meter elevation on the islands of Hawaii and Maui. The shrub is very hardy with compact form and bright waxy red new growth. Three cultivars: Kilauea, Red Button, and Nene were selected for ornamental use. This study tested *in vitro* cold storage and cryopreservation of these cultivars for germplasm preservation. Shoot cultures in StarPac tissue-culture bags

were maintained in good condition in 4 °C cold storage for more than a year. For cryopreservation shoot cultures were cold acclimated with alternating temperatures and photoperiod, and shoot tips were cryopreserved with three protocols: controlled rate cooling, encapsulation dehydration and PVS2 vitrification. Shoot tips were moderately desiccation sensitive and when encapsulated in alginate beads could only be dried to 31% moisture content before losing most viability. All three protocols had regrowth high enough for long-term storage of the germplasm (>40%). Regrowth following controlled rate cooling was 42 to 50%; encapsulation dehydration 60 to 75%; and PVS2 vitrification 45 to 70%. The addition of ascorbic acid (vitamin C) to the pretreatment medium during cryopreservation improved shoot regrowth in some treatments. These studies show that Ohelo berry can be successfully cold-stored or cryopreserved with any of these techniques.

S12.023

Cryopreservation of Peach Shoot-Tips

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The encapsulation-dehydration, vitrification and droplet vitrification methods were applied to *in vitro* grown shoot apices of three peach (*Prunus persica* (L.) Batsch) cultivars, Summer Grand, San Giorgio and Babygold 6. In the application of the encapsulation-dehydration method, apices were pre-grown for 1, 3, 5 or 7 days in liquid medium supplied with various sucrose concentrations (0.5, 0.75, 1.0 or 1.25 M). Desiccation was performed with silica gel for 0, 4, 6, 8, 9 or 14 hours. All the cultivars showed re-growth after cryopreservation with encapsulation-dehydration (from 43% to 52%) The highest response with this method was obtained maintaining encapsulated apices for 3 days in liquid medium supplied with 0.5M sucrose and desiccation of beads in silica gel to 20% of water content (9 hours of treatment), followed by immersion in liquid nitrogen (LN). In the vitrification and droplet vitrification techniques shoots were treated with the PVS2 solution for 0, 30, 60 or 90 minutes. Afterwards, explants were either immersed in LN in 2.0 ml cryotubes containing ice-cooled PVS2 solution, or singularly transferred to a droplet of PVS2 solution and placed on a strip of aluminium foil and immersed in LN. In the first experiments the droplet-verifications method seems to be more suitable than simple vitrification for cryopreservation of peach.

S12.024

Storage Stability Using Cryopreservation: A Case Study in Papaya

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Ex situ conservation of *Carica papaya* and its crop wild relatives, the *Vasconcellea* spp., has been the subject of ten years of research in our laboratory. This paper will summarise the achievements and key findings of this work. Both clonal and seed material have been investigated to allow storage of elite clonal material as well as maximum genetic diversity. Clonal material from several genotypes has been successfully stored using micropropagation followed by shoot tip cryopreservation, with >70% regeneration after 12 months storage in LN. The regeneration and acclimatisation of cryopreserved plants for field testing confirmed that true-to-type plants were produced. Genomic DNA markers also confirmed that no changes were associated with any stage of the cryopreservation protocol, whilst small changes in DNA methylation status were associated with cryoprotectant and LN treatments. Storage of papaya seed at a range of seed moisture contents and temperatures indicate that seed is essentially orthodox, but requires dormancy breaking treatments for germination. However, germination post storage for 12 months was >68% at ultra-low temperatures (LN), but <5% at conventional seed storage temperatures (-20 °C). This is in line with recent evidence that cryopreservation enhances storage stability and longevity, even in orthodox seeds, when compared with standard seed bank approaches. This highlights the importance of cryopreservation for the long-term management of genetic resources of both clonal and seed genetic resources of papaya.

S12.025

Cryopreservation of *Pelargonium* Species: A Comparative Study of Encapsulation-Dehydration and Droplet-Vitrification Protocols

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In order to guarantee safe, long-term conservation of the AGROCAMPUS OUEST – INHP *Pelargonium* collection, apex cryopreservation studies have been undertaken. An encapsulation-dehydration procedure has been first developed. More recently, studies were undertaken in order to adapt the droplet-vitrification procedure to this genus. For both procedures, protocols were optimized using *P. xpelatum* ‘Balcon Lilas’ as model accession. Survival and plant regeneration were obtained with the two newly determined protocols. However, droplet-vitrification gave higher and more reproducible regeneration results. The two protocols were then tested on 20 or more genotypes representative of the diversity of our collection. With encapsulation-dehydration protocol, apex survival was obtained for 19 accessions out of the 20 tested. For the 10 genotypes tested 3 times or more, survival rates were between 28% and 84% (with an average of 50% survival). With droplet-vitrification, for each accession, at least 3 repetitions were performed on different days, reaching a minimum of 24 cryopreserved apices per genotype. Apex survival was obtained for each accession out of the 28 tested. An average of 65% survival rate was obtained ranging from 14.8% for *P. fragans* to 90% for *P. capitatum* and *P. xhortorum* Neurot. Plants were regenerated for each genotype, except *P. xpelatum* Papa Crousse. The genotype dependent tolerances at each step of the process were compared for a sample of accessions. To compare the two protocols, a histo-cytological study was performed on *P. xpelatum* ‘Balcon Lilas’ apices. Samples were fixed after each step of both protocols in order to observe the modifications at the cellular level. Important differences were noted concerning starch accumulation and nucleus aspect.

S12.026

Dormant Bud Cryopreservation: Secondary Buds Can Significantly Affect the Estimation of Post-Thaw Survival

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Dormant bud cryopreservation for the genetic conservation of fruit crops of temperate regions has significant benefits in that it is essentially a field-level technique that does not require resource-costly *in vitro* culture. A protocol for *Malus* (apple) that was developed in the N. American continental climate has latterly been employed in the maritime climate of Denmark. Early in the first step of the protocol (incubation at -40 °C) a single freezing event occurs within the buds and thereafter progressive weight reduction is recorded, suggesting both cryodehydration and bulk tissue water loss are occurring. When cooled subsequently to -300 °C and then into liquid nitrogen, no further exotherms or water losses are detected. Given that the distribution of ice within the bud may be an issue during cryodehydration, bulk water loss and thawing, bud volumes were considered. Bud volumes were calculated by stereological measurement for three apple cultivars, exhibiting high, medium and low survival following cryopreservation. The highest survival correlated positively with volume yet for c.60% of such samples the primary meristem was lethally damaged and survival came from secondary buds. Control populations exhibited secondary bud outgrowth in a maximum of 25% of cases. Such data has not previously been recorded and is of significance as buds with larger volumes may be better differentiated to produce such secondary structures.

S12.027

Evaluation of Genetic Stability in Wasabi Plants Regenerated from Shoot Tips Cryopreserved for 10 Years

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Cryopreservation for plant germplasm is becoming a routine technique for long term preservation. Some studies pertaining to the genetic stability of cryopreserved plants were reported. However, there is very little evidence about the genetic stability of long-term cryopreservation in plants. In this study we investigated, from a molecular, morphological and biochemical approach, the genetic stability of plants regenerated from wasabi shoot tips cryopreserved by vitrification over a 10 year period. We confirmed that wasabi shoot tips cryopreserved for 10 years could maintain a survival rate of greater than 90%. Few statistically significant difference was observed in some of the leaf's contents. These differences do not seem to be based on genetic variation, and will disappear by the following cultivation. Genetic relationship among regenerated plants above were determined by random amplified polymorphic DNA (RAPD) PCR. Total DNA was extracted from each leaf of plant by PVP method with some modifications. For RAPD analysis, 20ng genomic DNA and 100pmol of 10-mer oligonucleotide primer. The results show that there were no variations in PCR patterns. Thus, with the experimental conditions tested, wasabi plants derived from shoot tips cryopreserved for 10 years were found to be genetically stable.

S12.028

Raspberry Cryopreservation by Droplet Vitrification Technique

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Over the past decades, plant cryopreservation technologies have evolved rapidly, opening the door to the long-term storage of valuable genetic resources of many crop species. Thus, from the original slow-cooling approach, research has moved to more reproducible and users-friendly techniques which enable complete vitrification of intra- as well as extracellular liquids through the direct immersion of explants in liquid nitrogen. Among these techniques, the droplet vitrification (DV) is relatively new, and has been applied to only a limited number of species (Sakai and Engelmann, 2007). In the present work, we applied a cryopreservation method that utilizes droplet vitrification (Panis *et al.*, 2005) to *in vitro* grown raspberry (*Rubus idaeus* L.) apices, cv. Latham and cv. Autumn Bliss and compared this with the vitrification one-step freezing (OSF) method. Excised meristems were transferred into the loading solution for 20 min at room temperature, treated with the vitrification solution (PVS2) for 30 min at 0 °C and then frozen into individual droplets of PVS2 placed on a strip of aluminium foil. After 20 min, cryopreserved explants were rapidly thawed in the recovery solution, for 15 min, and subsequently placed onto a regeneration medium, where they were kept in darkness. After 7 days the explants were transferred to a fresh medium and cultured under similar condition as normal *in vitro* grown shoots. For the cultivar Latham, the application of the DV method results in 46.7% and 18.75% of the explant respectively surviving and regrowing after liquid nitrogen immersion, while very low survival rates and no regrowth was observed using the OSF method. Similar results have been also obtained with the other cultivar Autumn Bliss. Optimization of the protocol is now in progress. These promising results prove the value of these technique for the conservation of raspberry genetic resources.

S12.029

Innovation in Conservation: How New Information Technology Tools Improve the *ex situ* Management of Plant Genetic Resources

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Genebanks are in the business of conserving, a conservative business by nature. However, many new technologies highly relevant to the PGR community have become available over the past years, especially in the fields of genomics and computer science. The effect of the second category of technologies on the *ex situ* management of plant genetic resources will be explored. After a low initial level of standardization in genebank documentation systems, a strong increase of access and exchange of information can be observed. Important elements behind this increase are standardization - creating the possibility to interpret each others information - and web services - creating the possibilities for machines to access each others information via the Internet. Some aspects of these developments such as the application of ontologies, persistent identifiers and anticipated developments such as the increased use of open source software, will be presented. Also for the user of PGR and associated information a lot has changed and continues to change. Ongoing developments include on-line querying and ordering facilities and the application of GIS technology for querying and trait prediction. A final element that will be described is the development of virtual genebanks: thanks to IT tools, the management of PGR and the interfaces to these PGR can be decoupled. As a result, anyone can develop a website giving access to PGR, including the possibility to order material in one or more genebanks anywhere in the world.

S12.030

Pear Diversity (*Pyrus* spp.) in Northwestern Spain by Microsatellites

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Spain is among the ten largest pear producers of Europe. Its main production is based on foreign cultivars. However, the traditional culture of the northwest region has maintained most of the local cultivars in the old orchards. Between 1978 and 1981, the Centro de Investigaciones Agrarias de Mabegondo (CIAM) in Galicia established a Germplasm Bank to collect local varieties of pears, in order to conserve genetic resources of this fruit. In the current study, 216 accessions from CIAM were analyzed and compared with 19 commercial varieties of *Pyrus* spp. (with 15 polymorphic microsatellites, each one located in different *loci*). Of the 216 accessions collected in the Germplasm Bank, 130 genotypes were differentiated (60%) and 40% were identified as repeated, due to clonality. A total of eight accessions were recognized as commercial cultivars in the Germplasm Bank (4%), being these 'Castell' (1), 'Mantecosa Hardy' (2), 'Roma' (1), 'Blanquilla' (1) and 'Williams' (3). Thirty-three genotypes were identified as repeated in the Germplasm Bank, between them some homonyms and synonyms not described previously; with 100 accessions involved (39%). The most frequent genotypes were those corresponding to 'Mantecosa', 'Urraca', 'Sevillana' and 'Donguindo', all of them reported previously as important for pear crop in Northwestern Spain. These results will allow the restructuring of the Germplasm Bank, eliminating duplications, and providing the correct identification of unique genotypes. As an effect, the study of local crops, in particular the known cultivars of Galicia can diversify Spanish production.

S12.031

Genetic Diversity Revealed by Fluorescent - AFLP Markers for Apricot Germplasms in Shandong, China

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Apricot is originated in China. Shandong is a major province in apricot production in china and has abundant apricot genetic resources. During the long term of evolution and selection, a lot of local apricot cultivars such as Badanshuixing in Tai'an, Honghebao in Jinan, Guanyelian and Yangjiyuan in Qingdao were formed. The genetic diversity of 26 apricot cultivars in Shandong province was analyzed by the fluorescent-AFLP marker techniques in this study. The results showed that the average number of polymorphic *loci* amplified by 8 pairs of fluorescent-AFLP primers was 103.25, the mean percentage of polymorphic *loci* (P) was 87.37%. The total number of polymorphic *loci* was 826. All average Nei's gene diversity index (H) and average Shannon's information index (I) was 0.1935 and 0.3100 respectively. In which indicated that genetic diversity of Shandong apricot was abundant. Apricot cultivars in Shandong were divided into 4 groups at 58.58% level of similarity coefficient. Shangdong apricot local cultivars distributed in all four groups, in which Yubadan and Yangjiyuan cultivars had farther genetic distances with other Shangdong apricot local cultivars and American apricot. So Yubadan and Yangjiyuan apricots were important for broadening genetic diversity of Shandong apricots, breeding new apricot cultivars and creating new germplasms.

S12.032

Development of an Application for Reliable Automated Cluster Calling in Taqman-Based SNP Genotyping

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Single-nucleotide polymorphism (SNP) genotyping using Taqman or related technologies is likely to replace older electrophoresis-based methods for genotyping of horticultural crops in the near future. Being a high-throughput method, massive datasets are generated, requiring sophisticated analysis and storage solutions. We developed a plugin application for the BioNumerics software, which performs a fully automated cluster calling and provides a flexible user interface for visualization and manual overruling of the calls. The plugin performs its automated cluster calling using a multi-seed partitioning algorithm, with the ability to store optimal seed points per SNP marker. Confidence values are reported per batch and for each individual SNP call. Thresholds for calling, algorithm parameters and report options can be specified by the user. All data, including calls, plant sample information and experiment settings, are stored in an industry-standard relational database (e.g. SQL Server, Oracle or MySQL), effectively allowing the construction of large genotyping databases over long periods of time. Additionally, users can take advantage of the sophisticated backup and user management tools available in the database management system and integration with existing storage platforms can be achieved. SNP scores can be further analyzed in BioNumerics using clustering and dimensioning reduction techniques (PCA, MDS and discriminant analysis) or with statistical tools such as multivariate analysis of variance (MANOVA). These analysis tools greatly facilitate common tasks in plant breeding, such as finding the hybrid that is most similar to the recurrent parent during backcrossing and comparing seed lots with representatives of known cultivars in trueness-to-type analysis. In addition, SNP genotyping results can be compared in the same software with results from other techniques such as SSRs, AFLP, Multiplex Ligation-dependent Probe Amplification (MLPA), high-resolution melting curves and phenotypic characterizations.

S12.033

Characterization of Some Local Sweet Cherry (*Prunus avium* L.) Cultivars in Iran Using SSR Markers

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Sweet cherry (*Prunus avium* L.) is one of the most important temperate fruit

crops in Iran. Iran is one of the largest producers of sweet cherries in the world. Therefore, evaluation of sweet cherry cultivars in Iran to for production potential and to provide linkage maps of them is one of the most important steps towards improved breeding of the species. In this research, simple sequence repeats (SSRs) were used to evaluate sweet cherry. A total of 120 alleles were detected by 11 SSR primer pairs, with an average of 10.91 putative alleles per primer combination, and a range from a minimum of 4 to a maximum of 18 alleles detected per primer pair. The size of alleles ranged from 105 to 291 bp. Heterozygosity values ranged from 0.71 to 0.92 and average about 0.86 per primer combination. SSRs' polymorphic fragments were used to calculate a similarity matrix and to perform UPGMA cluster analysis. As results of cluster analysis, 26 cultivars were divided to 7 clusters. Most of the cultivars grouped according to their pedigree.

S12.034

Molecular Phylogenetic Framework of *Citrus* L. and the Origin of Cultivated Types

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Citrus is one of the most important fruit trees in the world and has a long cultivation history. The taxonomy of the genus *Citrus* L. has long been controversial and the genetic origin of its cultivated types has remained unresolved. In this study, amplified fragment length polymorphism (AFLP) fingerprints, nuclear internal transcribed spacer (ITS), and three plastid DNA regions (psbH - petB, trnL - trnF and trnS - trnG) of thirty-seven citrus biotypes of the true citrus fruit trees group (*Aurantioideae*, *Rutaceae*) were analyzed in an attempt to reconstruct the phylogenetic relationships among the key "non-hybrid" wild species of the genus *Citrus*, and identify the genetic origins of its cultivated types. The molecular phylogenetic trees of the true citrus fruit trees group and the key "non-hybrid" wild species of *Citrus* were constructed based on three combined data sets. Our results showed that all the basic biotypes of *Citrus*, i.e., *Papeda*, pumelo, citron and tangerine were of monophyletic origin. Within the genus, *C. medica*, *C. grandis* and the *Papeda* species were more closely related, and they together formed a sister relationship to the genotypes of *C. reticulata*. Based on this framework, the genetic origins of twelve most important cultivated types were identified. Our results provided complete new evidences for the origins of grapefruit, bergamot, Femminello lemon and Eureka lemon, added additional molecular evidences for the origins of Rangpur lemon, and have confirmed the hypothesis for the origins of sweet orange, Valencia sweet orange, sour orange, Goutoucheng, Mexican lime, Guangxi local lemon, and Rough lemon.

S12.035

The Genetic Finger Printing of the Irish Apple Collection Using Single Sequence Repeats in Multiplex Systems

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The vast majority of apples in the Irish Heritage collection are of unknown origin and have only local names. It was therefore decided to fingerprint the collection for comparison with other data bases. Following initial attempts at using SSR's for finger printing the Irish apple collection, it became evident that whilst the methodology worked, it was of limited value unless all the major collections used the same SSR's. Consequently a series of SSR markers were grouped into three sets of four makers each (at East Malling Research Station) where each set of markers had similar requirements for PCR. There multiplexes were then applied to the Irish Heritage Collection so that the data could be compared to that generated for the UK apple collection at Brogdale. This paper reports on the results to date.

S12.036

Assessment of Olive Variability in Latium (Central Italy) through SNPs, SSRs and Morphological Traits

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Olea europaea L. cultivars, grown in Latium region (Central Italy), have been characterized by means of molecular markers as microsatellites and SNPs (Single Nucleotide Polymorphism) and morphological features. High Resolution Melting (HRM) analysis was employed to identify new polymorphisms in functional genes involved in plant and fruit development, and in the synthesis of secondary products. This method allowed the detection of single base sequence variation and to discover genetic mutations, studying the melting or dissociation behavior of DNA fragments, long 300-600 bp. New SNP markers were identified in key genes encoding environmental receptors (phytochrome A), enzymes involved in the synthesis of metabolites influencing the olive oil quality (lupeol synthase and cycloartenol synthase) and cellular function (glycosyltransferase). The identification of SNPs by HRM was confirmed by direct Sanger sequencing of only those amplicons predicted to carry nucleotide changes. Two in-del mutations and 38 SNPs were identified, consisting by 22 transversions and 16 transitions; most mutations were silent or had slight detectable effect on the encoded polypeptide. Genotyping data obtained by microsatellite and SNP markers, were evaluated by PCA and used for constructing a dendrogram by UPGMA cluster analysis. Molecular relationships among cultivars were discussed in relation to the morphological features. The markers developed in this research could be usable for resolving cultivar identity, for providing *loci* for map construction, association studies and markers assisted selection breeding.

S12.037

Identification of the Olive World Germplasm Bank of Cordoba by Molecular (SSR) and Morphological Markers

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Plant germplasm collections include varying levels of genetic redundancy, which hinders their efficient conservation and utilization in breeding programs. For this reason the correct description and identification of the genetics resources conserved in the gene banks is an essential step to improve the accuracy and efficiency of the collection management. A total of 506 accessions of olive from 22 countries of origin included in the Olive World Germplasm Bank of Córdoba (OWGBC) (Spain), were analyzed by 14 SSR markers showing a high genetic diversity. A total of 237 alleles were amplified with an average number of alleles per *locus* of 17. Forty of these alleles were unique alleles (allele present only in a single genotype); the observed heterozygosity (H_o) ranged from 0.967 to 0.121 with an average of 0.766 and the expected heterozygosity (H_e) showed slightly lower values than H_o with a mean of 0.734. The accumulated PI value for all *loci* was 3.172×10^{-16} suggesting a very low probability to find two identical genotypes. Three thousands ninety-five different genotypes were identified in the whole collection by using pair-wise comparisons of the 14 *loci* SSRs profiles. The existence of homonymous, synonymous and mislabeling cases among the olive accessions analyzed were revealed by comparing their genetic profiles and morphological descriptions. Discrepancies and similarities between molecular and morphological identification of the olive cultivars are discussed.

S12.038

Approaches to Determine the Origin of European Plum (*Prunus domestica*) Based on Nucleotide Sequences of Chloroplast DNA

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The genus *Prunus* consists of about 400 species of trees and shrubs and many *Prunus* species are cultivated for their edible fruit and/or for ornamental purposes. The hexaploid European plum (*Prunus domestica*) has overlapping native ranges with other *Prunus* species in Europe and is considered to be a cross between blackthorn ($2n = 4x = 32$, *P. spinosa* L.) and cherry plum ($2n = 2x = 16$, *P. cerasifera* Ehrh.). But the origin of European plum is still disputed. Microsatellites (SSRs) are abundant and well established in the nuclear genomes of both animals and plants. Recently, microsatellites in chloroplast genomes have been found and shown to be highly useful markers for phylogenetic and molecular genetic studies in several plants. A set of 10 chloroplast SSRs (cpSSRs) from 4 non-coding regions were chosen for cpDNA analysis from a total of about 100 individuals of *P. domestica*, *P. spinosa*, *P. cerasifera*, *P. salicina* and interspecific crosses of *P. domestica* × *P. cerasifera*, *P. domestica* × *P. spinosa* and *P. cerasifera* × *P. salicina* and used for molecular phylogenetic approaches to help clarify the origin of European plums. Primers were labelled with Cy5 and Cy5.5 and the PCR products were detected and analysed by capillary electrophoresis using a Beckman CEQ 8000 DNA sequencer (Beckman Coulter, Inc.) by comparison with internal size standards. Cluster analysis was performed with GelCluster V1.0 (BioSci-software), to obtain dendrogram grouping output. Polymorphic information content (PIC) and primer frequency distributions from these analyses together with genetic distances are described and discussed with reference to the putative origins of *P. domestica*.

S12.039

Analysis of Genetic Diversity of Local Olive Varieties in Turkey by AFLP Markers

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Olive (*Olea europaea* L.) has great genetic variation in Turkey as important crop due to its economic value. There are many varieties, types and landraces having good potential as a source of genes for productivity, oil quality and fruit characters which are difficult to differentiate by using morphological characters in different ecological conditions of Turkey. Therefore, genotypic identification studies are important for olive breeding, olive industry and genome analysis. In this study, local olive genotypes which grown in Adana, Artvin, Aydin, Balikesir, Bursa, Canakkale, Gaziantep, Hatay, Kilis, Izmir, Manisa, Mersin and Tekirdag provinces of Turkey were investigated. Most of the genotypes in this study were used first time for evaluation of their genetic structures. Genetic diversity of local genotypes was estimated using amplified fragment length polymorphism (AFLP) markers. Sixty three olive genotypes were analyzed with five *EcoRI*-*MseI* primer combinations. Informative AFLP markers were generated and scored as binary data. Five primer pairs used in AFLP analysis detected 173 *loci*, of which 135 (78%) were polymorphic. Similarity coefficient matrix was computed to cluster the data (software package NTSYS-PC v. 1.80) and to draw precise relationships among the 63 studied olive genotypes. The generated dendrogram revealed three major groups.

S12.040

Comparison of Genetic Diversity among Peach Cultivars Based on Agronomic Traits and SSR Markers

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Peach [*Prunus persica* (L.) Batsch] is one of the most important fruit species in temperate regions, which includes many variably agronomic traits. Agronomic traits has been used to analysis genetic diversity of different peach germplasm for many years, but in recent years, DNA molecular marker techniques such as SSR (simple sequence repeats) exhibit great potential. In this study, the genetic diversity of 50 peach cultivars from China, Japan and America was analyzed with 14 agronomic traits and 13 SSR markers. The mean similarity coefficient of the agronomic traits was tested ranged from 0 to 0.9 with an average of 0.4, and that detected with SSR varied from 0.52-0.97 with an average of 0.75. The results demonstrated that there is no evident difference between the cophenetic correlation coefficients of agronomic and SSR data. Cluster and principal component analysis with phenotypic indicated that the tested cultivars were divided into three groups based on flesh traits. However, SSR analysis grouped the cultivars into two major clusters which based on their origins.

S12.041

Assessment of the Genetic Variability within and among Portuguese Apple Cultivars Revealed by SSRs

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The objects of this study was to analyze the genetic relationship of Portuguese apple cultivars and to compare them with the most common standards present in Europe and in the United States. Samples were collected from apple plantations located in different areas of Portugal (mainland and archipelagos). Of the 12 SSR marker analyzed, 10 primers pairs detected polymorphic DNA fragments. All tested cultivars were discriminate by their allelic profiles. The phylogenetic tree revealed positive correlation between cluster groupings and geographic distances. Portuguese cultivars derived from a common gene pool, and a high gene flow happened among the regions. Results from this study could help the evaluation of the genetic diversity present in Portugal and support the germplasm conservation for apple improvement.

S12.042

Application of Next Generation Sequencing and High Throughput Genotyping Techniques for the Management of Genetic Resources

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Former generations of DNA markers (hybridization-based, RFLPs, and polymerase chain reaction (PCR)-based markers, RAPDs, AFLPs and SSRs) have enhanced genetic resources management over the past two decades. Each marker system meets some of the desirable criteria for use in plant diversity analysis: are reliable, reproducible, codominant, quite polymorphic and evenly distributed across the genome, but most of them are not low-cost or highly scalable. Single nucleotide polymorphisms (SNPs) markers, which are generally developed from sequence information, meet the aforementioned criteria and have also potential for high throughput low-cost genotyping. Next generation sequencing (NGS) technologies (massively parallel DNA sequencing platforms represented mainly by Roche/454, Illumina/Solexa, and ABI/Solid) enable genome/transcriptome sequencing/resequencing to become inexpensive, routine and widespread. These techniques are increasing rapidly the range of crops for which high amount of sequences are available. For example, NGS techniques have been used in fully sequenced model species (*Arabidopsis thaliana*, *Oryza sativa*, *Medicago truncatula*, *Vitis vinifera*), in other deeply studied species with complex genomes and high amount of genomic resources available (*Zea mays*, *Hordeum vulgare*), and even in non-model species for which genomic resources

are unavailable (*Brassica napus*, *Olea europaea*, *Eucalyptus*, *Amaranthus*, *Cucurbita* etc.). In many cases, diverse lines are sequenced (parents of mapping populations and germplasm panels) enabling the discovery of SNPs representative of the genetic diversity of the species. New SNPs collections are being used with high-density platforms that can simultaneously genotype hundreds of DNA samples across hundreds or thousands SNPs. This high throughput genotyping is being used for linkage map construction, genetic diversity analysis, marker-trait association and marker-assisted selection.

S12.043

Use of Microsatellite Markers to Develop New Eastern Black Walnut Cultivars in Missouri, USA

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Eastern black walnut (*Juglans nigra* L.), is valued both for its edible nuts and high quality timber. Production of black walnut nutmeats is centered in the western portion of the species' native range and primarily derived from wild sources. An applied breeding program focused on the development of improved black walnut nut varieties was initiated at the University of Missouri in 1996. Initially, a base population of 84 nut cultivars was assembled and represented by 161 total trees in three replicated clonal repositories. Microsatellite markers have been repeatedly used to define levels of genetic diversity within this collection, as well as facilitate breeding program activities. All base population trees were fingerprinted using 10 microsatellite markers in 2004. Results suggested that the collection actually contained 61 different cultivars, rather than 84 as originally defined. This 27 percent error rate was attributed to past propagation errors, due to the fact that most scion sources were obtained from non-commercial sources. These fingerprint results were also compared using phenological descriptors. In cases when two trees shared similar, but not identical fingerprints, date of first pollen shed and first pistillate bloom more accurately reflected actual cultivar differences in comparison to season length and mean pistillate bloom date. To facilitate breeding program goals, microsatellites have been employed to identify full siblings within a large open pollinated seedling family for the purpose of creating the first mapping population for the species. To date, 493 seedlings have been genotyped representing 17 known pollen parents. In addition, microsatellites were used to confirm the parentage of 122 control pollinated seedlings. Overall, for nearly 10 percent of these hand-pollinated trees (12/121) the assumed parentage was at least partially incorrect, which demonstrates the value of using currently available microsatellite fingerprinting techniques to conserve and improve this important hardwood tree species.

S12.044

Construction of Two Mapping Population and Breeding of Excellent Strains of *Litchi*

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Aiming at genetic mapping and breeding new variety with extremely early or late season, seedlessness and large size fruit, two F1 populations of 'MaguilixJiaohesanyuehong (MS)' and 'MaguilixWuheli (MW)' were constructed in 1998 and 2003 respectively. In the present paper we introduce the performances of these two populations. The juvenile phase, growth traits and fruit qualities of these two F1 populations exhibited continuous variation and extensive separation. By means of comprehensive evaluation of the fruiting individuals, four outstanding strains with good seasonality and/or excellent fruit qualities were selected, i.e.: i 'MS-48', which displayed extremely late season (maturing in mid-to-late July in Guangzhou), extra large-sized fruit (average fruit weigh up to 58.9g, bigger than the existing largest fruit strain in China and abroad), high recovery rate (72.5-77.5%), delicious and with good storability. ii 'MS-68', which displayed very late season (maturing in early-to-mid July in Guangzhou), extra large-sized fruit (average fruit weigh 57.3g), relatively high TSS content (16.8%) and good flavor. iii 'MW-04', which displayed late season (maturing from late June to early July

in Guangzhou), relatively large-sized fruit (average fruit weigh 42.0g), very high seed-abortion/seedlessness rate (93%), high recovery rate (78.0-81.3%) and enjoyable flavor. iv 'MW-37', which displayed high frequency of twins fruit (per single flower), mid-to-late season (maturing in mid-to-late June in Guangzhou), high seed-abortion/seedlessness rate (76.9%), high recovery rate (71.3-74.5%) and very crispy flesh texture. These strains were all new hybrids from severely control pollinations and were helpful to improve litchi variety structure and prolong the fresh fruit supplying period in China.

S12.045

Genetic Similarity Studies in *Arbutus unedo* L. (Strawberry Tree) Using Microsatellites

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Strawberry tree grows around the Mediterranean basin. The edible fruits are used to make an appreciated spirit called "medronheira". Based on studies performed in other *Ericaceae* (*Vaccinium*) SSRs were used to screen 27 wild-growing strawberry trees from nine different Portuguese locations, selected according to their fruit production. Eleven primer pairs were selected based on the information obtained with *Vaccinium* studies: level of polymorphism, number of alleles per locus, number of species amplified, allele scoring quality and repeat motif. Nine of these primers gave amplified products, and no amplification was obtained with the remaining primers. Five of the SSR loci were polymorphic, with 75% mean expected diversity (He), 11.6 mean number of alleles, and 71% of average polymorphic information content (PIC). A mean homozygote excess was found (7%) albeit not statistically significant. Locus CA421F displayed values higher than average: 22 alleles, He=0.90 and 88% PIC. Conversely, locus NA741 showed the lowest PIC (51%), number of alleles (6), and expected diversity (0.57). Estimation of null allele frequency was about 7.6% on average. The linkage disequilibrium test proved that all the loci were independent. Pair-wise genetic similarity was investigated using the Lynch (1990) coefficient, which revealed among genotypes band-shared up to 82%. The Mantel test confirmed the UPGMA tree topology ($r=0.75$; $P<0.001$). No consistent geographic pattern clustering was found, confirmed by the lack of correlation between the genetic and the geographical distance matrices. Reduced gene flows, due to fragmentary distribution and/or to the reproductive system of this species, are a plausible explanation for the results we obtained. They, also, indicated that SSRs markers are useful for further genetic similarity studies, germplasm fingerprinting and conservation.

S12.046

Molecular Phylogenetic and Evolutionary Studies of the Genus *Verticordia* (*Myrtaceae*) by PCR-RFLP

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Nuclear ribosomal DNA ITS region and chloroplast DNA non-coding region trnF-trnVr were studied using PCR-RFLP method to review the phylogenetic relationship of selected species in *Verticordia*. Eighteen species from three subgenera were clustered into two main groups with one representing subgenus *Eperephes* and another representing two subgenera, *Chrysoma* and *Verticordia*. This suggested that two subgenera rather than three are supported in *Verticordia* by this approach. Restriction site variation was found in both nuclear and chloroplast genomes but no amplified fragment length variation was found in trnF-trnVr. Intra-individual variation was detected in ITS region possibly due to restriction site variation among loci, alleles or repeat units. Chromosomal evolution was studied by correlating chromosome numbers with restriction site variations. A negative correlation was detected which suggests that the chromosome number was evolved from 11 downwards.

S12.047

Botanical Characteristics and Molecular Marker Classification of Red Kwao Krua (*Butea superba* Roxb.)

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Red Kwao Krua (*Butea superba* Roxb.) is a protected medicinal plant species of Thailand. In traditional medical practices of Thailand the tuberous roots of Red Kwao Krua (RKK) are used to treat various illnesses and maintain male hormones. Tuberous roots were obtained from forests in six provinces in northeast Thailand in the years 2004 to 2006. Molecular classification by the RAPD technique using 49 clones and 40 primers together with 10 botanical characteristics were performed on the samples. The RAPD technique and botanical classification showed that there were two groups (subgenera) at 32% relatedness. 1) Red Kwao Krua (*Butea superba* Roxb.) included 2 subgroups at 70% coefficient. Subgroup 1 was RKK from Nakhon Ratchasima and Kalasin provinces. Subgroup 2 was RKK from Nakhon Ratchasima and Sakon Nakhon provinces. 2) Tow Pan Say [*Spatholobus parviflorus* (DC.) Kuntze] included 3 subgroups at 84% coefficient from Chaiyaphum, Buriram and Mahasarakham provinces. The dendrogram of botanical characteristics also showed 2 groups at 19% relatedness and were related to the classification by DNA Markers. Mistakes in harvesting traditional medicinal plants are very common: it may be necessary to regulate the traditional medicine industry using modern analytical methods to characterize the products.

S12.048

Implementation of AFLP and SNPs to Characterize the CROCUSBANK World Collection of Saffron and *Crocus* Genetic Resources, CROCUSBANK

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The World Collection of Saffron and *Crocus* Genetic Resources, CROCUSBANK, has been recently created (EU co-funded) with the aim to preserve, characterize, utilize and exploit the endangered saffron crop and its allies. The bank compiles saffron accessions and *Crocus* wild species from native and cultivation areas world wide. The work presented here contributes to the molecular characterization of the bank, thus being applicable in identification of genetically linked characteristics (yield, drought and salt tolerance or disease resistance) and also in species or varieties determination for proper rationalization of the bank. For this purpose, AFLP and SNPs techniques have been employed. The AFLP methodology allowed us to discriminate between species but not between *C. sativus* from different origins. SNPs' inspection was done by genomic DNA high-fidelity PCR of a small set of 6-10 *Crocus* accessions, using primers from 50 *C. sativus* ESTs derived from a stigma library. Forty primers generated specific and reproducible PCR products, which were cloned and sequenced. The average of candidate SNPs found in a single fragment ranged from 0 to 6, and only 2-3 per fragment were further assayed with TaqMan Genotyping probes, in order to genotype a broader set of accessions. This approach allowed us to identify markers present in *C. sativus* and a few of its wild *Crocus* relatives, despite the inherent technical difficulties to analyze a triploid genome. In parallel, up to 50 *Crocus* and saffron accessions have been evaluated in different DNA fragments by High Resolution Melting (HRM) assays. The HRM has provided additional data supporting our TaqMan and direct SNPs sequencing results. All together, these data have proven useful for the evaluation of the bank and it also provides additional evidence for the intriguing issue of determining the hybrid origin of *C. sativus*.

S12.049

High Levels of RAPD and SSR Marker Diversity in Landraces of Watermelon (*Citrullus lanatus*) in Southern Africa

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In Africa, watermelon cultivation is prevalent in drought-prone, semi-arid areas with an annual rainfall below 650 mm. Here, watermelon is grown as a staple food (edible seeds), a dessert (edible flesh), and for animal feed. The fruit flesh can be eaten fresh or cooked. The rind can be pickled or candied, while the seeds are baked or roasted for consumption. Cultivation is based on seed-propagated landraces and farmer varieties that have been integrated with the indigenous knowledge, agricultural practices, food habits and cultural dynamics of the rural communities. Previous studies have reported on relatively low levels of genetic diversity in cultivated watermelon. The investigated material has, however, consisted mainly of US plant introductions and modern watermelon cultivars linked to breeding programmes for disease resistance. By contrast, germplasm maintained in the putative centre of origin in Southern Africa, could be expected to display considerably higher variability. Three different sampling strategies have been used to collect plant material of both wild and cultivated forms of cow-melons (*Citrullus lanatus* var. *citroides*) and of sweet watermelons (*C. lanatus* var. *lanatus*, only known from cultivation); (1) in-depth sampling in the fields of one village in Zimbabwe, (2) medium-scale sampling across the water-melon growing districts in Zimbabwe, and (3) broad-scale sampling across Southern Africa (Botswana, Namibia, South Africa, Zambia and Zimbabwe). Two molecular marker methods are used, random amplified polymorphic DNA (RAPD) and simple sequence repeats (SSR) also known as microsatellite DNA. Similarity matrices obtained with RAPD and SSR, respectively, have been highly correlated, suggesting that for some applications, the less demanding RAPD can be a useful alternative, especially in developing countries. Considerable amounts of genetic diversity was found at all levels, including within-accessions (half-sib families), and contrary to previous reports, sweet watermelon accessions contain as much variability as cow-melon accessions.

S12.050

Genetic Variation in Swedish Bilberries (*Vaccinium myrtillus*) Ascertained with ISSR Markers and Polyphenolic Content

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During the summer 2008, 150 bilberry shoots were collected at three locations 500 km apart, in the South, middle and North of Sweden, lat 57°, 60° and 64° respectively. The shoots were collected in 10 5x5 squares 10 m apart at each location. Each shoot was planted in a pot with acidic soil, and kept in an experimental garden at Balsgård, in the South of Sweden. During the autumn, leaves from these shoots were collected for DNA analysis using 7 inter simple sequence repeat (ISSR) primers which produced 112 polymorphic bands. Fourteen samples did not produce any reliable bands, and were thus excluded from the analysis. Of the remaining 136 samples, 95 were unique genotypes. The resolving power (Rp value) of the primers varied between 12.1 and 4.3 showing the primers were well able to distinguish between the genotypes. Shannon's diversity index, as well as Jaccard's similarity index, showed the variation decreased towards the North, Shannon's index from 0.413 to 0.324 and Jaccard's index from 0.584 to 0.683. During one day in August 2009, leaves were collected from 5 random plants from each location of the collection from 2008, for analysis of total phenol content using Folin-Ciocalteu method. The plants had then been in same soil and at the same place for one year and the environmental influence was therefore considered likely to be reduced. The result showed in contrast to the genetic variation, an increase in total phenolic content towards the North, 162.4, 176.7 and 211.7 mg/g dw, respectively.

S12.051

Conserving Wild Plants for Food and Medicine - The Work of Botanic Gardens

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Botanic gardens play an important role in conserving plant diversity making a major contribution to the implementation of the Global Strategy for Plant Conservation (GSPC). This strategy, agreed under the auspices of the Convention on Biodiversity in 2002, has 16 ambitious targets to be met by 2010. The targets are currently being revised to reflect the reality of climate change and to make explicit their links to the Millennium Development Goals. An important component of the GSPC is the conservation of wild plants for food and medicine. This keynote will address both the overall achievements and challenges of the GSPC and the practical ways that botanic gardens are supporting the conservation of plants important for rural livelihoods. It will emphasise the potential for stronger collaboration between botanic gardens and other agencies for the conservation and sustainable use of plant diversity.

S12.052

Traditional Seed Conservation Practices of Small Farmers in India

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In the face of continuous introduction of improved varieties and new crops, farming communities in India have been successful to some extent in conserving their traditional germplasm. This is mainly achieved through storage of seed and planting material besides regular cultivation of crops that are conserved. The love for native germplasm among rural communities is so high that they have preserved them for posterity without the help of modern methods of storage and advanced scientific knowledge. Methods used for the conservation of many horticultural crops show how traditional practices incorporate the principles of plant material storage. Many of these practices are aimed at preventing loss of moisture and maintaining internal temperature, the primary requirements under tropical conditions. For example, farmers of the Konkan region of Maharashtra separate the seed of jackfruit (*Artocarpus heterophyllus*) from the pulp and dry them under shade. Upon drying, they are coated with red soil, dried and stored in air tight containers in a cool place. Seeds stored in this manner retain viability until the next planting season. Similarly, custard apple (*Annona squamosa*) and jamun (*Syzygium cumini*) seeds are depulped and coated with cowdung slurry and dried. Further, these seeds are stored in airtight containers having wood ash and neem leaves. The storage practice for yam of *Dioscorea alata* is to clean the surface of soil and debris, coat with cowdung slurry and store in a vertical position in a container having wood ash with neem leaves. Another practice is seen in *Momordica dioica* where the entire fruit is allowed to dry before harvesting and storage. These are examples of methods adopted by farmers to conserve their indigenous genetic resources. The paper is a documentation of storage methods and the scientific basis of a collection of horticultural species.

S12.053

Conserving Australia's Unique Rainforest Fruits and Wild Relatives

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Australian rainforests have many unique edible fruits including wild relatives of many worldwide economically important species such as citrus and macadamia. One of the key risks of projected climate change is its effect on eastern Australian rainforests. The interaction of climate change with other threats, such as weeds and

habitat fragmentation, is thus likely to impact on already vulnerable environments such as the World Heritage listed Gondwana Rainforests of S. Queensland and N. New South Wales. Seed banking is a cost effective way of conserving vulnerable species *ex situ* for access to unique diversity for utilisation (e.g. plant breeding and restoration programs). However the seeds of many rainforest species cannot survive the desiccation required in standard seed banking procedures (i.e. non-orthodox seed storage characteristics). This poster reports on preliminary findings on biology and storage characteristics of seeds of significant and/or threatened rainforest fruits such as native pepper, citrus and native ginger. For species identified to have non-orthodox seed storage characteristics, living collections and technology development (e.g. cryostorage) need to be prioritised for *ex situ* conservation.

S12.054

Micronesia Go Local Campaign for Preserving Plant Genetic Resources

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The Federated States of Micronesia (FSM) has experienced a drastic shift to imported processed foods since the 1970s. These dietary and lifestyle changes have led to a neglect of the traditional food system and an epidemic of nutritionally-related diseases (e.g. vitamin A deficiency, diabetes, heart disease, cancers). In Pohnpei State alone, there are over 48 documented giant swamp taro, 50 banana, 133 breadfruit, and 171 yam varieties, but there is concern about the loss of traditional knowledge and plant genetic resources. Many local yellow-fleshed banana and giant swamp taro varieties and other foods have been analyzed and are rich in provitamin A carotenoids and other nutrients and have important health benefits, but are neglected. As part of a global health project led by the Centre for Indigenous Peoples' Nutrition and Environment (CINE), a Pohnpei case study was implemented in a target village aiming to increase local food production and consumption. In the first phase of the project, the traditional food system was documented and compiled in a book along with the other CINE case studies. The Pohnpei intervention implemented from 2005 to 2007 was successful in increasing the number of banana varieties consumed, frequency of banana and giant swamp taro consumed, dietary diversity and improving the attitude in the community towards local food. Methods used included: a participatory, inter-agency, ethnographic approach including social marketing, slogans and acronyms, workshops, container gardening, mass media (radio, newspaper, email, video), postal stamps, genebank collections, posters, charcoal oven production, competitions, recipes and leaflets. The "Let's Go Local" campaign promoted local foods for their "CHEEP" benefits: Culture, Health, Environment, Economy and Food security. Efforts have now been initiated to expand the project to other communities in Pohnpei and FSM, and there is interest also in establishing "Go Local" campaigns in other Pacific Island countries.

S12.055

Towards a Strategy for the Conservation and Use of Underutilized Crops in the Pacific

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Pacific Islanders traditionally have enjoyed diverse ways to achieve food security, however numerous factors have contributed to a weakening of local food production from poor investment in agriculture to urbanization to the availability of cheap food imports. An increasing reliance on imported food products has not only affected the health of Pacific communities, but also threatened food security, as shown with the recent food crisis. Climate change poses further challenges to food security, through its impact on food production, health, infrastructure, the ability of countries to import food, and the ability of households to purchase food.

Against this background, members of the Pacific plant genetic resources network (PAPGREN) met to address the following constraints: (1) Lack of information/documentation on underutilized species in the Pacific; (2) No regional priority list of underutilized species; (3) Lack of policy support from various government agencies; (4) Poor awareness at all levels about the value and potential of underutilized species. A regional strategy on "Crops for the Future in the Pacific" was developed containing the following key elements: 1. Generation and collection of knowledge/research 2. Communication and dissemination 3. Policy advocacy 4. Market development 5. Partnerships 6. Capacity building and institutional strengthening The priority crops identified for the region include breadfruit (*Artocarpus altilis*), bananas of the Fe'i group and/or Pacific plantain, Polynesian chestnut (*Inocarpus fagifer*), *Pometia pinnata*, giant swamp taro (*Cyrtosperma merkusii*), bele (*Abelmoschus manihoti*), the lesser aroids *Alocasia* and *Xanthosoma* and, particularly for the atoll islands, *Pandanus* spp. Priority research and development activities on these crops include increased regional exchange of information on ongoing activities to determine gaps and to identify areas of future research focus. Particular emphasis is put on increased capacity for plant breeding and the use of traditional knowledge on the use and conservation of these important crops.

S12.056

Farmer Participatory Development of Cercospora Leaf Spot Disease Tolerant Varieties in Ghana

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Involving farmers in the development of improved cowpea varieties is the most effective way of enhancing acceptability and adoption. A study was conducted to involve farmers in the planning, evaluation and selection of cowpea varieties that possess farmer-preferred properties and tolerance to *Cercospora* leaf spot disease. In this study, farmers and researchers identified the incidence of *Cercospora* leaf spot disease in the forest and forest-savanna transition zones as one of the main constraints of cowpea production causing excessive leaf spotting and defoliation leading to yield losses of 50 to 70% on farmers' fields. Four improved varieties: Nhyira, Asetenapa, Ayiyi and Bengpla and 8 (eight) *Cercospora* leaf spot disease tolerant lines were involved in the breeding programme. The *Cercospora* leaf spot disease tolerant lines included collections from across the country and from the International Institute for Tropical Agriculture (IITA). A preliminary evaluation at CSIR-Crops Research Institute, Fumesua showed that IT97k-499-35 had a 20% and 2 while Asetenapa had 60% and 3.5 for mean incidence and severity scores respectively. The remaining lines showed a mean incidence and severity score of 0% and 1 respectively. The eight (8) *Cercospora* leaf spot disease tolerant lines have been crossed to the 4 improved varieties yielding F1 progenies followed by a first backcross generation.

S12.057

Diversity and Utilization of Indigenous Leafy Vegetable in Urban and Peri-Urban Production Systems in Côte d'Ivoire

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Indigenous leafy vegetables are important sources of vitamins and essential minerals. They constitute a major diet for the growing resource-poor urban populations. However, the limited knowledge on improved agronomic practices combined to the high urban population pressure threatens their development. To promote a sustainable production system, a study was undertaken to assess the diversity of indigenous leafy vegetables in two major cities, Abidjan and Yamoussoukro, in Côte d'Ivoire. The farming practices and the market value of indigenous leafy vegetables were analyzed. The dry matter content, as well as Vitamin C and Phosphorous concentration of each accession, were determined. The study revealed a large diversity of indigenous leafy vegetables produced in the urban and peri-urban areas. More than

26 species were inventoried. Almost all cultivars were land races and seed production and preservation remain rudimentary. The farming practices varied with plant species and include pure, mixed and spontaneous cultivation methods. The indigenous leafy vegetables are low-yielding in farmer's production systems. Whereas men dominate the production (80%) sector, the marketing of indigenous leafy vegetables was mainly conducted by women (98%). A total of 56 plants accessions were collected. Morphological characterization of these accessions was realized. That morphological characterization shows an intra-specific diversity in each species. In this study, some accessions of amaranth, celosia, nightshades and jute mallow with good characteristics such as high leaf number, large leaf, earliness, flowering lateness and high production were identified. Selection of these accessions was undertaken for further improvement program. Based on a combined analysis of production potential, nutritional values and the market value of the accessions, the most important indigenous leafy vegetable species were identified. Three groups of importance and risk levels were identified. A strategy to preserve the indigenous leafy vegetables in Côte d'Ivoire is proposed by, the selection of best types or varieties.

S12.058

Ethnobotanic and Genetic Variability Studies in Barhal (*Artocarpus lakoocha* Roxb.) in Central Uttar Pradesh

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Lakoocha (*Artocarpus lakoocha* Roxb.) is a large, deciduous, underutilized fruit tree native to Sub Himalayan region. Lakoocha is used by rural folk as medicine for skin ailments, boils, as purgative and high carotenoid content is beneficial as antioxidant. Fruits/male flowers are used for culinary purpose. Unripe fruits with seeds are used in pickles and vinegar. Ripe fruits are used for making 'halwa' and 'jellies'. Leaves are used as fodder for lactating animals. 23 genotypes having desirable traits were collected from 17 different villages of Kasia Block and physico-chemical analysis of fruits following standard procedures was done. The genotypes collected showed considerable variation in vegetative parameters and fruit physico-chemical studied. The tree characters studied showed considerable variation viz., height of tree (4.80-14.80m), canopy of tree (N-S) (4.20-12.6), canopy spread (E-W) (1.67-4.2), trunk girth (0.31-1.67m), trunk diameter (0.099-0.053), trunk cross section area of the trees (0.007-0.221m²). Variation among genotypes for foliage characters ranged in leaf length (18.13cm-27.30cm), leaf breadth (9.58cm-16.38cm), petiole length (1.90cm-3.43cm), internodal distance(1.83cm-3.50cm). The fruit characters viz., fruit length, fruit breadth, fruit weight and fruit volume were studied and these were found to be ranging from 3.31cm to 7.51cm, 4.23cm to 7.32cm, 51.00g to 210.33g and from 47.33cm and 204.33g, respectively. Specific gravity (1.076-0.997), pulp weight (26.00-156.33), peel weight (6.667g-25.00g), pulp: peel ratio (2.327-9.39), number of flakes (6.67-29), core length (1.10cm-2.63cm), core diameter (0.83-2.30cm), no. of seeds/fruit (5-25.00), seed length (0.80 cm-1.57cm.), weight of seeds/fruit (3.3/fruit-19.66/fruit), seed breadth (0.80cm-1.47cm), TSS (22.360B-13.20B), acidity (0.77%-2.67%).

S12.059

Current Efforts in Bambara Groundnut Breeding in Ghana

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Bambara groundnut (*Vigna subterranean* (L) verdc.) is an indigenous legume mainly grown for subsistence by small holder farmers in sub-Saharan Africa. In spite of the importance of bambara groundnut, it has not received much research attention compared to other legumes, especially regarding its genetic improvement. In recent times, there has been a renewed interest in improving the productivity of Bambara groundnut in Ghana. Consequently, a collaborative research effort between CSIR-Crops Research Institute in Ghana and the University of Nottingham, UK led to the first ever successful cross in bambara groundnut. This process of hybridization is a significant scientific achievement which will open several opportunities

and possibilities for breeding improved varieties of bambara groundnut. Until this achievement, the institute had only been involved in evaluating existing landraces collected from across the country. Consequently, the Bambara breeding programme at the CSIR-Crops Research Institute, Ghana obtained 67 F3 bambara seeds of a cross between two bambara parental lines: Dip C × Tiga Nicaru. The F3 seeds and seeds of the parental lines were sown as F3 generation in 2008 and evaluated in the forest and semi-deciduous agro-ecologies of Ghana. Eighteen (18) selected lines constituting 27% of the total lines obtained showed farmer-preferred characteristics such as an almost cream to complete cream seed colour, high yields, large seed size, high number of pods per plant and number of seeds per pod. Results for days to 50% emergence and flowering ranged between 7 – 9 and 38 - 46 days respectively.

S12.060

Promising Practices of Medicinal Plants Conservation among Gond and Halba, the Forest Dwelling Tribes of Bastar, Chhattisgarh

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The present paper aims at critically examining the outcome of community based interventions on medicinal plant conservation initiated by a consortium of NGOs at Bastar and how the synergy between un-codified folk system of medicinal knowledge and codified local medicine system was streamlined. The paper also critically assesses the lessons learnt from such interventions towards reforming policy and practices in regard to community based medicinal plant conservation. The evaluation of various project components such as building on medicinal plant knowledge of local healers, village botanists programme, village Biologists workshop and developing community Biodiversity Registers validate that the use of local medicinal plant knowledge of Shamans locally known as Boigas has facilitated identification of rare and endangered plants. The biological resources in the indigenous territories of Chhattisgarh consist of various natural sources of agricultural, medicinal, ecological, veterinary and cosmological potencies which ensure equilibrium between local environment and social health of the tribal communities inhabiting in the forest villages. The traditional community based approach to medicinal plant conservation influence the customary practices, cultural resources and local knowledge systems not only among Gond and Halba communities but also among other indigenous communities. The above cultural practices, both customary and non-customary, prevalent among Gond and Halba tribes of Chhattisgarh are not only inherited territorially but also continue to evolve under influence of individual innovations and local environment. There is an urgent need to promote supportive policy and legislative framework from the insights and success stories of community based approach to medicinal plant knowledge and conservation.

S12.061

Promotion of Indigenous Vegetables in Asia

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Diversifying crop production and diets is important to combat malnutrition among the poor in developing countries, generate income, and sustain ecosystems under threat due to human intervention. Crop and diet diversification could be achieved by making better use of resilient indigenous vegetables that easily adapt to degraded, drought-prone, flooded, or saline land, areas of which are increasing due to climate change. Despite the recognized importance of indigenous vegetables to combat malnutrition and poverty, many remain underutilized due to a lack of information on their use, health benefits, field performance, and input requirements. A lack of varieties or lines for widespread distribution and uncertainty on how these plants can fit into common production systems further curtail their use. The underutilization of indigenous vegetables endangers their genetic resource base. AVRDC - The World Vegetable Center is currently undertaking project activities in Indonesia, the Philippines, and Taiwan to promote the conservation and use of indigenous vegetables. The Center focuses on the rescue, improved conservation, and seed increase of promising lines, variety trials and participatory evaluation of selected

accessions, collecting, and training personnel in germplasm management. Priority crops differ from country to country and comprise, among others, the following species: *Moringa oleifera* (drumstick tree), *Ipomoea aquatica* (water spinach, kangkong), *Amaranthus* spp. (amaranth); *Brassica oleracea* var. *alboglabra* (Chinese kale), *Brassica rapa* subsp. *campestris* (edible rape), *Ocimum basilicum* (basil), *Glycine max* (vegetable soybean), *Hibiscus sabdariffa* (roselle), *Basella* spp. (Malabar spinach), *Cucurbita moschata* (pumpkin), *Psophocarpus tetragonolobus* (winged bean), *Momordica charantia* (bitter melon), *Luffa acutangula* (ridged melon), *Lagenaria siceraria* (bottle gourd), *Benincasa hispida* (wax melon), and *Trichosanthes cucumerina* (snake melon).

S12.062

Famine Food Plants - Genetic Resources for Staple Crops for the Future

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Famine food plants are a little-known category of underutilized species. These plants are sought out in times of famine and other conditions of environmentally-induced food scarcity. Because they are adapted to some of the most extreme weather conditions Nature can create, they represent a valuable source of plant genetic material, for the development of new, affordable staple crops, for environmentally at-risk eco-zones. In addition, some of the very few famine food plants that have been analyzed, show unexpectedly high nutrient values. As nutritional value will be a base-line criterion, for identifying candidates for selection, growth trials and breeding, further work is needed, to ascertain the nutrient composition, of the large corpus of these as-yet unstudied famine food plant species.

S12.063

Preliminary Selection of Mangaba (*Hancornia speciosa* Gomes) Genotypes with High C Vitamin Content

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The mangaba (*Hancornia speciosa* Gomes) is a native fruit tree from Brazil. Despite its cultivation being yet practiced under undeveloped systems it is of great social, economic, and cultural importance in areas of its occurrence. Its fruits represent incoming source for many families of coastal tableland areas and are very appreciated by consumers by their sensorial characteristics. There were evaluated 12 fruits from 100 individual trees at five natural areas of mangaba occurrence (Aracaju, Barra dos Coqueiros, Pirambu, Itaporanga d'Ajuda, and Estancia) in Sergipe State, Brazil, aiming to identify and select genotypes with high C vitamin content. Fruits were picked at a uniform maturation stage and had their physic and chemical characteristic quantified in the Embrapa Coastal Tablelands Eco-physiology Laboratory. The trial was carried in a completely randomized design with five treatments (populations) and 20 replications. Data of literature show C vitamin content of around 200 mg/100g fresh matter (MF) in mangaba fruits. In the present work averaged values higher than 400 mg/100g MF were found for all studied populations, with some genotypes presenting 600 mg/100g MF. Fruits originated from the natural population of Itaporanga d'Ajuda presented the highest C vitamin contents (690 mg/100g MF). These results prove the efficiency of a preliminary selection of mangaba genotypes considering the functional attributes of the specie.

S12.064

Morphological Variation in *Ziziphora tenuior* L., a Medicinal and Aromatic Plant

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The uses of traditional medicinal plants in relieving symptoms of disease and curing

various infections dates back to many centuries. In recent years considerable interest has developed in Iran and other countries in the collection and extended use of indigenous and introduced plants for medicinal purposes. *Ziziphora tenuior* is an annual member of family *Lamiaceae*, widespread throughout Iran. Traditionally this species is used as an aromatic flavor, antiseptic, antibacterial and antiparasitic agent. The most important components of its essential oil are Pulegon, Limonell and Cineole. It grows usually in cold and semidry regions with 250-350 mm annual precipitation at about 1000-1800 m altitude. To investigate morphological variation of this species, 365 specimens belonging to 22 populations were collected from different parts of Iran. Thirteen quantitative and qualitative characters were studied and analyzed by cluster analysis using Ward method. According to the results all the characters examined showed significant variation among the populations. Some characters showed significant correlations with each other. Number of branches showed positive correlation with stem length and number of flowers per inflorescence, stem length had positive correlation with leaf length and inflorescence length, and leaf length with leaf width and inflorescence length. Based on studied characters, specimens of some populations including Qazvin, Karaj, Fars, Yasuj, Sanandaj and Zanjan clustered together showing a closer similarity with each other. The most similar population groups are those from Chitgar, Sorkhehesar, Robatkarim which are collected from close places. Although from different geographical regions, interestingly Yazd and Mashhad located in the second group, and Kerman and Thaleghan in the final group. The remaining populations showed a high degree of variation. According to the results a high degree of inter-population morphological variation was also present.

S12.065

Advances in Breeding Borage for Gamma Linolenic Acid Production

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Borage (*Borago officinalis* L.) is an annual plant that has been used from ancient times for its culinary and medicinal properties. Nowadays, international interest in borage has been renewed because the high percentage of gamma linolenic acid (GLA) contained in its seeds. This fatty acid is an intermediate of prostaglandin E1 and its derivatives; therefore supplementation of the human diet with vegetable oils containing GLA is currently used for the treatment of health problems related to deficiencies in this fatty acid. The most common commercial sources of GLA for pharmaceutical uses are evening primrose and borage. Large-scale commercial production of borage seeds is difficult because of its indeterminate growth habit and its seed shattering habit that reduce the seed yield. The objectives of this work were to show the results obtained in the different approaches followed at IAS-CSIC in order to increase the GLA production in borage. The material evaluated consisted of different accessions of: a) wild (blue flowered) and cultivated (white flowered) Spanish borage, b) wild Argentinean borage, collected in different geographical regions, and c) genotypes of borage selected from the two different mutants obtained after EMS treatment of the seeds: mutant B with high number of ovules, and mutant C with partially opened flowers and seed retention. There are important differences in oil content and fatty acid composition among the accessions studied, and genotypes with GLA content higher than 28% of the oil have been selected both in Spanish and Argentinean borage accessions. GLA content is negatively correlated with oleic acid content, and positively correlated with erucic acid. No correlation has been found between GLA and oil content. Seeds from both mutant B and C showed normal oil and fatty acid composition, but higher seed yield potential.

S12.066

Germination Rate of Outbreed and Inbreed Seeds in Four *Papilionoideae* - *Fabaceae* from Northwestern Argentina

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Crotalaria pumila Ort., *Desmodium pachyrrizum* Vog., *Macroptilium erythroloma*

(Benth.) Urban, and *Indigofera parodiana* Burk. (*Papilionoideae*, *Fabaceae*) are potentially ornamental species and may be used for forage given its high nutritional value. They have a mixed breeding system and are pollinated by small and medium-sized Hymenoptera. Previous studies confirmed their capacity for autonomous self-pollination. Here, we compare the germination rate under natural (free) pollination and autonomous self-pollination. We studied natural populations located in Lerma Valley, Salta Province, Argentina. The populations are located in an Eastern Andes seasonal rainforest (Yungas). In January and February of 2009 we marked 25 flowers and 25 floral buds from 30 individual plants per species and population. In each plant, we performed two treatments: (a) natural pollination (control) in which the marked flowers were exposed to the natural agents of pollination, and (b) autonomous self-pollination in which the marked floral buds were bagged throughout their flowering period. In March and April of 2009, seed pods were collected. Seeds were scarified with concentrated sulfuric acid for 5 minutes. Three replicates of 25 seeds for each treatment were then sown in Petri dishes and germinated at 25 °C ± 2 °C with 24 h artificial light. Germination was monitored daily for 30 days, where germination was defined as radicle emergence of 1.0 mm. In all the species, the germination rate of outbreed seeds was greater than that of inbred seeds, but these differences were not statistically significant, except for *D. pachyrrizum* ($X^2 = 6.32$; d.f. = 1; P = 0.012). This suggests that the outbreed progeny of *C. pumila*, *I. parodiana* and *M. erythroloma* have no advantage over the inbred progeny.

S12.067

Use and Conservation of Underutilized Crops in Mozambique

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MOZAMBIQUE

Indigenous Vegetables and Fruits play an important role in food security, food diversification, in nutrition and as food alternative particularly when drought affects the normal production. In Mozambique there are different species of indigenous vegetables and fruits differently utilized and in some places some of them are purely unknown or underutilized. Before Independence of Mozambique (1975), some surveys were conducted and literatures produced as example on "Plantas Silvestres de Moçambique com interesse alimentar" by de Carvalho, 1968, on "Listagem de Plantas Alimentares Expontâneas em Moçambique" by Calane da Silva, 1991, on "Survey of Literature on Mandate Vegetable Species of the SADC Plant Genetic Resources Centre, occurring in Mozambique" presented to SPGRC, by Tembe, 2002, on written presentation "Indigenous Vegetables and Legumes, Importance, Use and Marketing in Gaza Province, by Tembe, edited at *Acta Horticulturae* n° 752, Proceedings of the 1st International Conference on Indigenous Vegetables and Legumes Prospectus for Fighting Poverty, Hunger and Malnutrition", 2006. Lutheran World Federation, Mozambique, on implementation of its Community Development Program, conduct activities on Food Security and Nutrition, by introducing improved production technologies, introducing new crops like exotic vegetables, introduction of improved crops varieties and giving also high importance to Indigenous Vegetables and Fruits, like *Amaranthus* spp, *Corchorus* L., *Momordica* L and others, to Indigenous Fruits like *Vangueria tomentosa* Hochst, *Selerocarya caffra* Sond, *Strychnos espinosa* Lam and others, by improving the use, doing processing and improving conservation of both vegetables and fruits, involving training of community groups of women. On vegetables, the improvement of use and consumption is by introducing ingredients like coconut milk, pounded nuts from Canhi (*Selerocarya caffra*, indigenous fruit), pounded cashew nut, use of tomato, onion as traditionally indigenous vegetables are just boiled in water or with some pounded groundnut. Persistent drought in the country, particularly at Southern part, affects fresh vegetables becoming scarce and to mitigate these effects, women are trained on processing of both exotic and indigenous vegetables leaves by drying on the sun which improves conservation for long and to be used when due to drought there is shortage of fresh vegetables. For Indigenous fruits, women are trained on processing into juice and jam to improve use and conservation. These improvements bring diversification on preparation and use of vegetables and fruits and bring an important aid value to the leaves and fruits by improving nutritional values and as opportunity for income generation business. This Presentation recommend the need to call for

the attention of politicians and scientific institutions, to consider these subjects as part of research, production, processing, use and marketing policies, strategies and programs definition, to give better value to the underutilized crops as they are local available resources and can coexist with exotic crops to contribute for food security, nutrition and economics of the households, communities and the countries.

S12.068

More Efficient Conservation and Use of Vegetable Genetic Resources in Europe: ECPGR Achievements

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The European Cooperative Programme for Plant Genetic Resources (ECPGR) is a regional network funded by the European countries and coordinated by Bioversity International. The Vegetables Network (<http://www.ecpgr.cgiar.org/Networks/Vegetables/vegetables.htm>) is one of the crop specific ECPGR networks. It consists of six Working Groups (WGs), *Allium*, *Brassica*, Cucurbits, Leafy vegetables, *Solanaceae* and Umbellifer Crops, in which representatives from 42 European countries participate. Sharing responsibilities for the *ex situ* conservation of European germplasm is the highest priority of the Vegetables Network. It is foreseen that the rationalization of the collections will lead to higher cost-efficiency, and improvement of germplasm documentation and quality. These efforts will be continued within the framework of AEGIS (A European Genebank Integrated System) (<http://www.aegis.cgiar.org/>). Challenges for the Vegetables Network will be to identify the so-called Most Appropriate Accessions (MAA) for each crop for their inclusion in the decentralized European Collection, as well as to define agreed crop specific technical standards for conservation. Among the achievements of the Network in recent years, there are the development of Central Crop Databases, Minimum quality standards for collection management of seed-propagated crops and cryopreserved material, safety duplication improvement and definition of minimum characterization descriptors. Several EU-funded projects have initiated and accelerated the activities of the WGs. Apart from further improvements within the frame of AEGIS, the Network is planning a number of other initiatives, such as improving collaboration at the global level (*Allium*), developing molecular characterization protocols (lettuce), filling the gaps in the conservation of wild relatives (*Brassica* and Umbellifer crops), and improving its communication with the scientific community and the public at large.

S12.069

Strategies for Effective Maintenance of Vegetable Germplasm

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Vegetable crops are valuable assets of nature to people. They are popular amongst large group of people due to their high nutritative value and good taste. They supply essential nutrients and minerals for proper functioning of body. A wide genetic diversity exists in nature for different species of vegetable crops. It helps the plant

to survive under heterogeneous environment. Also it provides raw material to plant breeder in evolving new varieties with objectives of improved yield, quality and resistant to various biotic and abiotic stresses. The increase in crop production is primarily due to gene contribution from existence gene pool. Genetic uniformity makes a crop vulnerable to epidemics of insects pests and diseases as it happened in past. Conservation of genetic resources is vital not only crop improvement but also for the very survival of the species in time and space. Over exploitation and replacement with hybrids and improved cultivars as well as exotics caused a greater harm to native genetic diversity. The need for germplasm conservation of vegetable crops is widely realized due to increased economic importance, narrow genetic base, gaps in existing collections, and prevalent of genetic erosion. The main approaches for germplasm conservation are *in situ* and *ex situ* means. However an ideal one should be simple to practice, capable of retaining genetic stability and economical to use. Various storage factors were contribute to it and they were employed to minimize the deterioration thereby extends storability in different vegetable crops. As a result seeds remained viable for 25 years in onion, French bean, pepper, cluster bean, okra, radish, watermelon, pumpkin and for 20 years in tomato, eggplant, muskmelon, cucumber. The seedlings emerged from stored seeds were healthy, vigorous and free from any deformities. The details of finding shall be discussed during presentation.

S12.070

North American Continent - A New Source of Wild *Lactuca* spp. Germplasm Variability for Future Lettuce Breeding

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The wild relatives of crops represent essential germplasm for plant breeding. That's why the expeditions to unexplored areas of the world have been organized. In the years 2002-2008, we undertaken the missions in the USA and Canada to search for wild and weedy *Lactuca* species. Altogether 16 states in the USA (Arizona, California, Colorado, Idaho, Iowa, Minnesota, Montana, Nevada, New York, North Carolina, Oregon, South Dakota, Utah, Washington, Wisconsin, Wyoming) and two provinces in Canada (Ontario and Quebec) were visited. In total seven wild and weedy *Lactuca* spp. (*L. serriola*, *L. saligna*, *L. virosa*, *L. canadensis*, *L. biennis*, *L. floridana*, *L. ludoviciana*), and interspecific hybrid (*L. canadensis* × *L. ludoviciana*) were observed and collected in 200 locations. The largest number of localities was visited in California (56), however, the largest number of *Lactuca* species (5) was recorded in Iowa. *L. serriola* was the most common, observed in all states and provinces visited, except of North Carolina and New York. From an orographic viewpoint, *L. serriola* was recorded from 6 masl to 3524 masl. *L. saligna* was recorded only one time in Salinas (California). *L. virosa* was recorded in Washington. The typical habitats of *L. serriola*, *L. saligna* and *L. virosa* were along transport corridors (e.g. roadsides, road ditches, parking sites, grassy slopes) and ruderal places. Native North American taxa *L. canadensis*, *L. floridana*, *L. ludoviciana* were recorded only in Iowa. *L. biennis* was found only in Canada (Saint-Patrice-de-Sherrington, Southern Quebec). *Bremia lactucae* and *Golovinomyces cichoracearum* were recorded on some *Lactuca* plants. The collected seed samples have been maintained in the *Lactuca* working collection at Palacký University and used in research of germplasm biodiversity and to enrich diversity for lettuce breeding.

S12.071

Sweetpotato (*Ipomoea batatas*): A Challenging Crop for Food and Nutrition Security under Sustainable Agricultural System

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Micronutrient malnutrition, primarily due to diets, poor bioaccessibility and

bioavailable carotenes and minerals is now widespread in child and woman health especially in poor and developing world. The crop-based approaches with sweetpotato of both white and orange-fleshed types have great potential in alleviating nutrient deficiencies. Sweetpotato has proved to yield higher amounts of micronutrients and calories per unit area under adverse climatic conditions and low-input regimes than any other crop. One of our alternative options for combating the malnutrition is to enhance food-based strategies that aim at modifying people's diet through orange-fleshed sweetpotato (OFSP) which have high level of β -carotene and excellent source of nutrients which combat vitamin-A deficiency syndrome. Although vitamin-A deficiency is not a critical issue in India, this crop substitutes other. We therefore project some Indian elite heirloom sweetpotato cultivars of both white and orange-fleshed types which are highly ranked for its nutrition profiles including carbohydrate, protein, minerals and β -carotene content. The storage root of OFSP cultivars like Kamala Sundari (Orange Beauty), 90-101, S-1221, S-61, ST-14 have attractive orange colour flesh with red to orange skin and rich in dry matter and carotene content than the white-fleshed cultivars. All India Coordinated Research Project on Tuber Crops, India gives efforts for inter-intra country germplasm characterization, participatory evaluation and selection of varieties with desired characteristics of orange-fleshed sweetpotato for livelihood development and nutrition education to the resource poor farmers and tribal communities. Production of virus free, healthy vine cuttings of sweetpotato has been developed to counteract virus infection of propagation material after a few years by frequently replacing it with healthy material.

S12.072

A Plant Type of Garlic (*Allium sativa* L.) with Biological Seeds and Hard Coated Clove-Lets

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Garlic, one of the oldest known spice crops, is normally sexually sterile. However it was found that one genotype, collected from the North Eastern Himalayas during 1999-2000, produces a large number of florets per umbel and has the potentiality of true seed production. The genotype had some other economic attributes over the other cultivars. Though the cloves of this genotype were less pungent than other genotypes usually grown for commercial purposes. Another characteristic feature of the genotype was, that it produced very hard coated outer clove-lets, (19 to 27 in number per plant), either from the base of the main clove or from the fibrous roots of the main cloves. The breadth of its leaves was more (2.1 cm to 4.6 cm) than other types. The plants were grown at an altitude of 4,500 feet above the mean sea level, and studied for three years during the winter seasons of 2003-04, 2004-05 and 2005-2006 respectively, at two locations (Kalimpong and Dalapchand, district – Darjeeling, West Bengal, India). The genotype produced very large sized single cloves i.e. without any clove-lets, weighing up to 102 gm (57.02gm- 102.00gm), when planted at a closer spacing of 10 cm \times 10 cm. Observations were recorded on plant height (up to the tip of the umbel), plant girth and number of leaves/plant, on twenty five randomly selected plants in each plot in each location and in each year. Days to 75% flower initiation, days to maturity, and crop yield (weight of cloves and weight of inner and outer clove-lets together) were recorded on individual plot basis. Number of florets per umbel, yield per plant, length and circumference of the cloves, number of inner and outer clove-lets/clove, length and breadth of inner and outer clove-lets, weight of inner and outer clove-lets, number of inner and outer clove-lets per clove, length and circumference of inner and outer clove-lets were recorded after the harvest of the crop, again on twenty five randomly selected plants. At normal spacing the cloves produced 4 -9 large inner clove-lets per clove. The weight of such single clove-lets was as high as 2.53 g. It produced very tall spikes and the height of the plant was as tall as 136 cm. The spikes produced more than 750 florets per umbel. The florets produced fertile pollen grains and the percentage of fertile pollen grains was 84.72 per cent. The color of the florets was creamy white. These florets set true seeds (26.63%). The color of the seeds was brown. However the germination percent-

age of the seeds, under natural condition, in all the three years was nil. The outer clove-lets showed viviparous germination. Plants raised from those outer clove-lets showed that the yields were comparable, with the seedlings raised from the usual inner clove-lets. The scale of the cloves and the inner clove-lets were thin and could be peeled easily. However, the scale of the outer clove-lets was hard and thick, and difficult to peel. The genotype took about 180 - 200 days to mature. The yield per plant of this genotype was 122.20 - 158.63 g after sun drying. Though the seeds obtained from this plant type did not show any germination, yet it leaves a great scope to study the plants in great details. Proper drying of the seeds, immediately after harvest may give some interesting results. The plants has some other economic characters, which could be taken into considerations if the germination of these seeds is be achieved in future.

S12.073

Effects of Heat Stress on the Antioxidant Enzymes in Broccoli (*Brassica oleracea*)

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The objectives of this work were to clone the catalase (CAT) gene from broccoli and measure the regulation of CAT gene expressions under heat-stress conditions. Heat-tolerant (Shin-Fa, SF) and heat-sensitive (Green Magic, GM) varieties of broccoli were used. Plants were subjected to high-temperature treatments (30, 35, and 40 °C) and unstressed condition (25 °C, as the control) for periods of 0, 6, 24, 48, and 72 h in a growth chamber. Our results show that the genotypes responded differently to heat stress according to their various antioxidant enzymes and physiological parameters. SF plants had significantly higher CAT activity, water potential (WP), and a variable fluorescence/maximum fluorescence (Fv/Fm) value, but lower electrolyte leakage (EL) than did GM plants with 35 and 40 °C treatments over time. CAT gene expression profiles were well matched with the data for CAT enzyme activities in SF plants under heat stress, suggesting that CAT is involved in hydrogen peroxide detoxification which helps overcome the stress induced by heat. Full-length of the broccoli CAT cDNA was 1768 bp, including 1476 bp of an open reading frame in the CAT gene. A phylogenetic analysis of the deduced amino acid sequences of CAT indicated that plant CATs diverged into two major clusters. Genomic DNA Southern blot analysis revealed that the CAT genes belong to multigene families.

S12.074

Root Colour as an Important Feature of Carrot Origin and Genetic History

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During carrot history and cultivar development, new types based on root colour appeared. The center of origin of the cultivated carrot is believed to be in the Afghanistan region, with mainly purple carrots. Spread through Middle East and Spain, the first cultivated carrots in Europe in the 1100s exhibited purple or yellow roots. White and orange coloured carrots were first described in the 1600s, with the subsequent development of the orange type. In the 1700s, the pink carrot was developed in Asia, probably from the Afghan type. If the main dates of description of a coloured type are well known, the original material from which it has been developed is still in discussion. For example, several hypotheses exist about the origin of the orange type, first described in Holland. We have studied the diversity among coloured types of carrot with combined approaches: historical data, pigment variation, carotenoid biosynthesis genes diversity. Our results confirm some hypotheses. A general scheme on the origin of the different coloured types will be presented. The impact of this genetic history on genetic resources management and core-collection definition will be discussed.

S12.075

Evaluation of Salt Tolerance in Tomato Cultivars under *in vitro* Culture

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The response of six cultivars tomato (*Lycopersicon esculentum* Mill.) callus to salt stress was investigated. Callus relative growth rate (RGR), dry matter percentage (DM), osmotic potential and proline were evaluated. Significant differences were found among cultivars. 'PS-10' had the highest RGR, while 'Roma' had the lowest under salt levels during the incubation period. With increasing salinity levels in the media decrease RGR and increased DM and osmotic potential in all treatments compared with the control. In all cultivars proline levels increased in response to salinity stress. High callus formation correlated with low proline content. 'PS-10' and Imperial have highest callus formation and lowest proline content. Significant differences were reported in regeneration rate under salt treatments. 'PS-10' the highest and 'Roma' the lowest had regeneration rate. It is concluded that more the salt tolerant genotype more is the reduction in osmotic potentials and proline and less turgor of the callus tissue.

S12.076

Study on the Physiological Responses of Tomato Related Species to Flooding Stress in Summer

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This research was to explore the effect of flooding stress on the growth and physiological status of *Solanum* species in summer. The antioxidative enzyme activities and antioxidant contents in roots of sixty-day plants of 'ASVEG #6' (*Solanum lycopersicum* L.), L4422 (*S. pimpinellifolium*), L3683 (*S. habrochaites*) and L1947 (*S. peruvianum*) were analyzed after flooding for 0, 12, 24, 48 and 120 hrs in summer. The results showed that chlorophyll fluorescence Fv/Fm and NPQ values reduced after flooding for 24 hrs. L1947 had the lowest biomass. At 48 hrs flooding, increases were observed on ascorbate peroxidase activity in L1947 and α -tocopherol content in L4422. Thirty-day seedlings of 9 lines of *S. peruvianum*, 6 lines of *S. chilense*, 7 lines of *S. pimpinellifolium*, 16 lines of *S. lycopersicum* L. var. *cerasiforme*, 16 lines of *S. habrochaites*, and 'ASVEG #6' as control were used as experimental materials. The growth characteristics, physiological status, antioxidative enzyme activities, and antioxidant contents of roots were analyzed after flooding for 48 hours in summer. *S. habrochaites* exhibited lower stomatal conductance than 'ASVEG #6'. After flooding, the stomatal conductance of *S. chilense* L6048, L6049, and *S. habrochaites* L6123 increased, while *S. lycopersicum* L. var. *cerasiforme* L0144 decreased. Total ascorbate content in *S. lycopersicum* L. var. *cerasiforme* L0172 also increased. Among 54 germplasms tested, total ascorbate content of *S. pimpinellifolium* L4422, ascorbate peroxidase activity of L0172, alcohol dehydrogenase activity of *S. lycopersicum* L. var. *cerasiforme* L0172 and *S. habrochaites* L6218 exhibited the highest level after flooding.

S12.077

Characteristics and Breeding of *Rubus* Genetic Resources that Are Underutilized in Japan

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The genus *Rubus* consists of more than 250 species that are widely distributed from tropical to subarctic regions. Raspberry (*R. idaeus* and *R. strigosus*) and blackberry (*R. ursinus* and hybrid) are cultivated as major horticultural crops in many parts of the world. More than 35 *Rubus* species are native to Japan. These and foreign cultivars were not utilized for breeding and cultivation until the 1990s. Disease and insect damage to crops is phenomenal due to the warm humid summers that facilitate proliferation of pests and diseases. Effective control

of these by the use of pesticides is not well developed as only a few pesticides have been approved by law for this minor crop, and their commercial production is not extensive. However, imports of raspberries and blackberries have increased in Japan over the last decade due to changing dietary habits. This study investigates the diversity in the native Japanese species, their importance, and potential as a genetic resource. Species such as *R. crataegifolius*, *R. ikenoensis*, *R. mesogaeus*, *R. palmatus*, *R. parvifolius*, *R. pectinellus*, *R. phoenicolasius*, and *R. vernus*, were collected as whole plants or seeds, mainly from the Tohoku district in the northeast of the mainland. The collection included seeds from Hokkaido, a region located in the northern island, collected in 2005 and 2009 during an expedition in collaboration with the National Clonal Germplasm Repository (NCGR), Corvallis, USA. In addition, cultivars were introduced from NCGR, Corvallis. The collected plants were conserved and evaluated for growth habit, genetic relationships, and fruit constituents. They were also used to develop a molecular marker. It is hoped that the native Japanese species will contribute to breeding and cultivation in the future by providing genetic resources.

S12.078

Diversity and Species Richness on Genus *Amelanchier* Medik. (*Rosaceae*) in Mexico

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The genus *Amelanchier* Medik. (*Rosaceae* subtribe *Pyrinae*) represented several fruit shrubs which are mainly distributed from Canada in North America to Honduras and Guatemala in Central America. Six species of this genus are reported in Mexico, among them stand out the three species belonging to section *Malacomeles*. However, but it is not known where most biodiversity of this genus is located in the country. Therefore, this biogeographic study was to determine areas of highest biodiversity based on species richness and diversity indices of the genus *Amelanchier* in Mexico. We found that most biodiversity is concentrated in the northeast region of Mexico in the biogeographic region known as Sierra Madre Oriental, where the highest diversity based on the Brillouin's diversity index and highest species richness based on the observed species richness and Chao's estimator of richness were located. There is also an area in the highlands of Chiapas, that presented a high diversity but low species richness, which could be interesting for the collection of germplasm of this genus. However, *A. denticulata* and *A. nervosa* were the two predominant species in Mexico and the most interesting from the point of view of collecting plant genetic resources.

S12.079

Analysis of Genetic Identity of Apples, Pears and European Plums Prospected at Valderejo Natural Park (Álava, Spain)

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Valderejo Natural Park, located in the Purón river valley at the occidental corner of Álava (Basque Country, Spain) is an isolated region that became almost uninhabited around 1970s due to rural exodus. Agriculture was practised there since ancient times, mostly grain crops, with a certain backyard horticultural production. Isolation and depopulation have allowed natural preservation in the valley and, in the last years, within the program of "Recovery and Conservation of the Fruit Genetic Heritage kept in Valderejo Natural Park", several surveys have allowed collecting apple, pear and European plum germplasm. The objective of the present study is to determine genetic identities and potential relationships of the apple, pear and European plum prospected material with both commercial standards and ancient cultivars kept in Germplasm Banks using SSR markers. Each accession was characterized with at least 8 highly polymorphic SSR markers that have been previously used to fingerprint the commercial standards and the

ancient cultivars. For each species, the commercial standard cultivars were chosen to represent a wide genetic pool, and the ancient cultivars are the collections curated in the Germplasm Banks at Universidad Pública de Navarra (UPNA, Pamplona), Estación Experimental de Aula Dei (EEAD, Zaragoza) and Universidad de Lleida (UdL, Lleida) in Spain. Genetic relationships among the genotypes were calculated using UPGMA cluster analysis of the similarity matrix obtained from the proportion of shared alleles. The genetic information was assessed using number of alleles per *locus*, observed and expected heterozygosity, and the probability of identity.

S12.080

Distribution, Habitat, and Characteristics of Wild *Lonicera caerulea* (Blue Honeysuckle) in Canada

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Over 800 accessions of *Lonicera caerulea* have been gathered in 250 locations from 9 provinces in Canada. It was found in all provinces West of the Rocky Mountains but absent in British Columbia. It was widespread in the Boreal and Acadian forests but only a few accessions were found in the Great Lakes/Saint Lawrence Forests. In the interior parts of Canada, *L. caerulea* was usually an understory plant on the edge of a forested area in or near wetlands high in organic matter from either peat soils or decaying trees. In those regions, Black Spruce (*Picea mariana*), Alders (*Alnus* sp.), and Willows (*Salix* sp.) were useful as indicator plants of habitats worthwhile to search for *L. caerulea*. In maritime eastern Canada *L. caerulea* was often found with peat soils in open areas with rock outcroppings even on hillsides or in low lying boggy areas, sometimes only meters away from salt water. In all locations where it was found, there was a high diversity of plant species. It was never found to be a dominant species and was often quite rare in any given location, which indicates this species is highly unlikely to become invasive. Perhaps 700 'likely' locations were searched where it was not found. Diversity in plant habit, height, leaf colour and resistance to leaf diseases was noted. Fruits tended to taste good, were very small and either round or pear shaped but only about 40 accessions had fruits at the time of collection. Accessions are currently field planted at the University of Saskatchewan.

S12.081

Morphological Characterization of Some Sweet, Sour and Duke Cherry Genotypes

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Sweet cherry, *Prunus avium* L., originated from around the Caspian and Black Seas is one of the important fruits in the world. Sour cherry, *Prunus cerasus* L., is also originated from very similar place to that of sweet cherry and is a natural hybrid between sweet cherry and ground cherry (*Prunus fruticosa*). The hybrid between sweet and sour cherry called duke cherry (*P. ×gondouinii*) is cultivated on a much smaller scale in the world. Duke cherry trees are intermediate for their tree and fruit characteristics compared to their progenitors. For determining similarities and differences between these species, 50 genotypes (sweet, sour and duke cherries) were studied based on morphological characters. These characters included 29 traits of fruit and 2 traits of leaf that were measured over two years and analyzed by SPSS software, and the Ward method used for cluster analysis. Results showed that most of sweet cherry traits, such as fruit weight, TSS and flesh firmness, had significantly higher values than the other two species. Fruit doubled and fruit cracking were observed only in sweet cherries. Duke cherries had higher similarity to sour cherries than sweet cherries. In the dendrogram, genotypes of each species were placed in the separate groups, but some sour cherry genotypes were clustered with duke cherries. This survey revealed distinct characters of sweet cherries compared to the other two species.

S12.082

Lingonberry Germplasm Characterization for Berry Characters, Anthocyanin Contents and Antioxidant Activities

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Lingonberry (*Vaccinium vitis-idaea* L.), an important fruit crop in many northern latitude countries, is a medicinal plant rich in anthocyanin antioxidants. Two subspecies of *V. vitis-idaea* have been recognized, the larger lowland race as *V. vitis-idaea* ssp. *vitis-idaea* (L.) Britton and the dwarf arctic-montane race as *V. vitis-idaea* ssp. *minus* (Lodd) Hult. Fruits from a lingonberry germplasm containing different hybrids and cultivars of *V. vitis-idaea* ssp. *vitis-idaea* and *V. vitis-idaea* ssp. *minus* wild clones, were evaluated for fruit characteristics, anthocyanin contents and antioxidant activities. Wide genotypic variation for all characters was observed within and between sub-species with *V. vitis-idaea* ssp. *minus* being superior to *V. vitis-idaea* ssp. *vitis-idaea* for anthocyanin content and antioxidant activity. The hybrids were better than those of their respective parents for anthocyanin content but not for antioxidant activity. No correlation was found between the anthocyanin content and the antioxidant activity. The fruits at developmentally early stage had increased antioxidant activities but decreased anthocyanin contents.

S12.083

Heterosis and Combining Ability for Earliness Yield and Fruit Quality of Some Egyptian Melon Inbred Lines Via Line x Tester Analysis

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Six sweetmelon (*Cucumis melo* var. *aegyptiacs*) inbred lines "Ana-3, Esm-4, War-4, Fal-5, Mas-4 and Kha-4" were utilized into line x testers top crosses with three netted melon (*Cucumis melo* var. *reticulatus*) inbred lines "Kur-2, Gw-4 and Hira-2" during fall season "August-November" 2008. The eighteen nested genotypes resulted from top crosses, were evaluated in comparison with their respective parents, during 2009 hot summer season "June-September", in new reclaimed sandy soil in open field. Plant growth performance at flowering stage, number of days from sowing to flowering, early, total yield and fruit quality characters were determined. Also field tolerant for drought stress and gummy stem blight during fruit maturity period were recorded. Mean squares due to crosses and their partitioning into lines and testers, were investigated for all studied traits. Also mean squares due to lines x tester's interaction were determined. Additive and non-additive genetic variance, general and specific combining ability for all studied traits were investigated according to line X tester analysis methods. Moreover heterosis over mid and best parents was calculated for all studied traits.

S12.084

Comparative Analysis of the Genetic Diversity Maintained in Apple Germplasm Banks from Northeastern Spain

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Nowadays, Spanish (and European) apple production relies on a reduced number of cultivars, mainly on 'Golden', 'Gala', 'Red Delicious' or 'Fuji' groups and on few others mostly derived from crosses that include also these few progenitors. Since the 1960s, these varieties have drastically replaced the pool of ancient traditional

and well-adapted cultivars on which apple production was based earlier. As a consequence, many of the traditional or local varieties have been considered as “obsolete”, leading to a dramatic loss of genetic diversity. Acknowledgement of the need for collection and preservation of endangered apple germplasm encouraged the establishment of programs for conservation of these genetic resources that could constitute sources of diversity and useful agronomic traits, such as pest resistances or growing habits, in present and future breeding programs. Nowadays, three apple collections are curated in Northeastern Spain, namely in the Germplasm Banks at Universidad Pública de Navarra (UPNA, Pamplona), Estación Experimental de Aula Dei (EEAD, Zaragoza) and Universidad de Lleida (UdL, Lleida) totaling, respectively, 282, 65 and 104 accessions. A previous joint research fingerprinted the three collections with a common set of 10 SSR markers. The objective of this study is to assess the variability and diversity contained on each Bank and the degree of overlap among them. Diversity assessment was performed using the following indexes: number of alleles (total and effective) per *locus*, allele frequencies, observed and expected heterozygosity, probability of identity, Nei (DI) and Shannon-Weaver (H') diversity indexes. Genetic relationships among the genotypes were calculated using UPGMA cluster analysis of the similarity matrix obtained from the proportion of shared alleles. The differentiation between local populations of different geographical origin was calculated through the analysis of the molecular variance (AMOVA) and pairwise F_{st} values.

S12.085

Evaluation of Polyphenols Content and Antioxidant Activity of Six Different Cultivars of Pomegranates Peels, a By-Product of the Commercial Juice Industry

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In recent years, pomegranate fruit (*Punica granatum* L.) has taken great attention for its health benefits. Fruits are widely consumed fresh and in processed forms as juice, jams and etc. Pomegranate peel, a by-product of the commercial juice industry, is a rich source of polyphenol compounds. The objective of this research was to investigate and compare polyphenol compounds and antioxidant activity of six cultivars pomegranate fruit peels grown in Iran. In the present study, we evaluated total phenolics, total tannins, condensed tannins and antioxidant activity in ‘Aghaye’, ‘Faroghi’, ‘Rabbab-e-Fars’, ‘Shahvar’, ‘Shirin-e-Bihaste’ and ‘Shirin-e-Mohali’. The results showed that the significant differences ($p < 0.05$) were detected in all measured traits of pomegranate cultivars. The total phenolic content (mg tannic acid/g dry matter) varied from 380.06 to 441.18 in ‘Rabbab-e-Fars’ and ‘Shirin-e-Bihaste’, respectively. The values of total tannins (mg tannic acid/g dry matter) ranged from 170.09 (‘Aghaye’) to 220.98 (‘Shirin-e-Bihaste’). The concentration of condensed tannins (mg catechin/g dry matter) was varied from 2.94 (‘Shahvar’) to 4.03 (‘Rabbab-e-Fars’). The highest antioxidant activity was observed in ‘Shirin-e-Bihaste’ (84.50%), while the lowest value was found in ‘Rabbab-e-Fars’ (79.78%). The antioxidant activity was positively correlated with total phenolics ($r = 0.938$), total tannins ($r = 0.924$) and condensed tannins ($r = 0.773$). We concluded that the large amount of phenolics and tannins in pomegranate peels provides a good potential as a health supplement rich in natural antioxidants and merits further intensive study.

S12.086

The Use of Microsatellites to Analyze the Relationships and to Decipher Homonyms and Synonyms in Azorean Apple (*Malus domestica* Borkh.) Cultivars

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The Azores islands are rich in biodiversity. The climate is changeable, very windy and humid. Besides the high level of humidity *Malus domestica* is widespread all over the islands and well adapted. Most of the apples are represented by native populations. Generally they were called by local name and sometimes the same name could be attributed to different cultivars and the same morphological aspect could receive different denominations. Moreover, the absence of well-defined descriptions makes the morphological and genetic analysis extremely difficult. The aim of this study was to exploit the molecular and morphological variability present in *Malus domestica* in order to clarify the confusion characterizing the apple plantations in Azores. The samples were analyzed at ten SSR *loci*. The total number of alleles per *locus* and the percentage of heterozygotes found were high, reflecting the high variability present inside the pool of our samples. The high variability was supported also by the high value of the discriminating power. This means that using those primer combinations it was possible to discriminate most of the tested apple cultivars by their allelic profiles. The rooted tree organized most of the samples into fourteen main clusters, often with high bootstrap values. It was possible to individualize 60 synonyms and 32 homonyms. Moreover, it was possible to relate each individual to its original population and detect possible parent-offspring groups.

S12.087

Do We Preserve Unique Apple Germplasm?

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In many European countries, preservation of genetic resources in apple is directed mainly, or even exclusively, to so-called heirloom varieties, i.e. old and presumably indigenous cultivars. However, there is very little information about the extent to which such collections contain a unique and desirable set of genes as opposed to just a random sample of genes from the global population of apple germplasm. Molecular markers, especially SSR (Simple Sequence Repeats), are commonly used for quantification of the amount of diversity in national collections of various plant crops. Provided that the same SSR primers are used and that allele sizing is standardized, data obtained in different studies can be pooled, thus allowing us to estimate not only diversity but also the degree of differentiation among e.g. apple collections preserved in different national gene banks. We have now used primer pairs from a ‘standard set’, defined by the EC-PGR (European Community for Plant Genetic Resources) working group on molecular markers, to analyse a total of approximately 150 apple cultivars native to Sweden, Finland and Ireland, as well as some other European and non-European cultivars. Preliminary SSR data indicate that the Swedish and Finnish heirloom cultivars are very diverse, and are dispersed rather evenly among European and non-European varieties. By contrast, Irish heirloom cultivars appear to be more differentiated. Possibly the relatively short time of apple growing in Sweden and Finland is the main reason that heirloom cultivars originating in these areas show no evidence of above-average genetic similarity in spite of the rather extreme growing conditions. In Ireland, apple growing apparently has a longer history and thus several more generations of selection for, e.g., climatic adaptation may have taken place.

S12.088

Molecular Assessments of Genetic Diversity in Berry Crop Germplasm

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Although morphological characters have commonly been used to measure genetic diversity in many crop plants, molecular methods provide an additional tool for identifying germplasm and for assessing genetic diversity/relatedness in plant genotypes for practical breeding purposes and proprietary-rights protection. The introduction of molecular biology techniques, such as DNA-based markers, allows direct

comparison of different genetic material independent of environmental influences. This review presents the progress in-depth of various aspects of molecular diversity analyses in berry crop improvement program of Atlantic Cool Climate Crop Research Centre of Agriculture and Agri-Food Canada, St. John's, NL, Canada. Significant progress has been made in diversity analysis of wild cranberry, lowbush blueberry, lingonberry, and cloudberry germplasm, and in strawberry and raspberry cultivars and advanced lines developed in Canada. Inter simple sequence repeat (ISSR) markers detected a sufficient degree of polymorphism to differentiate among berry genotypes, making this technology valuable for cultivar identification and for the more efficient choice of parents in the current berry breeding programs. The paper also discusses the issues that still need to be addressed to utilize the full potential of molecular techniques to develop improved berry cultivars suited to the changing needs of growers and consumers. The combination of morphological and molecular approaches will produce a better description of the variation in berry germplasm collections and may reveal relationships between the markers and morphological traits.

S12.089

National Asian Pear (*Pyrus serotina* Rehd) Project in Iran, Phase 2: Cultivars Compatibility Evaluation under the Different Geographical and Environmental Conditions

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Asian pear cultivars are a new fruit in Iran. In order to evaluate some Asian (Japanese) pear (*Pyrus serotina* Rehd) cultivar under Iran environmental conditions, the national research project was established at the Department of Horticultural Science, Tarbiat Modares University (TMU), Tehran, Iran. In the project three phases was suggested and approved including Phase 1: Germplasm introduction, quarantine and propagation (Grant No. NRCI 4225); Phase 2: Cultivars evaluation for compatibility under different environmental conditions (Grant No. INSF ORG 84006) and Phase 3: Promotion and extension of specific suitable cultivars for new planting regions. Phase 1 of the National Asian pear project started in 1997 at TMU with successful introduction of nine Asian pear cultivars named 'KS'6, 'KS'7, 'KS'8, 'KS'9, 'KS'10, 'KS'11, 'KS'12, 'KS'13 and 'KS'14 to Iran. Imported cultivars propagated, checked for quarantine and added to the new established Asian pear collection orchard at TMU during 1997 to 2003. Phase 2 of the project has been started with planting of cultivars during 2004 to 2010 under different geographic and environmental conditions of Iran. The new planting sites are Tehran, Karaj, Nazarabad, Shahriar, Damavand, Lavasanat, Hashtgerd, Taleghan, Kordan, Mohamad-shahr (Tehran province); Shiraz, Norabad Mamasani, Abarkooh and Abadeh (Fars province); Ghazvin and Zanjan province; Rasht, Langarood and Sary (North of Iran); Mashhad and Sabzevar (Khorasan province); Isfahan, Kermanshah, Ormieh and Tabriz. During phase 2 of the project, Asian pear cultivars were evaluated for various compatibility criteria to the different environmental conditions such as vegetative, reproductive and pre and postharvest fruit characteristics. Currently, data obtained from this part of the project is under evaluation and analyze in order to select suitable cultivars for specific geographical and environmental conditions. The progress in the national Asian pear project in Iran and the future cultural and breeding program is discussed.

S12.090

The Management of the Apricot's Genetic Resources in Romania within the Request of a Healthy Nourishment

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The management of the Apricot's genetic resources in Romania has been subjected to the request of a healthy nourishment and has been focused on: 1. The preservation of a rich genetic fund (663 phenotypes from 7 geographical areas); 2. The

introduction of a large genetic variability by means of conventional and unconventional methods; 3. The setting up of genetic improving strategies based on genetic resources; 4. The selection and use of phenotypes belonging to different geographical areas as genetic apricot resources inducing the following: viruses tolerance, resistance to the apricot diseases (*Monilinia laxa* (Aderh et Ruhl) Honey, *Stigmia carpofilla* (Lev.) M. B. Ellis, *Cytospora cincta* Sacc.), resistance to the climate ups and downs, a higher agricultural productivity, an earlier and later ripping season, a better quality of fruit due to the higher content in glucides, in Mg, Fe, P, Ca, K, S, Na, Mn, Fl, Co, Br, in vitamins A, P and C; 5. The creation of a large inter and intra-specific apricot genetic fund; 6. The selection and validation of several new apricot phenotypes (25 varieties) with features identical and similar to the apricot ideotype.

S12.091

Evaluation, Utilisation and Conservation of Mango (*Mangifera indica* L.) Gene Pool of West Bengal, India

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The favourable soil and climatic conditions of West Bengal state of India are ideal for growing a variety of tropical and subtropical fruit crops. Mango is the major fruit of West Bengal covering an area of 80.90 thousand hectares which is about 41 per cent of total fruit cultivation of the state. The history of mango cultivation in the state is long and steeped in antiquity. The existence of relatively large number of varieties in West Bengal indicates the patronage extended to the mango by the 'Moughals' and 'British' rulers. Considering the imperative need to collect the existing gene pool of mango in the state, we carried out surveys for collection, evaluation, conservation and utilization of the gene pool. Over the last 16 years, we could identify 94 varieties growing in different districts of the state. The varieties were evaluated, vegetatively propagated and planted in the Research Station. The utilization of the gene pool has been suggested. Wide variations in fruit weight (70.0g in variety 'Subza' to 768.8g in variety 'Totapuri'), size (10.3 cm to 17.1 cm in length), pulp recovery (62.4 % to 83.7 %), bearing habit (regular/ alternate), maturity (early, medium or late) pests, diseases and physiological disorders status (susceptible/resistant) and keeping quality were evaluated. Based on different characters, the varieties were classified for (a) regular/ irregular in bearing (b) dwarf/ medium/ tall in tree size (c) early/ medium/ late in maturity (d) suitable for table fruit or for processing (e) varieties suitable for export, and (f) varieties that needs urgent attention before extinction.

S12.092

Minor Traditional Grapevine Varieties Exclusively Located in Zones of Wine D.O.P. in the West of Castilla y Leon (Spain)

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Over the last years the interest to identify and preserve grapevine minor varieties has increased a lot. Many of these varieties can get disappearance due to the renovation of vineyards with more spread varieties. Castilla y Leon, the most extensive region of Europe, has numerous and very different wine production zones, in which a high number of minor varieties exists, with very little vines and high danger of extinction. These varieties are distributed in a disperse way in vineyards of reduced size that are difficult to locate and identify. A meticulous and large prospection (962 vines, 176 vineyard plots) has been carried out throughout the last years (2003-2009) in the wine producing zones located in Leon, Salamanca and Zamora provinces (West of Castilla y Leon), getting up to 125 accession names, some of whose vines are prephyllloxeric, as it was indicated by Garcia de los Salmones in 1912. A reduced number of them corresponds to previously nondescribed varieties (exclusive genotype), according to the analysis by means of microsatellites

(descriptors 801 to 806 of O.I.V. codes). The ampelographic description of each variety was realised during four years according to the descriptors indicated by the O.I.V. The results show a set of varieties of exclusive genotype that are in a high danger of extinction, as for example Rio Abaixo, Tinto Jeromo, Verdejo Colorado and Verdejo Serrano. It would be necessary that these varieties, once located and characterized, are preserved in germplasm banks, to avoid the loss of this vegetal material and its corresponding genetic variability, at the same time that can be considered to evaluate their agronomic and enologic potential.

S12.093

Studies on the Mutation Breeding for Low-Seeded Nianju Tangerine Mutants

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Citrus seedless character is one of the main targets of citrus breeding and the important quality indexes of commercial citrus fruit. Nianju (*C. reticulata* Blanco) is a main cultivar in Guangdong Province which is of early bearing, high productivity, late-season and good quality characters, but has too many seeds. The objective of this study was to induce low-seeded Nianju cultivar. Mature autumn shoots were taken from the adult seedy Nianju trees and the leaves of the shoots were stripped. The shoots were irradiated with gamma rays from ⁶⁰Co at the doses of 0, 60 and 80 GY and dose rate of 0.23 GY/min in January of 2005. All the treated buds were single-bud-grafted on Hong Ning Meng (*C. limonia* Osbeck) rootstocks after irradiation. The survivor rates were 96.5%, 39.3% and 16.5% for the 0, 6 and 8 GY treatment respectively. The plantlets were grown in the orchard in May of 2006. Among them, 81 plants produced fruit in 2008. Six low-seeded mutants (2-1, 3-11, 4-10, 6-6, 6-11, 2-3) were screened, and the rate of low-seeded mutants was 7.4%. The average seed content of the mutants was 3-6 seeds/fruit, differing from the control plants which had 18.7 seeds/fruit. The average fruit weight was 41-60g (CK was 56g/fruit). The yields of mutants 3-11 and 4-10 were 7.1 kg and 8.3 kg respectively and that of the control was 8.5 kg. The superior characters of seedy Nianju were kept in the low-seeded mutants. One of the main causes producing low-seeded fruit was much higher rate of abortive pollen grains than that of the control.

S12.094

Evaluation of Some Lime and Lemon Accessions by Using Morphological Characterization in Hormozgan Province

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Citrus fruits are the most important subtropical crops in the world. In Iran, citrus fruits are cultivated in the North and South of the country. Limes and Lemons are major citrus crop in Southern Iran. There are high demands for consumption fresh citrus fruits, and also extracted juice and dried fruit. In this study, some physical and chemical characteristics of 18 genotypes of Lime and Lemon were investigated. The characteristics included fruit length and diameter, leaf length and width, and percentage of fruit juice and peel. Some characters of fruits juice such as pH, titration acidity and TSS and antioxidant activity were also determinate. According to the results of bivariate simple correlation analysis, there are significant positive and negative correlations between some important characters. Factor analysis showed that fruit weight, fruit length and fruit juice, leave and seed characters composed the main factor. The most effective characters were categorized into 9 main factors (with an Eigen value \geq 1) that contributed to 92.33% of total variances. Cluster analysis was performed by using these 9 factors and genotypes were divided at distance of 20 out 25 into 7 main clusters that included lime and lemon and unknown genotypes. The simple plot analyses with 2 main factors have divided all genotypes to four groups such as limes, lemons and two groups of unknown genotypes. Some genotypes of limes were different in some characters such as fruits shape and size, and peel thickness. Lemon groups contain 'Oreka', 'Lisbon' and 'Mayer' cultivars. Other group contains one genotype that was separated from other genotypes in

distance of 25 in cluster analysis and was separated from other genotypes in simple plot analysis too. It has large fruit similar to orange but very acidity likes lemons. Some unknown acid citrus were different from either limes or lemons.

S12.095

The Most Ancient Olives Trees of Andalusia (Spain), an *in situ* Germplasm Collection

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Olive growing is in time of change, many cultivars in traditional groves are being replaced by few cultivars in new orchards. This scenario may lead to genetic erosion. However, the value of these ancient trees and wild olive populations as genetic resource is still unexplored. A survey looking for the ancient olive trees of Andalusia (South of Spain) was made and 35 olive trees, 56 olive groves, 18 wild olives and 5 wild olive forest were sampled. A set of 14 SSR markers was used to identify and study the genetic variability and the genetic relationships among 160 of these monumental trees (29 wild and 131 cultivated). Only seventeen percent of the cultivated genotypes were identified as known current cultivars. Thirty-two percent of monumental trees were grafted and wild olives were in most cases used as rootstock. The phylogenetic analysis showed two major groups according to their status: cultivated or wild. The cultivated genotypes were associated between themselves according to their geographical origin. This correlation supports the hypothesis of local selection and domestication of olive cultivars. The information obtained in this work points out the value of the ancient and wild olives for tracing olive domestication history. Their high genetic variability and tolerance to biotic and abiotic stresses over the time could be also valuable in olive breeding programs. In this context, we propose *ex-situ* and *in-situ* conservation programs for preserving the genetic variability present in ancient and wild olives.

S12.096

Conservation of Pacific Plantain Bananas in French Polynesia

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There are 4 types of cooking bananas in French Polynesia. Three of them are Eumusa AAB type (Maoli, Popoulu, Iholena) and one is Autralimusa (Fei banana). These bananas were introduced in French Polynesia by the very first inhabitants (1600 BC). Cooking bananas were one of the staple food in the past centuries. Fei bananas are still appreciated in special dishes. The diversity of these bananas was enhanced by the climatic conditions within FP islands and the farmers' selection. In order to preserve the diversity of these unique bananas, FP started a conservation program in 2007. Accessions were collected in the islands of Tahiti, Raiatea, Tubuai, Rapa and the Marquesas islands. Two field collections are now in place in the Papara research station hold by the Ministry of Agriculture. The Maoli, Popoulu, Iholena collection has 108 accessions. The Fei collection has 80 accessions. The morphologic characterization of the accessions is ongoing. Virus indexing and tissue culture are the two main objectives in 2010 in order to secure the two collections. The tissue culture collection will be duplicate in the Center for Pacific Crops and Trees (CePaCT), facility of the South Pacific Community.

S12.097

A Plant Type of Jackfruit (*Artocarpus heterophyllus* Lam.) Bearing Fruits throughout the Year

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Jackfruit plays a significant role in Indian agriculture and culture. It was cul-

tivated in India 3000 to 6000 years ago. It is a popular and cheap fruit in Southern Asia and other warm countries of both the hemispheres. In Europe the fruit is sold in canned with sugar syrup. The tender fruits of the tree are used as vegetables and the ripe ones as table fruits. The traditional varieties bear fruits once in a year. Usually the flowering starts from mid November and extend till mid February, depending on the location and the variety. The tender fruits come to market from March onwards and continue till August. The fruits begin to ripen in the month of June. However the late varieties may ripen in October. A matured tree may bear more than 250 fruits depending on the size and the variety. On an average 50-80 tonnes of fruits can be harvested from a hectare of land. This tree produces the largest single fruit nearly 26 inches long, 40 inches in diameter and weighing up to 40 Kg. Fresh and tender fruits are usually not available from November onwards. However three trees were identified; in the *terai* region of sub Himalayan foot hills of Cooch Behar and Jalpaiguri districts of West Bengal, India, which bear fruits throughout the year. These trees were studied for three years from June 2006 to May 2009. Though it is reported that Jackfruit is a monoecious and highly cross pollinated crop, it was found that self pollination also took place, with no inbreeding depression for fruit size, to the extent of 84.57 percent, during the off season, i.e. in the months of April to early February. Percentage of fertile pollen grains was as high as 89.00 percent. Proper management of the crop i.e. application of 50.0 Kg of F.Y.M., 5.0 Kg of wood ash, 2.0 Kg of bone meal, and 50.0 gms of borax per matured tree (10 years and above), was found to increase the fruit yield per tree, substantially. On an average, 26 to 31 fruits were harvested per month per tree in the months of April to November. However it was only 16 to 21 fruits during the months of December to February. Thus a farmer could earn more than Rs. 2,500.00 per tree per year. The average weight of fruits varied from 3.2 kg to 4.8 kg., during the entire year. The following fertilizer-cum- manure dose was practiced, (F. Y. M. @ 50.0 Kg, wood-ash @ 5.0 Kg, bone-meal @ 2.0 Kg, N.P. K. @ 2.0 Kg and borax @ 50.0 gms) per matured tree. The manures and the fertilizers were well mixed before application to the soil. The mixture was applied in two split doses, first in the first fortnight of May, i. e. just after the onset of monsoon and second, in the first fortnight of October i. e. prior to the end of monsoon. Provision of irrigation was suggested during the dry months of the year (November to April). Air-layering was successful (21.67 per cent) when tried during the first fortnight of July. An application of 1000 ppm of I B. A. was found effective. The air layers were detached after proper rooting which was about 60 days of air-layering. Photographs depicting different stages of fruits in different months of the year have been presented for a clear understanding of fruit bearing through out the year.

S12.098

Genetic Resources of Pummelo (*Citrus grandis*) in West Bengal, India

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Survey for selection of elite clones of pummelo was initiated in 1999. Over the years more than 700 trees were evaluated and fifteen elite clones were identified as superior clones. Wide variability in yield (64-215 fruits/tree), fruit weight (625-2010 g), fruit shape, peel colour, number of segments (11-19/fruit), number of seeds (67-182/fruit), total soluble solids (5.0-9.6%), total sugar (3.6-7.6%), and fruit acidity ((0.12-1.10%) were observed among the genotypes. Genetic variability studies for fruit characters of fifteen selected genotypes reveals that variability was wide for fruit weight, peel weight, peel thickness, flesh colour, number of segments, number of seeds, segment weight and fruit acidity. High heritability estimates coupled with high genetic advance was estimated for TSS: acid ratio of fruit, peel weight, seed number per fruit, seed weight, fruit acidity as well as fruit weight and segment weight, suggesting that further selection for improvement in fruit weight, segment weight, acidity and TSS:acid ratio would be effective.

S12.099

Genetic Resources of Chinese Jujube (*Ziziphus jujuba* Mill.)

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Chinese jujube (*Ziziphus jujuba* Mill.), a native fruit tree of China, is becoming increasingly important for its extreme drought hardness and barren soil tolerance, nutritious and multi-use fruit as well as early bearing, high repaying and easy management. It is derived from the wild species *Z. acidojujuba* C. Y. Cheng et M. J. Liu and first cultivated in the middle and lower reaches of Yellow River 7000 years ago. Now, it is planted throughout China (23-44°N and 76-124°E) and concentrated in the northern dry lands with a total growing area of 1,500, 000 hectares and production of over 3,000,000 tones. It has been introduced into more than 40 countries all over the world and has reached commercial production in South Korea. Some 940 local cultivars/types and newly bred cultivars/strains have been reported, of which around 560 have been collected and well conserved in the National Chinese Jujube Germplasm Repository, Shanxi Pomology Institute. Considerable information on Chinese jujube germplasm has been generated which showed very high genetic diversity in terms of the adaptability to soil and climate, morphological and phenological traits, chromosome number and karyotypes, pollen morphology, isozyme, fruiting ability, seed fertility, functional component content, tolerance or resistance to biotic and abiotic stresses and genomic DNA. Besides, promising germplasms with distinctive traits such as high and stable yield ('Linyilizao'), male sterile ('Dongzao', JMS1, JMS2, JMS3), high embryo fertility ('Xuechengdongzao'), high Vc content ('Bingbingguan'), high resistance to the three destructive diseases-jujube witches' broom, fruit shrinking and fruit cracking ('Xingguang', 'Gikang 1', 'Yushuai', 'Yingyu'), triploid ('Zanhuangdazao') and tetraploid ('Chengguang'), high tolerance to drought ('Tongxinyuanzao'), high tolerance to cold and riping early ('Yueguang') have been identified. A series of innovative cultivars and strains with excellent traits from different routes have been released recently. The future research works are discussed.

S12.100

Comparison of Characteristics of Flower and Pollen between Tetraploid and Diploid Chinese Jujube (*Ziziphus jujuba* Mill.)

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Polyploid breeding is becoming more and more popular because of the higher resistance to biotic and abiotic stresses of the polyploidy cultivars. Recently, a great progress has been made on polyploid breeding in Chinese jujube (*Ziziphus jujuba* Mill.) which has encountered a lot of obstacles in cross breeding such as embryo abortion and small flower. Through induction of apical buds with colchicine of three diploidy cultivars named 'Linyilizao', 'Lajiaoza' and 'Dongzao' respectively, and by ways of pruning and top-grafting for several years, new germplasms with doubled chromosome number were acquired. The flower and pollen characteristics were studied in order to discover the differences between diploidy and tetraploidy germplasms. Compared with diploidy 'Lajiaoza' and 'Dongzao', the flower numbers per bearing shoot of their tetraploids decreased significantly by 49.2% and 52.1%, respectively. However, flower number per bearing shoot showed no significant difference between diploidy and tetraploidy 'Linyilizao'. The size of flower bud increased in tetraploidy 'Lajiaoza' and tetraploidy 'Linyilizao' while pollen number decreased in tetraploidy 'Lajiaoza'. The pollen vitality of the tetraploids decreased by 12.5%, 16.3% and showed no significant difference with their diploids. Pollens with four apertures only occurred in tetraploidy 'Lajiaoza', 'Dongzao' and 'Linyilizao' with percentage of 20.7%, 15.2% and 11.2% and the size of 451.2±24.7µm, 475.2±31.8µm and 438.0±46.6µm, respectively, while the diploids had much smaller pollens with three apertures.

S12.101

Identification of a New White-Berried Grapevine Cultivar as a Result from Bud Sport of the Portuguese Blue Black Cultivar Alfrocheiro Preto

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Bud sports are characterized by infrequent changes in phenotype affecting only some shoots of plants. However, the molecular basis behind the generation of such sports is not well understood. Among the bud sports, colour mutants with alterations in the red purple anthocyanin content of flowers or fruit are mostly common and several examples in horticultural plants are widespread. Bud sports have been maintained the specific traits through consecutive vegetative propagations by means of clonal techniques leading to a new variety. Since the past decades, the occurrence of mutation affecting berry colour in various *Vitis vinifera* cultivars has been reported. In the Portuguese National Ampelographic Collection (CAN) at L-INIA Dois Portos, an uncharacterized cultivar is found demonstrating bud sports at whole plant level by presenting completely white-skin bunches or, in a same plant, branches giving rise only white-skin berries and branches which present, in a same bunch, three different coloured berries (completely blue black or white berries and berries showing longitudinally white or blue black sectors). In order to identify such cultivar, genomic DNA was extracted from leaves and berries of bud sport plants and then analyzed at the following nuclear SSR (Simple Sequence Repeat) *loci*: VVS2, VVMD5, VVMD7, VVMD25, VVMD27, VVMD28, VVMD32, VrZAG62, and VrZAG79. The SSR analysis using the nine SSRs revealed that the fragments amplified (from all plants presenting bud sports) coincided in size with those from the Portuguese blue black-berried cultivar 'Alfrocheiro Preto'. The cultivar identification was further confirmed as for leaf morphological traits through the ampelometric leaf descriptors scores (OIV 2007). Therefore, it is possible to assume that bud sport white-berried is a new variety arose through a somatic mutation of 'Alfrocheiro Preto'. All these bud sports reported here are been used for further researches.

S12.102

Detection of Synonymies in Grapevine (*Vitis vinifera* L.) Varieties in North Portugal and Northwest Spain

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During the 2002 to 2009 period numerous grapevine samplings have been carried out in several zones of North Portugal, between the Douro river and the Spanish border as well as in northwest Spain, namely Galicia and the provinces of León, Zamora and Salamanca, trying to identify the sampled material and detect the existing synonymies. The total number of studied accessions was around 700, including a certain number of them that are in a marked danger of extinction. The varietal identification was made based on the characterization with the microsatellite molecular markers included in the OIV 801 to 806 descriptors, that correspond to the following *loci*: SSR VVS2, SSR VVMD5, SSR VVMD7, SSR VVMD27, SSR VrZAG62 and SSR VrZAG79, completed with the OIV descriptors referred to adult leaf, bunch and berry that are included in the second edition of OIV descriptors. A total of 26 varieties of *Vitis vinifera* were identified with different names in Spain and in Portugal; 13 white and 13 black grapes, detecting in all cases the corresponding synonymies in both countries. Likewise several cases of homonymies, that may cause errors of identification were detected. Several cases of synonymies mentioned in the bibliography that have not been confirmed or have been detected as erroneous are discussed. The most adequate names for denomination each studied variety are suggested in order to achieve a precise and accurate way of identification.

S12.103

Analysis of Genetic Diversity in Litchi and Longan Germplasm by SRAP Markers

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SRAP were used to analyze the genetic diversity of 32 accessions of litchi and longan germplasm from Guangdong, Guangxi, Fujian and Hainan province of China, which contained 28 litchi (*Litchi chinensis* Sonn), 2 artificial intergeneric hybrids (litchixlongan), 1 longli (*Dimocarpus confinis* How et Ho) and 1 longan cultivar (*Dimocarpus longana* Lour. 'Shixia'). Nine out of the 60 screened primer pairs produced efficient amplifications, which generated 339 *loci* with a polymorphic rate of 99.7% and average 37.7 *loci* per primer pair. The cluster analysis by NTSYS-2.10 software indicated that the similarity coefficients ranged from 0.481 to 0.993, suggesting a rather high genetic diversity among the tested accessions. Thereinto, the similarity coefficients of litchi germplasm ranged from 0.536 to 0.993, indicating a relatively high genetic diversity among most of litchi germplasm, while the other few displayed close relationships. According to the dendrogram, the 32 tested germplasm could be classified into five groups at 0.72 similarity coefficient. The first group involved 11 litchi cultivars/lines with early season and early-to-mid season, including 'YA1', 'YN5', 'Sanyuehong', 'Feizixiao', etc. The second group involved 16 litchi cultivars/line with late season and extremely late season, including 'Guiwei', 'Wuheli', 'Maguili', etc. The third group included two artificial intergeneric hybrids and their maternal 'Ziniangxi'. The fourth group only contained 'Longli', a tentative natural hybrid of longan and litchi genus. The fifth group was 'Shixia' Longan. This classification was consistent with fruit maturation period. In addition, the pedigree of some recent released new variety was uncovered in the present study.

S12.104

Contrasting Cyto-Nuclear Lineages in Southwest Mediterranean Olive Varieties Indicating a Diversification through Sexual Reproduction in Traditional Agro-Ecosystems

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In the West Mediterranean areas, cultivated olive seems to be originated from local population as attested by two specific maternal lineages but mainly from olive trees introduced from East Mediterranean and probably introgressed by local gene pool. To test this hypothesis, we analysed the genetic structure of 476 Mediterranean olive varieties using 12 nuclear SSR *loci* and chloroplast DNA polymorphism. We confirmed the East-West genetic structure and showed that western gene pool is mainly characterised by eastern maternal lineage. Our study suggests that cultivated olive results from the incorporation of hybrids between introduced varieties from East Mediterranean and local varieties. Hence, olive diversification seems to be a dynamic process mainly arose through sexual reproduction in traditional agro-ecosystems.

S12.105

A Contribution to Characterisation of Elite Cultivars of Winter Camellias as Revealed by Sequence Tagged Microsatellite Site (STMS) and Morphological Markers

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Genetic resources can be described as the total genetic diversity of cultivated species and their wild relatives. Their effective conservation and use depend on the availability of information and on the extent and distribution of genetic diversity

in species of interest. In recent years, numerous molecular techniques have been developed to provide markers and now are by far the most powerful and widely used. Among the several classes of DNA-based markers, the Sequence Tagged Microsatellite Sites (STMS) are highly polymorphic, multi-allelic, frequently co-dominant, highly reproducible, selective neutrality and random widely distributed in the genome. In order to improve the genetic knowledge in ornamentals and to solve taxonomical issues, STMSs were used to investigate *Camellia* genus. Overall, 44 cultivars belonging to *C. sasanqua* L., *C. xhiemalis* Nakai, *C. xvernalis* Makino and *C. hybrida* were characterised by means of 4 STMSs and 16 morphological traits referring to flower and leaf morphology, colour, and fragrance. STMS markers successfully amplified all the accessions and 58 alleles were scored. The distribution of the genetic variation, attributed by AMOVA, highlighted genetic overlapping among *C. sasanqua* cultivars and the hybrids of *C. xvernalis*, *C. xhiemalis* and *C. hybrida*. The Principal Coordinate Analyses (PCoA) based on joined molecular and morphological data sets showed a distribution of all accessions in agreement with their taxonomic classification. Taken as a whole, results demonstrated that this set of microsatellite *loci* are eligible in describing genetic diversity in winter camellias, being therefore an appropriate tool for germplasm characterization and management.

S12.106

Distribution and Characteristic of *Paeonia ostii* Populations in China

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Based on the investigation and literature study of wild and cultivar population of *Paeonia ostii*, this paper showed the distribution and habitat feature of *P. ostii*, discuss its possible distribution area. *P. ostii* mainly distribute in Henan and Anhui province. In addition, it also has the distribution in Baokang county in Hubei province, western of Hunan and Liangdang in Gansu province. Because the medicinal value of *P. ostii*, the artificial destruction is serious, which caused the wild resources serious outflowed. Simultaneously, it is the outstanding breeding and the production material, we should enlarge the protection and applied research.

S12.107

European Hazelnut and Almond Genetic Resources: Results and Perspectives of Networking Safenut AGRI GEN RES Activities

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Corylus avellana and *Prunus dulci* are commodities of international economic importance. Different Countries conserved the hazelnut and almond genetic resources. Moreover, traditional genotypes handed by father to son are conserved on farm. The exploitation and evaluation of such germplasm allow retrieving the main part of the genetic variability present even at low frequencies. Within EC AGRI GEN RES, the 'SAFENUT' project (Safeguard of almond and hazelnut genetic resources from traditional uses to modern agro-industrial opportunities), represents an example to coordinate the European genetic resources to share them in a more efficient manner.

The followed strategy included different actions: 1) centralization of available germplasm; 2) harmonization of the standard descriptors 3) recovery of local endangered varieties 4) molecular and biochemical characterization 5) creation of the almond and hazelnut core collection. The final results was to develop a European virtual inventory in order to share and spread all the information as well as to promote a wider application of traditional knowledge, agricultural practices. The project has been benefited from the participation of 11 partners from 6 European Countries. The present work summarizes the main results which focused on the centralization and harmonization of European hazelnut and almond germplasm and describes the survey carried out in different areas of traditional cultivation where novel ecotypes were pre-selected. In the frame of the biochemical evaluation, more than 150 hazelnut and 80 almond varieties were analysed for oil, tocopherol, phenolic and mineral content of the kernel. SSR markers were performed on 300 hazelnut and 154 almond varieties, to avoid synonymous and homonymous. With regard to traditional knowledge, a review of the existing hazelnut exhibitions was carried out. In the present work it is also described the SAFENUT European Database (<http://www.safenut.net>) and its applications.

S12.108

Genetic Characterization of Selected Macadamia Germplasm and Its Implications on Breeding and Conservation in Kenya

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Macadamia is the most important nut crop in Kenya with an estimated annual production of about 13,510 metric tonnes with an estimated value of \$4.8M. The cultivated Macadamia is derived from two species; the rough-shelled *M. tetraphylla* and the smooth shelled *M. integrifolia*. The Kenyan Macadamia industry relies on selection of superior trees from germplasm derived from open-pollinated seeds of the two species planted between 1946 and 1968. Twenty six Macadamia accessions sampled from five populations; Bob Harries, Thika, Kirinyaga and Embu and Meru in central and eastern Kenya respectively were analysed using 110 polymorphic AFLP markers using six AFLP primer combinations. The highest percentage of polymorphic *loci* of 80.0% was observed in the Bob Harries population and the least from Thika population of 67.3%. There were significant differences in the number of different alleles, number of effective alleles and expected heterozygosity with the highest values being observed in the Bob Harries population. Phylogenetic analysis based on UPGMA of genetic distances of the five populations revealed the Bob Harries population, the source of Macadamia germplasm in Kenya to be closely related only to the Kirinyaga population and distantly related to the Thika population, a preservation block of selected superior germplasm. Cluster analysis based on the 26 accessions did not distinctly separate them into species indicating high level of heterozygosity and genetic admixture of the original gene pool used in Kenya. Analysis of molecular variance further indicated significant genetic differences, PhiPT=0.043 at 95% confidence interval, with higher genetic variation within the populations (accessions), 96%, than among the populations, 4%, and high genetic differentiation among the accessions (theta=0.1909). The results of this study indicate high genetic diversity within the Kenyan Macadamia germplasm and provides information that can immediately be useful for enhanced breeding and more effective sampling of populations for conservation purposes.

S12.109

Characterization, Selection and Domestication of Wild Thistle for Processing Purposes

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Thistle (*Cynara cardunculus* var. *silvestris*) is considered a weed in Chile. It was introduced by Spaniards conquerors in century XVI as animal forage. Its petioles and sometimes flowers are consumed fresh in salads or cooked. It is distributed along the range zones within the Central Valley; it presents rusticity, character that could be used as gene source for other edible relative species, or just for domestication and direct use. The aim of this research was to characterize wild thistle populations morphologically and molecularly, to select agronomic and processing characteristics, and to start domestication. Sixty accessions were collected representing each agroclimatic zone where thistle was growing, pieces of rhizome and seeds were taken. Plants were regenerated in CRI La Platina, Santiago, Chile and managed under irrigation and fertilization regime. Each accession was characterized morphologically and other variables for processing purposes were considered, such as dry matter and sugar content. Simple statistic, cluster and discriminant analyses were carried out grouping the populations by morphologic and processing characteristics. After the first selection, a molecular characterization was carried out through the PCR technique RAPD. Molecular and morphological markers gave enough information to establish the genetic diversity present in the collection. Morphological results showed higher concentration of sugars in petioles during spring, reaching 5.8 ± 1.6 °brix, but presenting high coefficient of variation (27%) that indicates genetic variability for this character within the populations growing under similar conditions. Similarly, petiole dry matter varied from $9.2 \pm 1.4\%$ in early spring to $12.8 \pm 3.4\%$ in middle spring, but showing a low coefficient of variation; however during the 2009 spring it presented an average of $9.4 \pm 2.2\%$ and coefficient of variation rises to 24.1%. These results allowed to select 16 populations presenting characteristics useful for processing industry.

S12.200

Somatic Embryogenesis as a Tool of Conservation of Genetic Sources of Rare Genotypes of Autochthonous Population of Norway Spruce

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Tissue culture approaches, in particular somatic embryogenesis, is considered as the advantageous technique for *in vitro* propagation and gene conservation of conifers. Generally, the development of embryos and their conversion into plantlets is closely associated with changes in endogenous phytohormone levels. Beside the key roles of auxin and cytokinins, very important function in differentiation processes belongs to polyamines. The polyamine (PAs) contents and activities of polyamine biosynthetic enzymes in Norway spruce somatic embryos (*Picea abies* L. (Karst.), highly responsible genotype) were studied in relation to anatomical changes during their development - from proliferation to germination. Activities of enzymes and PA contents steadily increased during development from embryogenic suspensor mass until early cotyledonary stages. The enzyme activity subsequently declined in mature cotyledonary embryos, accompanied by sharp reductions in PA contents (with the exception of spermine, its level significantly increased during the desiccation phase). The predominant PA in somatic embryos of highly responsible genotype was spermidine. This knowledge led as to compare embryo PA levels of this genotype with less responsible ecotype of Norway spruce. Embryos of less responsible ecotype contained rather low level of PAs, predominant PA was putrescine and the content of spermidine was many times lower than in embryos of highly responsible genotype. The application of putrescine and spermidine into the growth medium in order to improve the efficiency of somatic embryogenesis of less responsible genotypes is discussed.

S12.201

In vitro Conservation and Cryopreservation of Endemic Taxa *Dianthus giganteus* ssp. *Banaticus*

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Dianthus giganteus d'Urv ssp. *banaticus* (Heuff.) Tutin (fam. *Caryophyllaceae*), is an endemic species in SW of the Carpathians, considered vulnerable/rare in Romania. Besides zoologic importance, its ornamental valences give it a plus for population conservation both *in situ* and *ex situ*. Maintaining several genotypes, belonging to different areas, in *ex situ* collections constitutes insurance for the conservation of certain species. The *ex situ* conservation methods used in our study are *in vitro* culturing and cryopreservation. The vegetal material was collected from natural sites. The initiation of the *in vitro* cultures was done from stolons and sterilized seeds, using three sterilizing methods based on sodium hypochlorite (Domestos) and mercuric chloride. The sterilization with sodium hypochlorite was less efficient (18 - 63% sterile explants), compared to the one with mercuric chloride (37 - 84% sterile explants), and the combined method gave the best results (60 - 100% sterile explants). As culturing medium three phytohormonal variants were used: (i) for initiation (NAA 1 mg/l; BAP 1mg/l), (ii) for multiplication (NAA 0,1mg/l; BAP 1mg/l), (iii) for rooting (without phytohormones). The study focused on the multiplication related to the culturing medium, alongside with the type of the explants and its position. For the cryopreservation experiments (in liquid nitrogen, at -196 °C) apical meristems from vitroplants were used. These meristems had 2-4 pairs of primordium foliar and a length of approximately 3-4 mm. We also studied the influence of the treatment with different sucrose concentration solutions on the regenerative ability of the apexes before and after cryopreservation. The highest regenerative rate after cryopreservation was obtained with the 0.5M sucrose solution (43% regenerated apexes).

S12.202

Effect of Light and Activated Charcoal on Growth of *Archontophoenix alexandrae* (F. Muell.) H. Wendl. & Drude (*Arecaceae*) Zygotic Embryos *in vitro*

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The *Archontophoenix alexandrae* species has great ornamental effect and has been cultivated for the palm heart extraction. It is propagated by seeds but this process is a problem in breeding programs due to high heterozygosity in the population. The knowledge of the *in vitro* germination will enable the work progress on somatic embryogenesis. This work had the objective to study the effect of light and activated charcoal on growth of *A. Alexandra* zygotic embryos. The work was conducted at University of São Paulo, Piracicaba, São Paulo State, Brazil. First, were studied the presence and absence of light in the zygotic embryos inoculation with 10 replications in a completely randomized design. In the second were studied 5 concentrations of activated charcoal (0, 1, 2, 3 and 4 g.L⁻¹) in culture media and 4 replications in a completely randomized design with 10 embryos per replication. The variables evaluated were length, width and embryo diameter (to the light effect in 30 days) and shoot length, root number, root average length and mass of total dry matter (for both experiments in 30 and 60 days). Means were compared by Tukey test. The oxidation rate of this species was not high (on average 12%), but in the dark and presence of activated charcoal, the oxidation rate was lower. There was greater shoot length in 30 days, but equal mass of total dry matter for embryos developed in the light absence, indicating shoot etiolating. In 60 days there was greater shoot length and mass of total dry matter indicating shoots etiolated by the light absence that kept the difference and more vigorous. Thus, the development of *A. alexandrae* zygotic embryos kept in conditions of light absence in the first 30 days after inoculation was beneficial. The activated charcoal did not affect the embryo development.

S12.203

Performance of Coconut Embryo Culture Accessions Introduced at International Coconut Genebank for Latin America and the Caribbean (ICG-LAC)

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The present paper is a partial report for the introduction of eleven accessions of dwarf coconut palm and one accession of giant coconut palm into International Coconut Genebank for Latin America and the Caribbean (ICG-LAC), a partnership between Embrapa Coastal Tablelands (CPATC) and Bioversity International, coordinated by Coconut Genetic Resources Network (COGENT). These accessions, originated from International Coconut Genebank for Asia, India and Oceania (ICG-AIO), situated at Ivory Coast, were introduced in Brazil through zygotic embryos in November 2008. The mature fruits and endosperm cylinders with embryos were submitted at Ivory Coast to a phytosanitary treatment process according to international technical recommendations for safe movement of vegetal germplasm. At CPATC Laboratory for plant tissue culture the embryos were excised and inoculated in Y3 culture medium. The first evaluation occurred after nine days of inoculation, and was verified that 1.38% of the embryos were contaminated by fungi and 15.30% by bacteria. After 12 months was observed variation of *in vitro* development within and among accessions and high percentage of non germinated and oxidated embryos. The normal germination percentage varied from 3.33% to 33.3% for, respectively, Niu Leka Dwarf and Malayan Green Dwarf accessions. The non germinated embryos percentage varied from 18.47% to 47.72% for Tahitian Red Dwarf and Sri Lanka Green Dwarf accessions, respectively. After 12 months, the contamination was higher for Niu Leka Dwarf (70%). The percentage of non germinated and oxidated embryos varied from 18.47% to 47.72% (Tahitian Red Dwarf and Sri Lanka Green Dwarf, respectively). The great number of collected cylinders with exposed embryos (not protected by endosperm) and cracked endosperm, in addition to the long storage time (7 to 11 days) from collection to inoculation, favored bacteria proliferation. The non uniform stage of fruits/embryos maturation contributed for high variation of *in vitro* development.

S12.204

In vitro Cold Storage of Sour Cherry (*Prunus cerasus* L.) Shoots is Affected by Carbon Source and Nitrogen Concentration

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In vitro cold storage of fruit crop germplasm is a useful tool in the preservation of heritage or commercial cultivars. Shoot cultures of sour cherry (*Prunus cerasus* L.) cultivars Dolgozdannaya, Moya Radost and Zukovskaya, were cold stored at 4 °C in either five-section tissue culture bags or in 150 ml glass jars. Carbon sources 3% sucrose, 2% or 3% mannitol, or 2% sucrose + 2% mannitol were tested in Murashige and Skoog (MS) medium with or without plant growth regulators (PGRs). Nitrate nitrogen at 100%, 50% or 25% of the normal MS concentration was also tested. Shoot cultures of the three cherry cultivars could be stored for over 30 months at 4 °C and remained in excellent condition in some treatments. There was significant variation in the storage duration with interactions of the cultivar, treatment, and container. Sucrose was the best carbon source for all three genotypes and allowed storage for up to 36 months. Shoots stored on 2% or 3% mannitol survived for only 6 to 12 months while the combination of 2% mannitol and 2% sucrose extended storage to 30 months for two of the three genotypes. The addition of abscisic acid to 3% sucrose MS medium significantly decreased storage length. Fifty-seven accessions of sour cherry germplasm were stored in tissue culture bags on 3% sucrose MS medium without PGRs and remained in good condition for 13 to 30 months. The 68 accessions of the *in vitro* *Prunus cerasus* germplasm collection are now stored in tissue culture bags with MS medium, 0.5 mg/l BAP, 0.1 mg/l IBA, and 3% sucrose.

S12.205

Establishment for *in vitro* Propagation and Conservation Protocols of Mangaba Tree Native of Brazil

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The mangaba tree (*Hancornia speciosa* Gomes) is a native plant found in different regions of Brazil, which has great importance because of its fruit produce pulp for use in the juices and ice creams marketing. This study aimed to apply the tissue culture techniques for asexual propagation and conservation of mangaba native populations of northeastern of Brazil. For *in vitro* propagation were studied the sealing bottles, explant types, culture medium and growth regulators for induction of *in vitro* rhizogenesis. For *in vitro* conservation the effect of mannitol and abscisic acid in interaction with different types of explants and sealing of vials were studied. After the work was possible to say that the best sealing types were PVC film and parafilm® to the establishment stage, parafilm® for the first and second subculture and PVC film just for the second subculture. The best explants for the first subculture were the median and basal nodal segments. There was no significant effect of the explants in the second subculture. The ideal subculture interval was 50 days. For rooting, at 60 and 90 days of *in vitro* culture, was a higher numerical value for the number of roots in the presence of 400 and 600-L⁻¹ of indol butyric acid. In the presence of mannitol the length of the shoots showed minor values of the control, but after 90 days was observed toxic effect of mannitol in the explants. The abscisic acid at 0.5-L⁻¹ presented better results for *in vitro* conservation of mangaba micro-cutting cultured in the vials sealed with aluminum paper.

S12.206

In vitro Propagation of *Colutea gifana*, a Rare and Endangered Plant Species of Iran

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In vitro methods provide a variety of tools to supplement traditional methods for collecting, propagating and preserving endangered plant species. In this study, an efficient protocol was developed for *in vitro* propagation of *Colutea gifana* a rare and endangered plant species with limited reproductive capacity that grows in a very narrow area of Iran. Single nodes explants were used for a series of experiments to select appropriate disinfection method and growth regulators for establishment, proliferation and rooting stages. Explants showed the highest establishment percent after 15 min treatment with 2% Sodium hypochlorite (NaOCl) cultured in MS medium plus 2.2 µM 6-benzylaminopurine (BAP) and 1 µM indole-3-butyric acid (IBA). BA was more effective cytokinin in comparison to Thidiazuron (TDZ) and Kinitin in proliferation stage. *In vitro* rooting of proliferated shoots were induced in half-strength MS medium with both tested auxins i.e. IBA and α -naphthaleneacetic acid (NAA). Eighty percent of the plantlets were successfully acclimatized to *ex vitro* conditions, exhibiting normal development. These plantlets can be used to replenish declining populations in the wild to conserve *C. gifana* from extinction and also for further studies about this species.

S12.207

Screening the *in vitro* Morphogenetic Reaction of Different Explants at Round Pepper (*Capsicum annum* L.)

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In Romania, as well as worldwide, peppers are one of the most important crop, the annual production in 2007 being, according with the FAO database around 184,939 tonnes (FAO, 2009). It is not a wonder why the efforts of specialist tagger for the continuous improvement of these plants, both for increased production or enhanced agricultural traits (resistance to pests and diseases, quality of production, etc). Development and optimizing of efficient protocols for *in vitro* regeneration can open new possibilities for speeding up the production of improved cultivars. Although, some information is available on the morphogenesis of *Capsicum* (Agrawal *et al.* 1989, Harini and Sita 1993, Fari and Andrasfalvy 1994, Steinitz *et al.* 1999, Kintzios *et al.* 2000, Ochoa Alejo and Ramirez-Malagon 2001), many of these investigations did not report satisfactory result in terms of enhanced number of shoots. The morphogenesis response seems to be highly dependent PGRs used in the media, which is again cultivar and genotypic specific. Genetic stability of the tissue culture raised pepper plants also needs to be addressed. In order to support the breeding activity, a primordial condition is the establishment of a viable and rapid multiplication technology, specific for each species, which should allow the regeneration of a sufficient number of plants in the shortest period of time. Different combinations of growth hormones were tested in order to achieve regeneration structures from young tissues of pepper. Kinetin (KIN) and α -naphthaleneacetic, Kinetin (KIN) and indolilactic acid (IAA), or benzilaminopurine (BA) and NAA or IAA, zeatine were added to basic medium composed of full strength MS salts, MS vitamins, 3% sucrose and 0.8% agar-agar. Young explants (shoot tips, hypocotyls and leaves) derived from aseptic germination of seeds were cultivated on these media.

S12.208

In vitro Study on Effects of Salinity on Growth, Proteins and Antioxidant Enzymes in Some *Onobrychis* Species

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Onobrychis Miller (*Fabaceae*) comprises 77 species in Iran. Some species such as *O. viciifolia* are very important as forage and soil improvement crop well adapted to dry and semi-dry regions and resistant under hard environmental conditions. The effects of salinity on some growth parameters, protein content and antioxidant enzymes were studied in three *Onobrychis* species with two base chromosome numbers in two ploidy levels including *O. viciifolia* ($2n = 28$), *O. melanothrica* ($2n = 16$) and *O. subnitens* ($2n = 14$). Seedlings of the species were subjected to different levels of NaCl stress for two months. Salinity affected the growth parameters and caused a reduction in germination percentage, relative growth rate (RGR), and relative water content (RWC) with a greater reduction in *O. subnitens*. Protein content in both *O. melanothrica* and *O. subnitens* increased up to 150 mM NaCl, but this increase in *O. viciifolia* occurred at 200 and 250 mM NaCl. *O. melanothrica* exhibited a decrease in peroxidase (POX) and poly phenol oxidase (PPO) activities under NaCl stress, while *O. viciifolia* showed a remarkable increase in these enzymes between 50 to 300 mM NaCl. In *O. melanothrica*, POX and PPO activities increased at 50 mM NaCl and then decreased at higher levels. The results showed that the differences in the antioxidant activities of seedling may, at least in part explain the greater tolerance of *O. viciifolia* comparing to *O. melanothrica* and *O. subnitens*. Then, *O. viciifolia* (tetraploid) showed a better protection mechanism against salinity induced oxidative damage than other studied species.

S12.209

Anther-Stigma Separation in Four *Papilionoideae-Fabaceae* from Northwestern Argentina

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Anther stigma distance is a trait highly associated with breeding systems. It has long been assumed that stigma exertion promotes outcrossing. We studied this trait in *Indigofera suffruticosa* Mill., *Galactia latisiliqua* Desv., *Rynchosia edulis* Griseb. and *Zornia contorta* Mohl. (*Papilionoideae, Fabaceae*). They are species with ornamental

potential and may be used for forage given its high nutritional value. We studied natural populations located in Lerma Valley, Salta Province, Argentina. The populations are located in an Eastern Andes seasonal rainforest (Yungas). In January and February of 2009 we marked 10 floral buds and 10 flowers at the end of anthesis (opened flowers) from 30 individual plants per species and population. Anther-stigma separation was determined at 7 a.m. in the floral buds and at 8 p.m. in the opened flowers. Two distances were measured with calipers: (a) from the base of the ovary to the top of the anthers, and (b) from the base of the ovary to the tip of the stigma. Both distances were measured to the nearest 0.1 mm. Stigma position was then calculated by subtracting the height of the anthers from the height of the stigma. Anther-stigma distance varied between -0.26 and 1.24 mm in floral buds and between 0.93-1.54 mm in opened flowers. *Galactia latisiliqua* and *Rynchosia edulis* presented the stigma below the anthers in floral buds and showed the lowest values in anther-stigma distance both in buds and opened flowers. At the end of anthesis, all species showed the stigma protruding above the anthers. *Indigofera suffruticosa* and *Zornia contorta* showed the highest separation in both floral stages. For all the studied species, the level of selfing was inversely correlated with anther-stigma separation.

S12.210

Regeneration of Cross-Pollinated Plant Genetic Resources

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Crop Research Institute, Department of Vegetables and Special Crops Olomouc, is a holder of Czech genetics resources of vegetable and medicinal, aromatic and culinary plants (MAPs). Its collections composed of 9 236 vegetable accessions and 836 MAPs accessions. 75 % of these materials are preserved as a seed samples in the long term conservation conditions of gene bank. About 1 500 accessions of vegetable and/or MAPs are regenerated every year. Regeneration of cross-pollinated species is carried out in technician isolation - stabile (150 pieces) and/or mobile (100 pieces) isolation cages are used every year. Controlled pollination is provided by honey-bees (*Apis mellifera* L.) and/or humble-bees (*Bombus terrestris* L.). About 200 honey-bee clusters and 20 humble-bee nests are installed every year and methods of its keeping and maintaining are used and proved in Olomouc since 1985. Factors as commercial accessibility, pollination affectivity, lifetime, aggressiveness as well as financial balance of acquisition were compared. Long term experiences brought knowledge of each pollinator's advantages and disadvantages for individual crops and many examples can be mentioned.

S12.211

In situ and on Farm Conservation of Central European Fruit Genetic Resources

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The area of the Czech Republic has been a centre of genetic diversity of many fruit species. Due to the continuing destruction of old plantations and alleys since the half of the last century, there has been serious threat of extinction of fruit landraces representing the most valuable part of genetic diversity at the territory of the Czech Republic. Because of the possible extinction of these indigenous landraces, the program for collecting and long term conservation of fruit landraces has started in nineties of the last century. This paper describes conservation that employs both *in situ* and on farm techniques in a complementary manner. *In situ* conservation is considered a conservation of wild species in the natural habitat. The authors extend the term for cultivated fruit species naturalized in the landscape. Important landraces found within 12 regions of the Czech Republic were registered, evaluated and *in situ* localized by GPS. In total, 848 accessions of fruit woody species were marked for *in situ* conservation. As another form of preservation, on farm plantations were established for conservation of important accessions in particular region. Planting material of these accessions was prepared on seedling rootstocks. Concrete

accessions (cultivars) for on farm conservation in given locality were selected according to the incidence and representation of genotypes in particular areas. This information was obtained by localization of fruit trees *in situ* and determination of concrete cultivars during collecting expeditions. At present, four on farm plantations KRNAP Vrchlabi (48 accessions), "Orchard of Reconciliation" Neratov (53 accessions), National park Podyji - Znojmo (13 accessions), National park Sumava (18 accessions) were successfully established in a traditional form of large orchard trees. Four established on farm plantations ensured long term preservation of landraces in original areas.

S12.212

Proposal of an Exploration and Collection Survey of Wild and Cultivated Olive (*Olea europaea* L.) Genetic Resources by GIS Techniques and Multivariate Models

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Last two decades have witnessed significant advances in the characterization of plant genetic resources mainly due to the development of DNA analysis techniques. However most of the exploration and field germplasm collection methods are based on inaccurate vegetation maps and local information. This study focus on the development of a new methodology for surveying cultivated (*Olea europaea* subsp. *europaea* var. *europaea*) and wild (*Olea europaea* subsp. *europaea* var. *sylvestris*) genetic resources by using of GIS (Geographic Information Systems) and multivariate models of spatial analysis. The preliminary results about the identification and characterization of environmental variables (topography, climate, lithology, geomorphology, etc.) which determine the distribution areas of olive in Andalucía (Spain) are discussed. Ulterior use of multivariate spatial models (GAM (Generalized Additive Models) or ENFA (Ecological Niche Factor Analysis)) will try to: a) obtaining a map of olive habitability according to limiting ecogeographic factors; b) identifying areas with different ranges of habitability for this specie based on specific environmental variables, and c) identifying areas that maximize the genetic diversity of the olive tree. In summary we propose this modelling system for maximizing the efficiency of the exploration and collection surveys of olive genetic resources for both conservation and breeding purposes.

S12.213

The Evaluation of Apple (*Malus xdomestica*) Genetic Resources by Molecular SSR Analysis

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Apples (*Malus xdomestica* Borkh.) belong to the main fruit species and they are the most important fruit in Europe. The apple genetic resources (1087 varieties), which included current and old world varieties, our regional and local varieties, are maintained in Research and Breeding Institute of Pomology Holovousy Ltd.. In present, the utilization of DNA molecular genetic methods is the best suitable method for evaluation of individual genotypes, so that we eliminated duplications and characterized genetic relationships. 195 current, old, local and special apple varieties were selected for molecular genetic evaluation. DNAs, which were used for molecular genetic analysis, were isolated from selected apple genotypes. SSR molecular genetic analysis of six microsatellite *loci* was carried out on DNA samples. We found high amplified polymorphism of SSR markers among apple varieties, as all amplified products (83) were polymorphic. Analysis results were statistically processed and based on it, analysis of genetic relationships of individual genotypes of apple varieties was proceeded. We mapped the wide molecular genetic biodiversity of apple varieties. The most of current apple varieties was clustered together into several groups. Special apple varieties were clustered separately in last group.

We found no differences between individual plants of varieties James Grieve and Mc Intosh, respectively. We also found no differences between normal and Red mutation plants of varieties James Grieve, Mc Intosh, Oldenburg, Melba, Mantet and Wealthy. In set of apple genotypes, we found duplications of Belréne with Parména gold winter and Glencross with Mantet, respectively.

S12.214

The Use of Resistance Molecular Markers for Evaluation of Apple (*Malus xdomestica*) Genetic Resources

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Apples (*Malus xdomestica* Borkh.) belong to the main fruit species and they are the most important fruit in Europe. Scab (*Venturia inaequalis* CKE.) and powdery mildew (*Podosphaera leucotricha* (Ellis & Everh.) E.S. Salmon) are the most significant apple fungal pathogens. A use of resistant plant materials is one of the best possibilities how to defend from their infection. Apple genetic resources include genotypes with different level of resistance to fungal diseases. There is a wide variability of resistance genes in genetic resources collection and the utilization of DNA molecular genetic markers is the fastest method for evaluation and characterization of individual genotypes and founding of new resistance sources. 226 resistance, current, old, local and special apple varieties were selected for molecular genetic evaluation from apple genetic resources (1087 varieties) of Research and Breeding Institute of Pomology Holovousy Ltd.. DNAs were isolated from selected apple genotypes for molecular genetic analysis. Molecular genetic analyses of nine markers to scab and powdery mildew resistance were carried out on DNA samples. Amplified products of molecular markers for individual resistance genes to scab (Vf, Vm, Vbj, Vr and Vh) and powdery mildew (Plw, PII and Pld) were detected in analyses of apple genotypes. Molecular marker of the most effective resistance gene Vf was detected in 29 scab resistant, 3 current apple varieties and 3 special genotypes. Molecular markers of scab resistance genes Vr and Vh were detected in 18 scab resistant, 37 current, 64 local and old varieties and 4 special genotypes. Molecular markers of scab resistance genes Vm and Vbj were not detected in any genotype. Molecular markers of powdery mildew were only detected in 2 old varieties and 4 special genotypes.

S12.215

Using Morphological and Genetic Markers for Identification of Clones within 'Khalili' and 'Askari' (*Vitis vinifera* L.)

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Two different marker systems were used to characterize intra-varietal diversity within Khalili and Askari cultivars. These two semi-seedless grapevine cultivars are important for table grape and raisin production. Some accessions of these two cultivars are morphologically different which are useful for their economical value. In order to evaluate these differences, we selected 14 SSRloci (VVS2, VVMD5, VVMD7, VVMD14, VVMD21, VVMD25, VVMD27, VVMD36, ZAG47, ZAG62, ZAG79, VMC6d12, VMC6c7 and VMC6g8) and 32 morphological markers to identify intra-varietal diversity. The 42 accessions of "Askari" and 15 accessions of "Khalili" were analysed. In "Askari" eight polymorphic markers (VVS2, VVMD5, VVMD21, VVMD36, ZAG62, ZAG79, VMC6d12 and VMC6c7) detected six divergent accessions (no. 3, 21, 31, 44, 46 and 114). Accessions no. 46 and 114 were identical. The remaining 36 accessions were identical. In "Khalili" four polymorphic markers (VVS2, ZAG79, VMC6c7 and VMC6g8) detected five divergent accessions (no. 2, 6, 8, 15 and 27). The remaining ten accessions were identical. Some of these accessions were differentiated by morphological markers and clustered in same groups in genetical and morphological dendrogram.

S12.216

Assessment of Genetic Diversity in Sugar Apple Germplasm Using RAPD Marker

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In Brazil sugar apple is an important crop mainly in semiarid regions. Diversity relationship in 64 sugar apple germplasm accessions collected from North of Minas Gerais state were analyzed by using RAPD marker. Using 20 selected RAPD primers 169 fragments were generated, of which 48 bands were polymorphic (28.7%) and 119 were monomorphic (72.3%) producing on an average of 2.4 polymorphic bands per primer. Primer OPF 19 generated eight polymorphic bands. Low percentage of polymorphism (<29%) was observed indicating low level of genetic variation between the 64 accessions evaluated. Genetic relationship using similarity Jaccard's co-efficient were estimated. Values between different pair of accessions varied from 0 to 0.17834 suggesting low diversity. Accessions from different cities were found to cluster together indicating no correlation between molecular grouping and geographical origin. The dendrogram revealed three clusters. The first cluster grouped C19 and G29 accessions collected from Verdelândia and Monte Azul, respectively. The second cluster grouped G16 and B11 accessions collected from Monte Azul and Coração de Jesus, respectively. The third cluster was formed by three sub clusters grouping 60 accessions. In summary the RAPD marker was effective in discrimination and revealed duplicates from germplasm collection. Despite the low genetic diversity some types may be used as parents in breeding sugar apple program.

S12.217

Molecular Characterization and Genetic Diversity of *Citrus aurantium* L. Germplasm from Central Italy

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The genus *Citrus* L. belongs to the subfamily *Aurantioideae* of the *Rutaceae*. Plants of *Citrus* genus are generally distributed in the Eastern hemisphere (Davies and Albrigo 1994) and probably introduced in the European areas by Arabs and by the Crusaders (Moore, 2001). In Italy, during past decades, the species *C. aurantium* called "sour orange" had an high economic and social importance: the fruits were used for food traditional dishes and for the essential oil extraction, moreover this species is the most used *Citrus*-rootstock. The aim of this work is to make a genetic characterization and to evaluate the level of genetic differentiation of 19 different accessions of sour orange from Umbria region (Central Italy) using microsatellite (SSRs) markers. Our results highlight a low genetic diversity for the examined *C. aurantium* accessions; the SSR markers identified four different genotypes and four *loci* with one private allele. The identification of *loci* displaying unique alleles could have practical applications as they may be used as an important tool for juvenile germplasm identification. This study can be considered a starting point for future gene conservations programs of *C. aurantium* species in the Italian Central region.

S12.218

Genetic Diversity of Red Skinned Chinese Sand Pear Cultivars Revealed by AFLP Markers

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A total of 38 Chinese sand pear cultivars and landraces (mostly red skinned types) native to Southwestern China were subjected to Amplified Fragment Length Poly-

morphism (AFLP) analysis. The six primer combinations (*EcoRI*-AAG/*MseI*-CAT, *EcoRI*-ACT/*MseI*-CTT, *EcoRI*-AAC/*MseI*-CAG, *EcoRI*-AAC/*MseI*-CTA, *EcoRI*-AAC/*MseI*-CAA, *EcoRI*-AAG/*MseI*-CTC) could generate 443 fragments from 38 pear accessions, of which 356 fragments were polymorphic. The percentage of polymorphism for six primer combinations was from 76% to 85% with a mean of 80%. Primer *EcoRI*-ACT/*MseI*-CTT showed highest fragment, polymorphic fragments, percentage of polymorphism and marker index. Six primer combinations could successfully distinguish 32 cultivars, of which one pair of cultivars might be homonym. Three pairs of cultivars could not be differentiated, which might belong to the synonymy cases or bud mutants. Dendrogram analysis clustered the accessions into five groups. 'Huobali' types, a very common name referring red pears harvesting in late July and early August in Yunnan Province, scattered in different groups, revealing the possibility of different origin. The red skinned sand pears clustered with the non-red skinned sand pears which could be presumed that the red skinned sand pears might be from the bud mutation or free pollinating seedlings of non-red skinned sand pears.

S12.219

Chloroplast DNA Analysis in Mandarins (*Citrus* spp.) and their Close Relatives Using Cleaved Amplified Polymorphic Sequence (CAPS) Marker

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Variability of four chloroplast DNA regions, i. e., *rbcl*-ORF106, *trnF*-*trnV*, *trnV*-*rbcl* and *trnK2*-*trnQ*, was examined in 39 mandarins and their close relatives and compared with two true citrus species, pummelo (one accession) and citron (two accessions). The combination of all polymorphic restriction patterns resulted in eight chloroplast genome types (CGTs), i. e., CGT1, CGT2, CGT3, CGT4, CGT5, CGT6, CGT7 and CGT8 for the accessions analyzed. Based on the estimated genetic dissimilarity matrix, a phylogenetic tree was constructed for the above chloroplast genome types in which four main clusters were separated as follows: 1) Mandarin cluster: which is sub-divided into five sub-clusters (CGT1, CGT2, CGT3, CGT4 and CGT5). In the sub-cluster CGT1 the mandarins, namely, 'Aoshima unshiu' (Satsuma mandarin), 'Clementine', 'Ponkan', 'Dancy', 'Mediterranean' (or 'Willow-leaf'), 'Avana tardivo', 'King', 'Late', 'Encore', 'Kinnow', 'Wilking', 'Fremont', 'Kara' were placed with 'Kishu', 'Mukaku kishu', 'Hira kishu', 'Fukure mikan', 'Shikaikan' and 'Ji mikan'. The mandarins 'Shiikuwasha' and 'Cleopatra' with identical restriction patterns were placed in sub-cluster CGT2. 'Sunki' was placed with 'Tankan' in the sub-cluster CGT3. The mandarins 'Tachibana' and 'Kouraitachibana' each was placed in a single-accession sub-cluster CGT4 and CGT5, respectively. 2) Calamondin (or 'Shikikitsu') (CGT6) cluster. 3) Pummelo (CGT7) cluster: in this cluster the accessions, 'Obeni mikan', 'Kunenbo', 'Yatsushiro', 'Keraji', 'Kabuchi', 'Kikai mikan', 'Oto', 'Binkitsu', 'Giri mikan', 'Kobeni mikan', 'Ogonkitsu', 'Ujukitsu' and 'Horai-kan' were not discriminated from each other as well as from the pummelo accession, 'Egami'. 4) Citron (CGT8) cluster: including 'Etrog' and 'Marubusshukan' cultivars. The highest (0.258) and the lowest (0.008) genetic dissimilarity values were found between CGT4 and CGT8, and between CGT2 and CGT3, respectively.

S12.220

Inheritance and Molecular Marker of Resistance to Marssonina Blotch (*Marssonina mali*) in Apple (*Malus domestica*)

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Marssonina blotch, caused by *Marssonina mali* (P. Henn) Ito, is one of the most serious leaf diseases in apple (*Malus domestica* Borkh.). It results in a large annual crop loss in apple production in Asian countries. In order to analyze the inheritance of and to screen for the SSR and AFLP markers for the resistance to marssonina blotch, leaves of 1401 6-year-old hybrid seedlings (Jonathan × Golden Delicious) were inoculated *ex situ* with spore suspension of *M. mali*. The number of infected spots per inoculated leaf segregated significantly among individual seedlings. From the frequency distribution diagram, two distributions were distinctly divided by the criterion of 230 spots / leaf. Chi-square test showed that the segregation ratio of the two distributions (1060:341) was not significant with 3:1, indicating that the variance of resistance is concerned with the segregation of one or two alleles of major genes, resistant is dominant against susceptible. According to the inheritance of the resistance, segregation population for SSR and AFLP analysis was constructed by 89 resistant and 102 susceptible seedlings. A 300bp and a 350bp specific fragment were amplified in resistant individuals by SSR primer CH01h10, which is located on linkage group 8, and AFLP primers E-TT/M-GAA, and the map distance of CH01h10-300 and E-TT/M-GAA-350 away from the target gene *locus* is 10.1cM and 3.7cM respectively.

S12.221

SSR Based Characterization of Grape (*Vitis vinifera* L.) Germplasm from Southeast Anatolia

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Southeast Anatolia located in close proximity to the center of origin of grapes is an important grape producing area of Turkey. The strategic location of this region together with its diverse ecological conditions has led to the development of local grape germplasm that is unique to this region. However, so far little has been done to genetically analyze this grape germplasm. In this study, we genetically analyzed 55 grape cultivars of this region using 14 simple sequence repeat (SSR) *loci* and a number of ampeological characteristics. We identified three cases of synonymous and four cases of homonymous cultivars. In addition, local centers of high genetic diversity within the region were identified. However, no correlation was found between the genetic relatedness and the geographical locations of the cultivars. The results reported here are important towards better characterization of the grape cultivars of the region and would aid future grape germplasm management and breeding efforts.

S12.222

Molecular Characterisation of *Prunus* Accessions of Traditional Cultivars Prospected in Western Andalusia, Spain

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Forty-six accessions of *Prunus* spp. belonging to 19 presumably local cultivars have been prospected in different orchards located in Olivares, Constantina (Sevilla) and Galaroza (Huelva) in Spain, within the frame of the project INIA RF2007-00027-C06-05. DNA was isolated from leaf samples and analysed with a set of 6-8 SSR *loci* previously selected by Wünsch (2009) for its good performance and transferability in different *Prunus* spp. All 19 cultivars were fully discriminated. No cases of synonymy or homonymy were found, i.e. all samples from different trees of the same cultivar were correctly assigned. The dendrogram obtained with the UPGMA clustering method and Dice coefficient shows five different groups according to the

different species: peach, apricot, plum (2 clusters) and cherry. The plum group was the more numerous as well as the more complex one: multiple amplification products (4-6 alleles/*locus* and sample) reveal the polyploid character of most of the samples.

S12.223

Microsatellite Markers (SSR) as a Tool to Assist in Identification of European Plum (*Prunus domestica*)

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European Plum (*Prunus domestica* L.) is a valuable commercial fruit. For economic as well as for scientific reasons it is important to distinguish European Plum cultivars for breeding, cultivation and germplasm collection and there is a demand for a rapid and reliable method to identify cultivars for registration, protection, cultivation and management. Normally morphological traits are used to identify cultivars but these traits are often differently expressed in different environments and with changed production practices. DNA-based markers are useful for germplasm identification, diversity analysis and verification of rootstock identity. Among molecular markers, microsatellites, or SSRs, are known for being highly polymorphic, codominantly inherited, abundant and evenly distributed in the genome. The most widespread technique for SSR detection is PCR with specific primers, as this approach is simple, reproducible and suitable for automation. SSRs provide a more reliable method for DNA fingerprinting compared with RAPDs. SSR markers have been used widely for cultivar identification, genetic mapping and phylogenetic studies. Our objective was, parallel to the usual morphological identifying methods, to fingerprint a subset of 45 European plum cultivars with 7 SSR primers, to determine if there was sufficient polymorphism to differentiate the cultivars, and furthermore, to establish one routine method for cultivar identification for European plums at the KOB, and then use this new method as a practical technique to differentiate two unknown samples from each other, to exam the authenticity and purity of cultivars for the nursery, import and export chains and to support quality inspection in the food trades. 7 SSRs revealed polymorphism. The polymorphism information content (PIC), frequencies of 7 SSR *loci* distributed found in 45 plum cultivars are described and discussed.

S12.224

Application of an Extended Set of Microsatellite DNA Markers for the Analysis of Presumed Synonym Cultivars of Apple

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South Tyrol (Northern Italy) harbours the largest continuous apple producing area in Europe with an annual production of more than 1.1 million of metric tons. However, 95% of the total apple harvest is made up of only eight globally distributed cultivars. Consequently, many old, regional varieties with unique characteristics have disappeared or are at risk of extinction. In the last years, several projects were carried out in order to identify, describe and protect the remaining local genetic diversity of apple in South Tyrol. Since accurate identification is an important prerequisite for the inclusion of local apple cultivars into germplasm collections, a data base with genetic profiles of confirmed reference cultivars was established as an alternative tool for determination of unidentified samples. Each reference cultivar was collected from at least two different germplasm collections and analysed at 14 microsatellite *loci*. In ten cases identical genotypes were found for cultivars with similar but distinct names. In order to rule out the possibility that the number of 14 microsatellite *loci* analysed was too low to distinguish closely related cultivars, these samples were analysed at 34 additional microsatellite *loci*. The results obtained are discussed, also with regard to some common genotyping errors.

S12.225

SSR Markers Analysis of the Sardinian Grape Germplasm

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The island of Sardinia has a long tradition in viticulture and a several number of grapevine cultivars documenting the *Vitis vinifera* L. genotype exchanges, introductions and cultivation. This study represent a detailed fingerprinting of a grapevine Sardinian collection, with the objective of improving the management of the local germplasm and provide information on the current state of grape genetic resource conservation. Genotyping of accessions (*Vitis vinifera* L.) was achieved by applying 12 highly polymorphic microsatellite loci (VVS2, VVMD5, VVMD7, VVMD27, VVMD32, VVMD36, VrZag21, VrZag25, VrZag62, VrZag64, VrZAG79). The genetic relationships within and between Sardinian and international cultivated grapevine were examined, to clarify the cases of synonymy and homonymy in the collection. The genetic profiles of 120 local, Italian, Spanish, French and Portuguese accessions were defined. A total of 109 alleles were found and 11 private alleles belonging to autochthonous and Portuguese cultivars were observed. Cases of synonymy were identified: the pairings 'Zirone Alzu' - 'Zirone di Spagna', 'Bi-anchedda' - 'Barriadorgia Bianca', and 'Grenache' - 'Tocaj' - 'Garnacha Blanca'. Our results did not confirm the previously recorded synonymy between the cultivars 'Cannonau' and 'Garnacha'. The Sardinian 'Cannonau' variety showed a different genetic constitution with only 82% of alleles in common. Model-based clustering method did not evidenced a clear separation between local and international cultivars clustered at similarity value of 0.11. The 'Bovale' group, including Sardinian and Spanish cultivars clustered in the two subgroups: 'Cagnulari' - 'Bovale sardo' - 'Graciano' and 'Carignano' - 'Bovale grande' - 'Mazuela', as previously reported. The information obtained can assist in the management of the grapevine collection and in defining an appropriate *in situ* conservation strategy, in order to valorize the traits potentially useful to viticulture and oenology.

S12.226

RAPD Markers Reveal Polymorphism among Iranian Mahaleb Genotypes

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RAPD markers reveal polymorphism among Iranian mahaleb genotypes Taraneh Gharooni, Zabihollah Zamani, Naser Bouzari To estimate diversity and relationships RAPD marker was used among 28 mahaleb genotypes from various geographical regions of Iran as well as 'Lambert' sweet cherry and 'Hamedan' sour cherry (as outgroup samples). Totally, from 100 tested random TIB MOLBIOL primers, 16 were polymorphic and produced 160 fragments among which 104 were polymorphic. Genetic similarity coefficients ranged from 0.1 between sweet cherry cv. Lambert and Ker4 mahaleb genotype to 0.88 between two mahaleb genotypes Karaj12 and Karaj05 as well as between the Karaj05 and Karaj14, all four from Karaj. Jaccard's similarity coefficient and UPGMA method were used for cluster analysis. Co-phenetic coefficient between similarity matrix and the dendrogram ($r=0.92$) showed the goodness of fit for dendrogram and the original similarity matrix. Based on respected dendrogram, the sour and sweet cherry samples were divided from mahaleb accessions at similarity level of 0.22 and produced one separate group. At the distance of 0.65 on the dendrogram, mahaleb accessions divided into 9 groups. Grouping of mahaleb samples at the most instances was in good accordance with their origin place of collection. This reflects the close genetic background of samples from one place. Although, some unexplainable similarity or distance cases were also observed. Mahaleb samples from Kermanshah had higher distribution in the branches of the dendrogram, reflecting the higher diversity of samples from this region. This study showed usefulness of RAPD markers for genetic diversity study in mahaleb genotypes.

S12.227

Molecular Characterization and Genetic Diversity Assessment of Some Romanian Grapevine Varieties

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The genetic diversity/relatedness among Romanian grapevine cultivars has been poorly investigated at the molecular level. Two molecular markers systems RAPD (random amplified polymorphic DNA) and SSR (simple sequence repeats) were employed for identification, genetic diversity and stability analysis of some autochthonous Romanian grapevine varieties. Sixty ten-mer primers were surveyed out of which 15 were selected for further analysis on the basis of their ability of producing stable, reproducible and highly polymorphic markers. To initiate the characterization of the Romanian grapevine genepool we have genotyped the grapevine cultivars at 9 microsatellite loci (VVS2, VVMD27, VVMD24, VVMD28, VMC5G6.1, VMC8G9, UDV117, UDV125, VVIP31) previously developed for grapevine. The SSR markers proved to be highly informative in all cultivars and therefore constitute a useful set for the genetic characterization of Romanian grapevine cultivars. Both techniques have demonstrated their utility in studying genetic relationships between the grapevine varieties analyzed allowing the identification and discrimination of all tested cultivars.

S12.228

Genetic Diversity of Tunisian Apricot Germplasm Assessed by AFLP and SSR Markers

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Apricot (*Prunus armeniaca* L.) species, already known at the roman age, belongs to the set of fruit tree heritage in Tunisia. The repartition of the germplasm was located throughout the country. A representative set of 81 accessions issued from the different areas of cultivation has been collected and characterized through both AFLP and SSR markers. A large genetic variability was evidenced with discrimination of all the studied accessions and identification of specific fingerprint to each accession. A total of 339 polymorphic markers were obtained (135 for SSRs and 204 for AFLPs). Neighbour joining dendrograms were established with each of the data matrices (AFLP and SSR ones) and with combined data set. Results evidenced the conservation of identified subgroups with clustering correlated to the geographical origin of the studied material. Thus, a clear distinction of the North and the oasis regions was revealed, while the accessions of the centre and the South were clustered in the same subgroup. Their phylogeographic relationship has been then discussed.

S12.229

Bayesian Model Clustering Analysis of Apricot in the Mediterranean Basin

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Apricot (*Prunus armeniaca* L.) was considered as an important fruit species espe-

cially in the Mediterranean Basin which enclosed the largest part of worldwide apricot production (nearly 80% according to FAO source). To estimate the genetic structure of apricot species in this Region, a representative sample of 471 accessions was collected throughout different Mediterranean countries from the different national germplasm in Algeria, France, Italy, Morocco, Tunisia, Turkey, and Spain. Their molecular characterization has been accessed through 25 SSR markers chosen according to their high level of polymorphism, their transportability between *Prunus* species, and localization on the whole *Prunus* genome. The statistical treatment of the dataset has been performed by using a Bayesian model clustering analysis. It allowed the assignation of the studied accessions into distinct genetic subpopulations related to their geographical origin. A gradient of decreasing genetic diversity from the East to the West of the Mediterranean area has been clearly evidenced, with an interesting new pattern based on the discrimination of 'North-Africa' subpopulation evidenced for the first time by several specific alleles. In addition, genetic variability parameters showed that 'Irano-Caucasian' and 'Adaptative Diversification' subpopulations explained the higher variance within group. Data are discussed in the context of species domestication process in this important region of apricot culture.

S12.230

cDNA-AFLP for Gene Expression Analysis: *Colletotrichum acutatum*-*Olea europaea* L. Interaction

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Anthraxnose in Portuguese olive orchards is mainly due to *Colletotrichum acutatum* pathotypes causing yield and quality losses. A differential gene expression study between *C. acutatum* and olive, in susceptible and resistant *Olea europaea* L. cultivars has not been performed so far. cDNA Amplified Fragment Length Polymorphism (cDNA-AFLP) technique can be used to profile gene expression following a special treatment or during a particular biological process. The aim of the study was to identify expression patterns of olive-*C. acutatum* pathogen interaction pathosystem. Three olive cultivars 'Galega' (susceptible), 'Cobrançosa' (moderately-resistant) and 'Picual' (resistant) were selected, and based on their known resistance to *C. acutatum* infection were used to understand the functional genomics bases under pathogen infection. Thus, selective amplifications with 100 *Mse*I/*Eco*RI primer combinations were analysed, allowing the visualization of about 5000 differentially expressed transcript derived fragments (TDFs) in 'Galega' (susceptible), 'Cobrançosa' (moderately-tolerant), and 'Picual' (tolerant) cultivars. 60 TDFs produced reliable sequences after sequencing. Important similarities with defense-related genes (e.g. lysin motif-type receptor-like kinases: LysM RLKs), was observed. However, other sequences had unknown functions through BLAST searching the GenBank database. These results were expected due to the fact that the olive genome has not been sequenced. Further studies are being conducted in order to understand which may constitute candidate genes.

S12.231

Genetic Diversity of Germplasm Resources of Litchi and Longan by SSR Analysis

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Twenty-seven pairs of SSR primers developed from litchi were applied to detect the genetic diversity of 60 accessions of litchi and longan from Guangdong, Guangxi, Fujian, Hainan, Yunnan and Sichuan province of China. These accessions included 47 litchi main cultivars or rare resources (*Litchi chinensis* Sonn), 1 longli (*Dimocarpus confinis* How et Ho), 2 artificial intergeneric hybrids (litchi×longan) and 10 longan cultivars (*Dimocarpus longana* Lour.). The 27 pairs of SSR primers

generated 447 polymorphic *loci* in all accessions, with an average of 16.6 *loci* per primer pair. Except for one extremely close affinity material, these SSR markers could distinguish the remained 59 accessions each other. The cluster analysis by NTSYS-2.10 software indicated that the similarity coefficients ranged from 0.14 to 1.00, suggesting a high genetic diversity among the accessions. Furthermore, some germplasm resources with very close relationships, such as 'Sanyuehong' and 'Jiaohesanyuehong', 'Baipixiaonuo' and 'Hongpidanuo', 'Heiye' and 'Jiangshalan', could also be distinguished clearly by these SSR markers. According to the dendrogram, 60 accessions could be classified into three groups at 0.59 similarity coefficient. The first group included all litchi accessions and 2 artificial intergeneric hybrids. The second group included all longan cultivars. The third group only included 'longli', a tentative natural hybrid of longan and litchi genus. This result was completely coincident with the traditional taxonomy. In addition, the classification of each sub-group under the first and second group was consistent with their fruit maturation period.

S12.232

Cultivar Identification in Cauliflower with Molecular Markers

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Cauliflower (*Brassica oleracea* L. var. *botrytis* L.) is one of the most popular vegetable crops worldwide. In this study, three molecular marker systems including RAPD, ISSR and SRAP were employed to identify cultivar and the relationship of 29 cauliflower cultivars and one broccoli and cabbage cultivar, respectively, and construct the fingerprint of these cultivars. The results suggested that nine RAPD primers amplified 98 distinct bands in the 32 cultivars and the polymorphic rate was 78.57%; 10 ISSR primers amplified 91 bands and the polymorphic rate was 63.74%; seven SRAP primers amplified 112 bands and the polymorphic rate was 78.57%. These results showed that higher polymorphism existed in 32 cultivars. Of all the primers, the primer me1/em4 produced 17 polymorphic bands and could distinguish 24 cultivars, while the primer me8/em8 produced 17 polymorphic bands and could distinguish 27 cultivars. 32 cultivars could be distinguished with these two primers. Cluster analysis and relatively genetic similarity coefficient indicated that the genetic relationship of these cauliflower cultivars was tightly associated with its origin and its characters. The study showed RAPD, ISSR and SRAP markers were powerful for cultivar identification and genetic diversity analysis in cauliflower.

S12.233

Random Amplified Polymorphic DNA Markers and Morphological Diversity among Four Populations of *Juglans regia* in Iran

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Genetic variation and structure of four natural populations of *Juglans regia* (family *Juglandaceae*) from Iran were assessed using random amplified polymorphic DNA markers and morphological characters. For RAPD analysis, 14 primers generated 180 discernible and reproducible bands across the four populations analyzed, out of which 174 (96.6%) were polymorphic. The Resolving power (Rp) varied with population, ranging from 2.17 to 8.14, with an average value of 5.44. Most of the genetic variation was found within population although pair-wise FST values and Bartlett's test indicated significant differentiation among populations. The variance components of within and between sampled locations detected with AMOVA were 90.52% and 9.48% of the total variance, respectively, which were both significant ($p < 0.001$). The results for *J. regia* are generally in accordance with those already recorded for other out crossing tropical tree species. For morphology analysis, the first PCs represented a total variance of 23.30% while the second and third PCs

contributed each to total variances of 13.72 and 11.40%, respectively. However, a dispersion plot of all the genotypes defined by the first and two principal components did not show any significant grouping. While the correlation between the Euclidean distance matrices based on morphological and RAPD data was low and non-significant ($r = 0.324$). The information obtained here could be valuable for devising strategies for conservation and breeding of *J. regia*.

S12.234

AFLP Analysis on the Plant-Type Mutants and Original Strain of *Salvia splendens*

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Plant-type mutants BN35-1 of *Salvia splendens* have excessive branches. They become ball-plant type without disbud when maturity. During seeding, axillary buds on cotyledon were bourgeoning and elongating. Then, The lateral branches of first and second order was elongating from the bottom of stem to top. At last, lateral branches of third order on the BN35-1 grew out and elongated. Leaves and branches were luxuriant. However, original strain BN35 have few branches, only axillary buds on 1-3 nodes under the main flower were bourgeoning and elongation growing. At last, BN35 possessed of lateral branches of second order, and leaves and branches were thin. In the paper, plant-type mutants BN35-1, original strain BN35 and 4 cultivars of *S. splendens* were studied using AFLP (amplified fragment length polymorphism) technique. 6 pairs of primer combinations were screened from 21 pairs. The results showed that BN35-1 and BN35 have a higher similarity, the similarity coefficient is 0.9861. 12 pairs of primers from 21 pairs amplified 49 polymorphic bands between BN35-1 and BN35. All of polymorphic bands should be treated as candidate gene segments which control the plant-type mutation or linkage with it. Study remains to be further.

S12.235

AFLP Analysis of the Phylogenetic Relationship of *Salvia* spp. in Beijing

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Twenty-third collections (belongs to 7 species) of *Salvia* spp. in Beijing were analyzed phylogenetic relationship by AFLP (Amplified Fragment Length Polymorphism) technique. 6 primer pairs with good repetition and highly polymorphic bands from 21 pairs were selected to amplify the genomic DNA. Total of 367 AFLP bands were obtained, in which 359 (97.82%) were polymorphic markers. At a genetic distance of 0.32, the collections could be clustered into 7 species: *S. splendens*, *S. farinacea*, *S. coccinea*, *S. plebeia*, *S. miltiorrhiza*, *S. umbratica* and *S. officinalis*. The respective genetic relationship between *S. farinacea*, *S. coccinea*, *S. plebeia*, *S. miltiorrhiza*, *S. umbratica*, *S. officinalis* and *S. splendens* were further order by order. The results provided reference for hybrid breeding inside and outside species of *Salvia* spp. in Beijing.

S12.236

Portuguese *Arbutus unedo* L. Genotypes - Genetic and Geographic Clustering Using RAPD Markers

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Arbutus unedo (Ericaceae) is a small tree characteristic of Mediterranean ecosystems. The edible fruits can be consumed fresh or used to make jellies and to produce a

spirit, which is the main valuable-derived product. The species is also interesting from an ecological perspective, since it is drought tolerant and fire resistant. In spite of its economic potential the strawberry tree genetic diversity in Portugal is unknown. In the current study we aim at determining the genetic variability of a group of selected individuals spanning the range area of the country. Twenty-seven individuals, selected based on fruit production, were screened with 20 RAPDs primers. Molecular data were used to uncover genotype genetic relationship and germplasm variability for conservation purposes and future base-breeding population. The set of 20 RAPDS primers generated 124 bands that were used to evaluate pairwise genetic similarity. About half of the *loci* (71) were polymorphic and the total expected heterozygosity was 0.27 ± 0.014 . The pairwise matrix computed with the Lynch (1990) similarity index was used to make an UPGMA dendrogram, and the tree topology was tested against the correspondent cophenetic matrix through a Mantel test. This test showed a moderate correlation yet significant ($r = 0.64$; $P < 0.001$). The cluster analysis revealed a similarity among trees up to 83%. The genotypes did not group according to their geographical origin. Indeed, no association was found between similarity and geographical distances, with a normalized Mantel computed statistic ($r = 0.01$; $P < 0.57$). The pollination system, partially based on small insects with reduced mobility, thus limiting gene flow, is one possible explanation for these results. The gathered data confirmed the suitability of RAPDs to describe genetic relationships and to fingerprint this selected group of individuals.

S12.237

Genetic Relationships among Some Italian and Spanish Traditional Tomato Cultivars as Revealed by (GATA) 4 Markers

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Spain and Italy were the first European countries where the tomato gained commercial importance. During more than two centuries, coincident with the spread of the tomato in Europe, Naples was governed by Spanish Kings, and was connected with the Iberian peninsula through important commercial relationships. This period of common history, together with several apparent morphological similarities among some traditional varieties from Southern-Spain ('de la Pera'; 'Muchamiel') and traditional varieties originated in the Campania Region, whose capital is Naples ('San Marzano' and 'Sorrento') led us to evaluate possible relationships among these Italian and Spanish traditional cultivars. In this work, we evaluated a set of 26 accessions of the traditional cultivars types 'Muchamiel' and the 'de la Pera' from the South-eastern of Spain and 'Sorrento' and 'San Marzano' from the South of Italy, using a (GATA) 4 oligonucleotide probe (Rao *et al.*, 2006, Plant Breeding 125:173-176). The number of DNA fragments found in the hybridization pattern of 'Muchamiel' and 'de la Pera' accessions was higher than that obtained in the accessions of 'Sorrento' and 'San Marzano'. In addition, the genetic variability among the accessions of 'Muchamiel' proved to be higher than the variability found among the accessions of 'de la pera'. Similarly, 'Sorrento' accessions showed larger variability than 'San Marzano' accessions. This is the possible consequence of a selection operated by Italian farmers of 'San Marzano' and 'Sorrento' accessions two very famous traditional varieties most appreciated on the market. The evidenced DNA polymorphism was used to evaluate the genetic distances among the analyzed samples and the dendrogram obtained showed a good clustering of the accessions belonging to the respective traditional variety. Our data confirm that the polymorphism of GATA fragment is a useful tool to highlight genetic relationships between traditional tomato varieties.

S12.238

AFLP Analysis of Genetic Diversity in Hazelnut Genotypes from Portugal

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Corylus avellana (hazelnut) is a deciduous, wind-pollinated monoecious tree or shrub. It's the fourth most economically important tree nut crop worldwide, following almond, walnut, and cashew. Genetic diversity of 31 Portuguese hazel trees (18 landraces from the North inner, North littoral and centre regions, as well as 13 wild genotypes), four commercial varieties and one Turkish hazel tree were evaluated by AFLP. For this purpose 7 primer combinations were used which produced 173 AFLP fragments. A total of 135 of the bands showed polymorphism (percentage of polymorphic bands =78%) and mean expected heterozygosity was 0.121. AMOVA results indicated that about 61% of variation in the data set was from genotypic variations within populations and the remaining 39% to differences among populations indicating some degree of population structure. The dendrogram generated with AFLP data from cluster analysis using UPGMA showed two main groups, one including eleven of the thirteen wild genotypes and one landrace from center. The Turkish hazelnut is the only member of one of the clusters. The PCA was in agreement with the dendrogram. A good correspondence between the clusters and the genotypes geographic origin was observed. These results illustrate the effectiveness of the AFLP technique in discriminating hazel trees. This would provide valuable information for decision making in future hazelnut breeding studies as well as germplasm management activities.

S12.239

Identification of Microsatellite *Loci* in *Opuntia* spp. and their Characterization in Cultivars and Species

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Cactus pear is a multipurpose and highly drought-tolerant crop species. In a very unstable climate situation as it is now, genetic resources undervalued so far may become crucial to cope with unforeseen threats to agricultural production. Among best suited crops to less-favoured environments, cactus pear has become one of the most well-known examples of exotic and sometimes invasive plant species in the regions of introduction from the native highlands of Mexico. Following a long process of naturalization and inbreeding, to the rise of different genetic pools specific to each area occurred. However, the difficulty experienced in identifying different *Opuntia* accessions has hindered breeding and germplasm evaluation. Among the taxonomic confusion regarding the delineation of the various species within the *Opuntia* genus, the molecular markers technique has been recently applied to complement morphological characters in assessing genetic diversity. In the present paper we report the development of 10 microsatellites isolated from *Opuntia ficus-indica*, using an enriched library from size selected genomic DNA ligated into SNX forward/SNX reverse-linker and enriched by magnetic bead selection with biotin-labelled (CT) 13, (GT)13, (AAC)10 and (AAG)10 oligonucleotide repeats. In order to evaluate the degree of polymorphism of these markers, a panel of cultivars from the germplasm collection established at the Experimental Station of the University of Sassari has been tested. DNAs from 14 *Opuntia* species (*Opuntia basilaris*, *O. dillenii*, *O. fuscicaulis*, *O. gymnocarpa*, *O. lindheimeri*, *O. matudae* Scheinvar (xoconostle), *O. polyachanta*, *O. nastrena*, *O. robusta*, *O. soberensii*, *O. stricta*, and the species *Nopalea cochenillifera*) were also used to test transferability of the developed primers. The level of polymorphism and the relatively high number of alleles detected ranging from six to 22 suggest that these markers can be used for both inter and intra-specific studies, as well as to analyze the polyploidy occurrence in the *Opuntia* genus.

S12.240

Molecular Identification and Genetic Variation Analysis of 13 Identical Herbaceous Peony Cultivars Using SRAP Markers

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Herbaceous peony is an important ornamental plant. Many cultivars indigenous to China are apparently similar, which hindered normal trade and parent selection in designing optimal breeding strategy. In the present study, thirteen morphologically similar herbaceous peony cultivars were employed for molecular characterization and evaluation of genetic variation with the use of sequence related amplified polymorphism (SRAP) technology. The results showed that eight screened primer combinations produced 81 amplified fragments, with an average of 10.13 fragments for each primer combination, ranging in size from 50 to 500 bp. Among the total 81 fragments, 64 (84%) were polymorphic. The primer combination Me8Em9 with the highest polymorphism (100%) could not distinguish all the cultivars, but seven selected fragments produced by two primer combinations (Me8Em9 and Me4Em8) allowed differentiation among the thirteen identical cultivars. According to the result of pairwise similarity comparison, genetic similarity coefficient among the cultivars ranged from 0.52 to 0.80. Further UPGMA cluster analysis demonstrated that the molecular classification profile based on SRAP data was neither related with flower form, nor guard petal color. Instead, it showed moderate relation with color of stamen-transformed petals. In conclusion, SRAP technology was successfully used in our study. It is not only potentially powerful for larger-scale herbaceous peony cultivars identification, but also informative and useful for future breeding program of herbaceous peony.

S12.241

Genetic Diversity Analysis of Balsam Pear (*Momordica charantia* L.) by ISSR Mark

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Analysis of 30 varieties balsam pear by ISSR mark. 172 bands have been amplified by 18 primers, in which, 132 bands are polymorphism and the polymorphic proportion was 76.7%. UPGMA analysis show that: These balsam pear varieties can be divided into four categories: (1) The varieties which from Yunnan, Sichuan and Chongqing in the Southwest of China are classified as a categorie, it can be divided into two sub-categories, the first sub-categories major from Yunnan, the second sub-category, including Chongqing and Sichuan varieties; (2) The varieties which from Guangdong, Guangxi, Hainan in the Southeast of China varieties classified as a categorie, the relationship of this populatio is more complicated, but also showed a certain geographical differences; (3) The variety 105-2 which from Guangdong as a separate category, maybe it's a special variety; (4) The variety 14-2-3 which from Hainan as a separate category. The result show these balsam pear varieties classification is similar as regional distribution difference, it's embody on other species just like tobacco (Xiao *et al.*, 2007), chimonanthus praecox (Zhao *et al.*, 2008) and so on.

S12.242

Molecular Characterization of Tomato Accessions from Southern Italy

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During last few decades the use of local varieties has declined due to the introduction of contemporary cultivars characterized by higher yield and bred for im-

proved agronomic value. However, these cultivars do not always have the quality characteristics typical of older, traditional lines. There is concern that the practise of intensive agriculture and adoption of new varieties risks the loss of important alleles in germplasm adapted to specific environments. The present study reports the molecular characterization of twenty tomato accessions from the Campania region of Southern Italy, using different DNA markers: (GATA) 4 minisatellite, SSRs and SNPs. GATA hybridization patterns revealed accession specific profiles and heterogeneous material. Homogeneous accessions were further analyzed at 14 SSR and 71 SNP *loci*. Of the 14 SSRs analyzed, 13 were polymorphic and three *loci* were able to discriminate all the samples. This result demonstrated that, despite the limited genetic diversity of tomato, DNA fingerprints based on SSRs permitted discrimination of not only different cultivars, but also morphologically similar types. A lower level of polymorphism was found using SNP markers. Only 12 SNP *loci* out of 71 showed polymorphism among the local accessions. SNPs were not able to generate a unique profile for each accession analyzed. SNP polymorphism of the Italian samples was compared with SNP polymorphism detected in a tomato collection (www.tomatomap.net) that includes samples of different geographic origin and suitable for different food products. The data were analyzed with the software Structure. The results showed that the Campania accessions formed a separate group, suggesting that they are genetically different from all the other samples. Although accessions of contemporary germplasm contained more allelic diversity, the Campania accessions represent a unique sub-population. Our observations point out the importance of preserving and protecting regional germplasm as it could represent a source of important alleles.

S12.243

Evaluation of Genetic Diversity among Some Genotypes of *Poa pratensis* L. by RAPD Molecular Markers

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Genetic improvement of plants needs a high genetic diversity. *Poa pratensis* has extensive divergence in natural old turfs and pastures in temperate zones of Iran, being useful for breeding programs. Molecular markers offer a powerful tool to assess genetic diversity. In this research RAPD markers were used to study the genetic diversity of 19 turfgrass genotypes (including 17 genotypes of *Poa pratensis* along with one genotype of each of *Lolium perenne* and *Cynodon dactylon* species). After screening 100 random 10-mer RAPD primers, 17 primers were chosen based on the clarity of their amplified fragments in PCR and clear bands with high reproducibility were used for scoring. From the 284 produced bands, 281 were polymorphic. Cluster analysis of RAPD data was performed using the UPGMA method based on Jaccard's similarity coefficients. Dendrogram at similarity distance of 0.24 gave 5 main clusters. The correlation coefficient between the data matrix and the co-phenetic matrix of cluster data was high ($r = 0.99$), indicating that the clustering dendrogram is highly fit to similarity matrix. A relatively high genetic similarity was observed between some commercial cultivars and Iranian local genotypes. Also, this research showed the high genetic diversity among studied genotypes as well as the high efficiency of RAPD markers for evaluation of genetic diversity among turfgrasses.

S12.244

Genetic Diversity in Pepper (*Capsicum* spp.) by RAPD Marker

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The *Capsicum* germplasm had a great diversity for morphological traits like fruit size, shape and color. This diversity offer opportunities to develop unique cultivars

for agronomic applications. The study of genetic divergence is a useful and effective tool to screening accessions in germplasm banks and to identify superior parents in breeding programs. The objective of this work was to evaluate the diversity genetic among 29 accessions of germplasm bank of Universidade Federal de Roraima by RAPD markers. The DNA extraction was made by CTAB protocol and then submitted to amplification by RAPD analysis and separation of amplification products by agarose electrophoresis. The grouping was made by UPGMA method. The DNA was extracted of the 29 accessions and amplified by 10 primers. Eight primers (UBC-135, UBC-146, UBC-155, UBC-168, UBC-228, UBC-247, UBC-253, UBC-296) produced polymorphic bands. The minimum distance were 0,08, between AC 06 and AC 273. The Accessions were grouped in four different groups. The Group 1 was formed by accessions 73, 6, 43, 74 and 49; the group 2 was formed by accessions 8, 61, 40, 75, 45, 59, 32, 25, 58, 3, and 56; The group 3 was formed by accessions 44, 22, 20, 5, 76, 72, 55, 69, 37, 24 e 70; and the group 4 was formed by accessions 71 e 67. RAPD markers were effective in to detect the diversity genetic among 29 hot pepper accessions.

S12.245

DNA Barcoding: Unsuccessful for Species Identification in *Fragaria* L.

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The USDA-ARS National Clonal Germplasm Repository (NCGR) in Corvallis, Oregon, maintains more than 1500 of *Fragaria* accessions representing approximately 24 species collected from 37 countries. Species designation currently depends on the published species descriptions which depend on geographical location and morphological traits that exhibit limited variation. Our objective was to assess if a simple DNA-based technique like DNA barcoding could verify the identity of *Fragaria* species. Four potential barcoding regions were tested in *Fragaria*. They consisted of the nuclear ribosomal internal transcribed spacer (NrITS) and three chloroplast regions (psbA-trnH spacer; IRB11 [YCF2/ORF2280 3' to ORF 79] and IRB14 [ndhB 5' exon to rps 7, 5' end]). IRB11 and IRB14 are located in the inverted repeat region B (IRB) of the chloroplast genome. The 'barcoding gap', between within species and between species variation, was absent preventing successful identification of *Fragaria* species. Cluster analysis using NrITS supported *F. mandschurica* as the maternal donor to the octoploids. Three diploid *Fragaria* clades (X, Y and Z) were identified using NrITS, while the chloroplast psbA-trnH contained little variation. The psbA-trnH spacer could only identify *F. bucharica* and *F. nilgerrensis* due to characteristic deletions in this chloroplast region. However, none of these sequences, singly or in combination, could identify each of the *Fragaria* species. Therefore, DNA barcoding using universal sequences is not an adequate technique for species identification in *Fragaria*.

S12.246

Identification of *Hydrangeaceae* Accessions of Wild Origin from Jeju, Korea Using Molecular Markers

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This study was designed to identify unknown accessions of *Hydrangeaceae* that have leaf morphological characters different from *H. anomala* subsp. *petiolaris* (Siebold & Zucc.) E. M. McClint. and *Schizophragma hydrangeoides* Sieb. & Zucc. in Jeju, Korea. A total of 29 accessions of *Hydrangea* and *Schizophragma* from the wild were collected and analyzed using randomly amplified polymorphic DNA (RAPD) and single nucleotide polymorphisms (SNPs) from the ribulose-1, 5-bisphosphate

carboxylase/oxygenase large subunit (rbcL) gene. Morphological characteristics of leaves and shoot apices of several accessions were also examined. Based on the RAPD markers, SNPs of the (rbcL) gene, and vegetative morphological characteristics, the unknown accessions were identified as *H. anomala* var. *petiolaris*. The RAPD markers and SNP analysis effectively identified all four unknown accessions as *H. anomala* var. *petiolaris*.

S12.247

Chloroplast DNA Diversity in Portuguese Grapevine Cultivars

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The chloroplast genome (cpDNA) analysis is an excellent tool in phylogenetic and phylogeographic studies. Investigations about grapevine domestication are being carried out based on chloroplast microsatellites (cpSSR) variation in samples of *Vitis vinifera* ssp. *sylvestris* and *sativa* from the whole area of the species distribution (from Iberian Peninsula to Middle East). Polymorphisms detected suggest the existence of at least two important origins for the cultivated germplasm. The objective of this study was to assess the diversity in the chloroplast genome of a group of Portuguese cultivars, with the purpose to know their regional genetic patterns as well as to infer their putative origin. A total of 38 varieties, representative of cultivated grapevine material in the North of Portugal, and three international varieties (out-groups), were studied using three cpSSR *loci* and analysing several fragments of the Large Single Copy (LSC) region of the cpDNA by PCR-RFLP or CAPS (Cleaved Amplified Polymorphic Sequences) methodology. The three cpSSR *loci* were polymorphic, two different alleles were found in each *locus*. Allele variants of the three *loci* combined in a total of three different haplotypes. The most frequent haplotype (70.7%) was the same previously reported as the most frequent in Iberian Peninsula and Occidental Europe. One of the haplotypes was unique in Rabigato variety. This haplotype was previously proposed to be an ancestral haplotype. Inference about the progenitors of the cultivar Touriga Franca was done. This cultivar could be the progeny of Touriga Nacional and Tinta Barroca. The cpSSR analysis revealed that Tinta Barroca could have been the female progenitor. Some fragments of the LSC region from *Vitis vinifera* cpDNA were amplified and digested with two restriction enzymes (HinfI and TaqI), according to CAPS methodology. No polymorphisms were found in the 41 varieties, but this analysis will be expanded to the rest of the LSC region.

S12.248

Morphological Evaluation of Some Chamomile Genera, *Anthemis* and *Matricaria* in Iran (*Matricaria* spp, *Anthemis* spp)

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For evaluation and identification some of the chamomile species in two genera *Anthemis* and *Matricaria*, were used morphological and phonological characteristics. The experiment was conducted in a Randomized Completely Blocks Design (RCBD) with three replicates, sixteen quantitative and qualitative traits were evaluated such as height, anthodium diameter, receptacle diameter, ligulae flower number, dry flower percent, 100 flower weight, time of flower beginning, yield, flower number per plant, 1000 seeds weight, stomata length, leaf length, leaf width, flower color and type of leaf and receptacle in 26 genotypes. Analysis of variance indicated that the cultivars were significantly different for all examined traits. Principle Components analysis (PCo) analysis placed 16 traits in five principle components that covered 91 percent of variance. The first principle components covered 39 percent of variance included main traits such as anthodium diameter, receptacle

diameter, ligulae flower number, 100 flower weight, yield, 1000 seeds weight, leaf length. Cluster analysis classified genotypes to 8 main groups and 2 independent genotypes. This study showed that morphological markers are one of the reliable tools to determined genotypes and species, yet.

S12.249

An Evaluation of Tolerance of Some Bean (*Phaseolus vulgaris* L.) Genotypes to Bean Common Mosaic Virus (BCMV) Using Principle Components and Biplot Analysis

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The common bean is the most important grain legume for direct human consumption in the world and BCMV is one of the world's most serious bean diseases that can reduce yield and quality of harvested product. To determine the best tolerance index to BCMV and recognize tolerant genotypes, 2 experiments were conducted in field conditions. Twenty five common bean genotypes were sown in 2 separate RCB design with 3 replications under contamination and non-contamination conditions. On the basis of the results of indices correlations GMP, MP and HARM were determined as the most suitable tolerance indices. The results of principle components analysis indicated 2 first components totally explained 98.52% of variations among data. The first and second components were named potential yield and stress susceptible respectively. Based on the results of BCMV tolerance indices assessment and biplot analysis WA8563-4, WA8563-2 and Cardinal were the genotypes that exhibited potential seed yield under contamination and non-contamination conditions.

S12.250

The Morpho-Physiologic Analysis of Biodiversity of Melon Germplasm Cultivated in Romania

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The present paper aimed toward the accomplishment of a complex study over the physiology and biochemistry of *Cucumis melo* species. The Romanian market is not very rich in the assortment of cultivated melons. From experience, we learned that the varieties with foreign origin, although well known and appreciated for their qualitative potential in their native countries, in the climatic conditions of Romania the results were not conclusive, some developing mediocre characteristics. The main purpose of our work was a deep comparative analysis of the melon assortment belonging to the varieties: *cantalupensis*, *reticulatus* and *inodorus*. We concentrated on the physiologic and biochemical processes that occur in fruits during their growth and development. The biologic material is represented by twenty five cultivars of *Cucumis melo* specie. The main objectives of analysis, made on the fruits of cultivars from the three varieties mentioned above, were the determination of the following characteristics: - resistance to transport and storage (through the determination of respiratory intensity, content of endogen ethylene, flesh conductivity, total dry content and water content); - content in sugar and titrable acidity; - content in carotene; - content in ascorbic acid; - content in proteins; - content in flavones; - aroma.

S12.251

Relation of Garlic Morphotype and Distribution of Viruses in the Czech Germplasm Collection

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The Czech collection of garlic genetic resources (648 accessions) was divided into the three main morphological groups according to the ability to produce the scape:

bolting (with the scape; 327 genotypes), semi-bolting (108 genotypes) and non-bolting garlic (213 genotypes). All accessions were surveyed for the presence of four different viruses, e.g. Onion yellow dwarf virus, Leek yellow stripe virus, Garlic common latent virus, and Shallot latent virus, by DAS-ELISA. These four viruses are widespread in the collection with an incidence ranging from 65 to 83% for genotypes and 39 to 61% for plants and most of the genotypes tested were infected with two to four viruses. The differences between individual viruses infection in three garlic morphological groups were studied. Statistically significant differences were not found within the virus infection of three morphological groups but some relations are obvious. For instance very rare infection of LYSV (35%) was ascertained in semi-bolting garlic compare other two groups (59% in non-bolting and 62% in bolting) and/or very often infection of SLV was found in bolting garlic (93%) compare to others (59% in non-bolting and 50% in semi-bolting garlic). Also the frequency of virus combinations was different in individual morphotype groups.

S12.252

Production and Identification of F1 Hybrids between Rape-Radish Addition Line and *Brassica oleracea*

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In order to transfer radish chromosome d carrying nematode-resistance into *Brassica oleracea*, sexual hybridization was carried out between rape-radish d addition line ($2n=40$) as chr.d donor and cabbage as the recipient ($2n=18$). F1 hybrids were obtained by means of embryo rescue. They were identified by morphological investigation, cytological and molecular marker technique. The result indicated that all F1 plants had the same morphological characters, partly resembled female parent, partly liked male parent. Cytological observation of somatic cells showed that the F1 contained 29 chromosomes. The presence of chromosome d in F1 plants was tested by the chr. d special marker OPF07-363bp which we have developed. As a result, all F1 plants presented this chr.d special band. Additionally, three pairs of SSR primers were used to identify the heterozygosity of F1 plants, and all results showed the F1 are hybrid. Therefore, it could be concluded that the F1 plants were true hybrids crossed by rape-radish d addition line and cabbage, and chromosome d could be transferred into cabbage by sexual hybridization.

S12.253

Identification Organoleptic and Functional Quality Profiles in Spanish Traditional Varieties of Tomato

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Despite the increasing importance of internal quality in breeding programmes and marketing of tomato, little information is available regarding organoleptic and functional profiles of traditional varieties of renowned quality. This is the objective of this work, consisting in the evaluation of internal quality of 51 traditional tomato accessions representative of the Spanish variability. Contents of total soluble solids, oxalic, malic, citric and glutamic acids, fructose, glucose and sucrose, vitamin C and lycopene were determined, obtaining the respective organoleptic and functional profiles. These profiles will be very valuable to establish breeding objectives, as these varieties are considerably appreciated by consumers, who are willing to pay higher prices for them. A considerably high level of variability has been found in the profiles obtained and no clear groups could be identified considering fruit morphology or local name. Variability was higher in traits affecting functional quality (coefficients of variation of 51.2% for vitamin C and 74.6% for lycopene content) than those affecting organoleptic quality (coefficients of variation ranging from 18% for total soluble contents to 38.8% for glutamic acid). Additionally, several accessions could be selected for their higher individual contents for further studies of internal quality. It is the case of accessions CDP08102 and CDP03547 for their high malic content, accession

CDP06315 for high fructose and glucose contents, accession CDP01523 for its lycopene content and accessions CDP02226 and CDP00336 for vitamin C content. Considering previous correlations among individual contents and consumer preference accessions CDP07554, CDP02666 and CDP03547 should be further evaluated for their overall flavour quality.

S12.254

Characterization of Spanish Genetic Resources of *Arachis hypogaea* L.

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Despite the importance of groundnut cultivation in Spain during the XIX and early XX centuries very little of this diversity is represented in genebanks and the degree of genetic erosion is extremely high. During the last three years a Spanish collection of groundnuts has been established, mainly from accessions collected on the east coast of Spain and the Balearic islands, the main producing areas at the height of groundnut cultivation. This work presents the results of the characterization of this germplasm. The collection, that included controls obtained from the USDA-NGRP, represented four of the five varietal types described in Spanish literature: Collaret, Cacahua, Moruno (from Valencia) and Palma (from Balearic Islands). Collaret and Cacahua made up most part of the collection as they can still be found in cultivation in the areas surveyed. The typification of both types coincides with earlier descriptions. A considerable degree of variation was found in pod and seed descriptors. Collaret is characterized by the small pod size, low number of seeds and deep pod constriction. Cacahua is characterized by big pods with slight or almost absent constriction and 3 to 4 seeds per pod. In both types, two subgroups could be identified considering vegetative traits. Palma and Moruno types were originally represented only by USDA controls. Moruno type was characterized by intermediate pod size and moderate seed constriction. One of the recently collected accessions with an ambiguous local name could be reclassified as this type with a PCA analysis. Palma type was grouped in the PCA analysis with the Collaret type, suggesting that it could be represent an introduction to the Balearic Islands from Valencia. Other accessions of the Balearic Islands corresponded to the Cacahua type and were closely related.

S12.255

Evaluation of Genetic Diversity in Pepper (*Capsicum* spp.) Genotypes Using Quantitative Traits

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Pepper is being used as a food flavoring, a coloring agent, a pharmaceutical ingredient and in other innovative ways. For having useful information about morphological traits for breeding of pepper genotypes, a precise determination and discrimination of the genotypes is required. In this study genetic diversity of 77 pepper genotypes collected from Gene Bank of Plant Improvement Institute and also from various regions of Iran and a few countries was investigated by using fourteen morphological traits in augmented design with three controls. Measuring of traits of pepper genotypes were performed based on IPGRI descriptor. Statistical calculations included correlation analysis, factor analysis and cluster analysis. Correlation analysis of traits revealed that the greatest positive correlation (0.863**) observed was between fruit width and fruit dry weight. Factor analysis was used to determine the number of main factors and results showed that 4 independent and major factors explained 71.2% of total variance. With using ward method for cluster analysis, genotypes in distance of 8 were divided into six sub-clusters, but we didn't find any relationship between this clusters and geographical distribution. This study showed that the evaluated genotypes had considerable genetic variation and morphological traits can be significant tools for classification and separation of pepper genotypes.

S12.256

Wild and Cultivated Tomato Fruits Characterization for Its Internal Quality

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The development of high organoleptic quality and functional value vegetables is becoming an increasingly important breeding objective. In the case of tomato reducing sugars, organic acids, vitamins and carotenoids play an important role in the definition of these characteristics. In this work a collection of cultivated and wild germplasm has been evaluated for sucrose, fructose, glucose, oxalic, malic, citric and glutamic acids, L-ascorbic acid, beta-carotene and lycopene. A total of 36 accessions have been evaluated, 10 of them during two years in order to evaluate environmental effects. A high variability has been observed between accessions in all the characters analyzed, both for organoleptic quality (coefficients of variation ranging from 27% for total soluble contents to 93% for glutamic acid) and functional quality (coefficients of variation higher than 100% for ascorbic acid and 72% for lycopene). Several accessions have been identified for its high content in individual compounds and could be an interesting source of variation for breeding programmes. Accessions CDP00237 of *Solanum pimpinellifolium* and CDP06889 of *S. lycopersicum* showed high values of ascorbic acid. Accessions CDP05226 and CDP07852 of *Solanum pimpinellifolium* showed high values of lycopene (20-30% higher than LA-3538 carrying hp1 gene). Accession CDP06673 of *S. pennelli* showed high content in sucrose. These accessions can be used in future breeding programs to improve internal quality of cultivated tomato.

S12.257

Germplasm Collections of Old Lettuce Cultivars and Landraces for Modern Breeding

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Lettuce (*Lactuca sativa* L., family *Asteraceae*) is the most important leafy vegetable species; about 900 of accessions are kept in the National germplasm collection of the Czech Republic since 1951. Repeated multiplications could lead to a genotypic shift of accessions and these changes can be manifested phenotypically. Old cultivars and landraces can be important donors of resistance factors to actual spectra of pathogens and their races. The set of 30 lettuce accessions (landraces and cultivars bred and/or traditionally grown on the territory of the Czech Lands) provided from the Czech national germplasm collection. Plants grown in the field were assessed for 22 traits (19 morphologic traits and 3 traits for developmental stages). Actual description of 23 accessions was similar to data from literature sources. Five accessions showed significant differences when compared actual data to literature information. These differences are fixed genetically. Original description of one accession was not found. One accession was completely heterogeneous in phenotypic expression of traits. This heterogeneity is probably caused by crossing with another accession(s) during regeneration. Identical set of 30 lettuce accessions was evaluated for response to 15 isolates (races) of lettuce downy mildew (*Bremia lactucae*) by *in vitro* tests in growth chamber. These isolates (races) are actually highly virulent largely distributed throughout Europe. Totally 21 accessions were either susceptible to isolates, either incompletely resistant, either developed heterogeneous reaction to some isolates. Remaining nine accessions were resistant at least to one of isolates tested. Race-specific resistance genes to *B. lactucae* were detected also in old cultivars of lettuce, same-named accessions expressed different reaction to *B. lactucae* isolate(s).

S12.258

Analysis of the Population Structure of the Wild Tomato Species *Solanum pimpinellifolium* in the Germplasm Collection of AVRDC - The World Vegetable Center

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The currant tomato (*Solanum pimpinellifolium*) is a close wild relative of cultivated tomato. It is a valuable source of genes for biotic and abiotic stress tolerance, nutritional quality, and yield traits in the research and breeding portfolio of AVRDC - The World Vegetable Center. Understanding the population structure is a prerequisite to fully exploit the potential of this species. The Center's *S. pimpinellifolium* collection consists of 321 accessions. For this population structure analysis, 192 accessions representing 15 countries were selected for study. Eighty-five percent of the selected accessions originated from South America (Peru, Ecuador) and Mesoamerica (Mexico); the former countries are considered the center of origin and genetic diversity of the tomato crop. These accessions were genotyped at 48 SSR *loci*, evenly distributed on the 12 chromosomes with an average distance of ~20cM. The genotypic data were analyzed using Bayesian analysis (STRUCTURE software) and hierarchical clustering analysis (DARwin) to elucidate the population structure. Both the Bayesian and hierarchical clustering analyses revealed the presence of four major subpopulations. As expected in a wild species with minimal human intervention, the constitution of these four subpopulations largely corresponded with the center of origin of the accessions. Genotypes grouped as follows: 87.3% of the genotypes from Peru grouped into two distinct clusters; 71.9% of the genotypes from Ecuador grouped into another cluster; and 96.7% of the genotypes from Mexico grouped into the fourth cluster. There were five Peruvian accessions in the Ecuadorian cluster and 10 Peruvian plus eight Ecuadorian accessions in the Mexican cluster. This data suggests a shared history of evolution of the species, firstly between Peruvian and Ecuadorian accessions and then between Peruvian, Ecuadorian, and Mexican accessions. This suggests that the species might have originated first in Peru and then migrated northwards to Ecuador and thereafter to Mexico.

S12.259

Isozyme Analysis of Tomato Hybrids and Parents on Leaf Curl Virus

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Tomato is affected by several pests limiting successful cultivation and yield loss. Tomato leaf curl virus (TLCV), disease affect plant growth and yield losses up to 100 per cent (Picco *et.al.*, 1998). The disease is confined to tropical, sub tropical Asian countries, Australia parts of Africa Middle East, South West Europe and the Caribbean Islands. use of insecticides increases cost of cultivation and residual effects which creates pollution hazard on non-targeted organisms, and health risk to consumers. Cultural practices to control TLCV must be combined with use of tolerant or resistant tomato varieties (Picco *et.al.*, 1998). Nematode incidence in tomato causes about 100 billion euro-losses (Sesser *et.at.*1981). Root knot nematode infection causes of typical root galls affect nutrients uptake and translocation of materials. Use of nematicides and soil fumigants, are expensive, labour intensive and creates ecological hazards. Development of resistant/tolerant cultivars would have proven commercially successful to control nematode incidence. Resistance of root knot nematode is degenerated due to changes in pathogenic variability and environment. The present study on Peroxidase and poly phenol oxidase isozyme analysis which confer resistance to TLCV and nematode incidence was carried-out with two hybrids CLN2123AXHN2 and HN2XCLN2123A and their parents CLN2123A and HN2 along with TLCV resistant check (Lakxmi), nematode resistant check (HisarLalit) and susceptible check CO.3. There was uniform appearance

of PO2 isoform in all the genotypes evaluated except CO.3 and more number of isoforms were observed in both direct as well as reciprocal cross. The polyphenol oxidase isozyme analysis revealed that susceptible check CO.3 exhibited two faint bands whereas two tomato hybrids recorded six isoforms at 96 hours after grafting.

S12.260

Aspects of Growth Behaviour and Evaluation of DMY of *Medicago arborea* under Different Management Utilisations for Pasture Improvement in Mediterranean Environment

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The paper shortly describes some biological and morphological characteristics of *Medicago arborea* (tree medic) a member of the family *Leguminosae* or *Fabaceae* and its biomass potential as forage plant for pasture improvement under Mediterranean conditions. In particular researches on the relation between seed germination and moisture content of the seed; DM evolution of the plant organs during the growing season (October-May); DMY/plant under real sheep grazing management, are reported.

S12.261

Classification and Identification of Bunching Onion (*Allium fistulosum*) Varieties Based on SSR Markers

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DNA markers are useful tools for phylogenetic analysis, variety identification and plant breeding. Bunching onion (*Allium fistulosum* L.) is an allogamous crop, and therefore has a high degree of genetic heterogeneity within each variety. We investigated DNA polymorphisms in 30 bunching onion varieties using 33 simple sequence repeat (SSR) markers. Cluster analysis based on allele frequency data at 29 SSR *loci* could classify most of the varieties into the predicted variety group, that is, 'Kaga', 'Senju' or 'Kujo'. This classification, based on SSR markers, was consistent with the morphological grouping. However, high genetic variability was observed within each variety as well as among varieties. It was considered impossible to determine the proper genotype for any bunching onion variety, and therefore, it appeared difficult to determine that an accurate genotypic identity for varieties can be determined using only DNA markers. Nevertheless, an assignment test was successful at correctly assigning 89% of individuals to their original variety. These results indicate that SSR markers are effective markers for variety identification in bunching onion.

S12.262

Chlorophyll Degradation in Varieties of Paprika Pepper

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The varieties of pepper for paprika can be classified into two groups according to their evolving process of chlorophyll degradation during fruit ripening. In the wild-type red varieties chlorophyll catabolism is concomitant with the start of carotenogenesis, whereas in the stay-green mutant varieties the chlorophylls coexist with that process, resulting in varieties of a characteristic brown color. Nevertheless, in some stay-green varieties, the fruit becomes completely red in the over-ripe fruit stage, with an increasing in carotenoids. The aim of the present study is to

investigate the behavior, in terms of evolution of chlorophylls in fruits, of seven varieties selected and bred by I.M.I.D.A. Horticulture Department: the wild-type red varieties Costal, Bola, Datler and Rodacol and the stay-green varieties, with the recessive gene *cl* (chlorophyll retainer) Negral, Datler Negro and Nedacol. The fruits have been classified in 8 ripening stages. In each of the stages we have studied the following parameters: total chlorophyll content, chlorophyll a, chlorophyll b and the chlorophyll a/b ratio. The results were as follows: 1. In the stay-green varieties there is not chlorophyll degradation at the beginning of ripening. A chocolate brown color of the ripe fruit results from lack of chlorophyll degradation and the simultaneous accumulation of red pigments. 2. In some stay-green varieties, such as Datler Negro, the chlorophyll values in over-ripe fruits range from 528.26 mg / kg at the beginning of maturation to 222.68 mg / kg in the over-ripe stage. Therefore, the route of catabolism of chlorophyll is not completely inhibited but delayed. 3. In the varieties Negral and Nedacol there are decreases in total chlorophyll content until stage 7, although an increase of chlorophyll b, from this stage, causes a higher content of total chlorophylls in the stage 8 (over-ripe).

S12.263

Qualitative Analysis of Volatile Compounds in Local Landraces of Tomato Cultivated in South Italy

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Volatile components extracted by solid phase micro extraction (SPME) from the full ripe fruits of 28 landraces of a local variety of Sicilian tomato (*Lycopersicon esculentum* Mill.) were analysed by gas chromatography-flame ionization detector (GC-FID) and gas chromatography-mass spectrometry (GC-MS). On the whole between 15-20 components have been fully characterized. However, all the aromatic profiles were characterized by the predominance of hexanol, hexanal, cis-3-hexenal, cis-2-hexenal, 2-methyl-5-hepten-2-one, 2-isobutylthiazole, together with several minor components. Significant differences among the several analysed samples have been detected, allowing to hypothesize the use of the different aromatic profiles as biological marker as a further parameter for the characterization and differentiation of this particular variety of Sicilian tomato.

S12.264

Characterization of the Local Variety 'Torquemada Pepper'

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In the Torquemada (Palencia) area there is a pepper growing tradition with a variety of thick pulp, highly appreciated as a seasonal product, usually eaten fresh. The crop is kept practically with traditional techniques. The seed obtained by the farmer, chosen from the fruits considered to be those that best respond to the characteristics of the variety, is used. Promoting the crop, seeking to attain quality products, starts with the understanding of the characteristics of the variety. In 2006 a study was initiated aiming at characterizing morphologically this variety and study its variability. Twelve seed samples were taken among the cultivators. The evaluation of these samples, along with 3 commercial varieties, has been done in a lab and in the field. The seed characteristics have been analyzed in the lab. The characters of the plant, the flower and the fruit have been evaluated in the field. The morphological characterization has been done following the guidelines of the *Capsicum* spp. descriptors of the IPGRI-FAO (International Plant Genetic Resources Institute). The statistical process of the results was done with the ANOVA test, using the SAS statistical package (V8). It was possible to observe differences in some characters of the plant, the inflorescence and the fruit. The more distinctive characteristics shown are the following: it's dark red when it reaches ripeness; very large size, with an average weight of 365 g; the longitudinal section is squared, with an average length and width of 10 and 9 cm respectively; very fleshy, the pulp has a thickness of around 8 mm, and sweet taste; with four very marked locules and a deep set peduncle.

S12.265

Embryo Rescue - A Useful Method for Obtaining Interspecific *Allium cepa* x *Allium roylei* Hybrids

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The bulb onion is one of the most important edible *Allium* crop. Unfortunately, *Allium cepa* is completely susceptible to downy mildew. Only *Allium roylei*, a wild species originating from northwest India is completely resistant to the infection caused by *Peronospora destructor*. With the aim of introgression to the bulb onion resistance to the pathogen, interspecific F1 *A. cepa* x *A. roylei* plants were obtained through *in vitro* cultures, via embryo rescue technique. Embryo rescue method is mostly used when proper seed setting is not possible, especially for plants obtained from interspecific crosses. The experiment was carried out using eight different accessions of the bulb onion and two ecotypes of *Allium roylei*. After pollination of *Allium cepa* plants with *Allium roylei* pollen, almost 36.000 ovules were placed on regeneration media, from which 1768 (4.9%) embryos started to develop into F1 plants. As a result of application of embryo rescue technique, after acclimatization to the *ex vitro* conditions, 906 F1 *Allium cepa* x *Allium roylei* plants were obtained. Verification of the hybrid character of the F1 regenerants was based on molecular markers. After PCR reaction with SIR-F/SIR-R primers (based on AY753557 onion cDNA clone), products of amplification were resolved in 1% agarose gel. Molecular analysis revealed that 97.6% of the examined regenerants were true F1 *A. cepa* x *A. roylei* hybrids.

S12.266

Morphological and Commercial Characterization of Lettuce Varieties from the Seed Bank in Amayuelas de Abajo (Palencia)

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The serious problem of genetic erosion that is nowadays taking place throughout the world also affects our territory, which is a centre for diversity for certain horticultural species. There is a seed bank collection in Amayuelas de Abajo (Palencia) which is part of the seed bank network of Castilla-León. The seeds supplied by farmers need to be characterised through a card for its identification and conservation. Different institutions and local action groups working on the recovery of landraces from every region in the central area are intended to be involved in this project. The study focused on the morphological and commercial characterization of lettuce varieties. 18 varieties were selected randomly. A total of 432 lettuce plants were planted at a density of 5.3 plants/m². Trials were conducted in the High Technical School of Agricultural Engineering (ETSIIAA) located in Avenida de Madrid 57, Palencia (España). From the results obtained we can conclude that the Capitata varieties present better quality than the Longifolias, considering the qualitative and quantitative point of view as well as the general behaviour of the crop. None of the Longifolia varieties was found to be very good and only a few can be marketed. Among the Longifolias there are varieties that have been assessed as bad due to susceptibility to diseases. We need to carry out more trials in order to be able to verify these first observations

S12.267

Nutritional Value of Local Italian Genotypes of Onion, Broad Bean, Artichoke, Tomato and Red and Yellow Peppers

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Biodiversity is the result of a very long evolution processes. Within its huge diversity, it represents a vital reserve for humanity and its preservation is an essential con-

dition for the survival of the whole "ecosystem". The globalization of market has induced the big seed supplier to select spreading stuff able to adapt to every kind of agricultural environments, and gave plant with high productivity, uniformity and long shelf-life, even though very often it doesn't reach any nutritional or organoleptic good level, in a way to penalize the "local" product. The re-establishment of the local ecotypes, duly revised, selected (according to agricultural needs as resistance and tolerance to pathogenic, etc.), reintroduced and produced in the former specific area, could represent a new thrust to the whole local production process. Local ecotype, besides, could face with resolution about the competition of imported products, focus its strength points on "locality", "seasonality" and "sensorial quality". Fruits and vegetable are also know for their health-promoting effects against oxidant compounds. The research program supported by Marche Region (Italy) has the main goal to identify, characterize and valorize regional Italian genotypes of onion, artichoke, broad bean, tomato and peppers. Agronomic studies have been performed to identify lines with higher productivity and quality among lines collected from the local germplasm. A total of 18 vegetable samples, including 4 broad bean, 7 peppers, 4 tomatoes, 1 onion and 2 artichoke were analyzed for their quality (Soluble Solids Content, Total Acidity) and nutritional parameters (Total Antioxidant Capacity, measured by Trolox Equivalent Antioxidant Capacity method). Results evidenced an interesting variability among the different genotypes in particular regarding their solid soluble contents and total antioxidant capacity.

S12.268

The Distribution of Male Sterile Cytoplasm in Garden and Leaf Beet Germplasm Resources

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Cytoplasmic male sterility (CMS) is used to produce hybrids of both garden and sugar beets. Hybrid seed production relies upon a single source of CMS, the so-called Owen cytoplasm, which was discovered in an old cultivar by Owen. We found that a mitochondrial gene termed preSAtp6, which encodes a 35kDa protein, is responsible for Owen CMS. Additional sources of male sterile cytoplasm have been described, some of which are considered to have different CMS mechanisms from Owen CMS. Example is the I-12CMS(3) cytoplasm derived from wild beets in Pakistan: the preSAtp6 protein was missing in the anther of I-12CMS(3)-cytoplasm plants and instead a CMS-associated ORF129 protein of 12kDa appeared. It is also worth mentioning that the 12kDa protein caused pollen disruption in transgenic tobacco plants when targeted to the mitochondria. Garden and leaf beet germplasm accessions were evaluated for the presence of normal fertile and male sterile cytoplasm using polymorphisms in the mitochondrial minisatellite *loci*. Eleven mitochondrial haplotypes were identified, of which two (named min 06 and min 04) were associated with Owen and I-12CMS(3) cytoplasm, respectively, and two others (min 09 and min 18) with normal cytoplasm. The results indicated that normal cytoplasm predominated in both the garden and leaf beet gene pools, whereas Owen cytoplasm rarely occurred. In addition, I-12CMS(3) cytoplasm was sporadically found in the beet germplasm accessions examined.

S12.269

Diversity in *Capsicum* landraces Cultivated in Brazil

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In Brazil many *Capsicum* landraces are cultivated. The *Capsicum* Active Germplasm Bank of Embrapa Temperate Agriculture, located in Southern Brazil, since 2003 develops activities of collecting landraces of peppers to *ex situ* conservation. Today,

this bank has 265 accessions, collected from farmers of different Brazilian regions. These accessions are landraces traditionally grown by farmers, who made selection and produce their own seeds for planting in their fields. This work has the objective to report the diversity of landraces conserved in the *Capsicum* Active Germplasm Bank of Embrapa Temperate Agriculture. In the bank there are accessions of *C. annuum*, *C. baccatum*, *C. chinense* and *C. frutescens* landraces cultivated in Brazil. Morphological characterization allows the identification of a wide genetic diversity for traits such as color, shape, size of leaves, plant height and growth habit. Among the accessions can be found landraces in which the fruits show different colors such beige, yellow, orange, red or purple. Fruits of different landraces have distinct shapes, as blocky, campanulate, elongate, almost round or triangular, with different sizes and pungencies (from sweet to very hot). By morphological and agronomic evaluation of these accessions was possible to distinguish them and indicate the most promising for use in breeding programs for specific purposes. In this collection there are accessions with great potential for ornamental use because they have traits highly desirable for this purpose, such as compact architecture of plant, erect fruits that show several colors during the ripening process. Other accessions are ideal for the preparation of pickles and others for the processing of spices. Landraces without pungency can be directed to the fresh market. This Germplasm Active Bank aggregate important diversity of *Capsicum* landraces grown in Brazil and can be used as a source of variability in different breeding programs.

S12.270

Effects of Different Genotypes and Growing Condition on the Morphological Traits of Seed Stalk, Seed Yield and Quality of Table Beet (*Beta vulgaris* L.)

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In the study two monogerm seed inbred lines, three multigerm seed inbred lines, twelve cultivars with multigerm and cv. 'Patryk' with high percentage of monogerm seeds were tested. The plants were grown in the isolated boxes, plastic tunnels and field conditions. Statistic differences were found between tested lines and cultivars, regarding to the height of seed stalk, mean yield of seeds per plant and seed quality. Also the condition of growing influenced to the studied traits of beet genotypes. It was noticed that both monogerm as well as multigerm plants grown in the plastic tunnels, got the highest seed stalk, while the seed yield of monogerm lines was much lower as compared to the multigerm objects. In case of all tested plants, the best seed yield was obtained for the plastic tunnels. Relatively poor seed yield of monogerm genotypes was a result of a very late ripening of seed stalks, that showed tendency to form a bush type of seed stalk with much more stalks as compared to the multigerm plants. Additionally, in the monogerm lines a fasciation of the leaves and seed stalk were observed. The weight of 1000 seeds of plants grown in the field and plastic tunnels was much higher than in the case of objects from isolated boxes. Germination capacity of seeds harvested in the plastic tunnels and isolated boxes was better as compared to the field plants. However, monogerm genotypes always got lower percentage of normal seedlings than multigerm ones. These results demonstrated the necessity of improving the monogerm inbred lines in order to accomplish in seed production.

S12.271

Resistance to *Meloidogyne incognita* in Accessions of *Malvaceae*

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The experiments were conducted in 2008, in Jaboticabal-SP, in a greenhouse with the aim of evaluating resistance to *Meloidogyne incognita* in accessions of *Malvaceae* (Vinagreira, Algodão IAC 03/979, Algodão PRO 277, Algodão IAC 24, Algodão

IAC 20-233, Algodão PR 136, Quiabo Colhe Bem IAC and Quiabo Santa Cruz 47). Ten plants were utilized per material, where each plant was considered a repetition. Resistance of the plants was determined at the time of transplantation of seedlings to ceramic pots, where the root system was inoculated with 3000 eggs of *M. incognita*. At 60 days after inoculation, the plants were tested for nematode resistance based on the reproduction factor. All materials, except the cultivars of quiabo Colhe bem and Santa Cruz 47, showed resistance to *M. incognita*. The utilization of these six materials as rootstock of commercial cultivars of okra can be promising, but compatibility of graft between the plants needs to be studied further.

S12.272

Characterization for Bioactive Compounds of Spanish Pepper Landraces

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After the discovery of America, Spain was the point of entrance to the Old World for several vegetables native to America, including peppers (*Capsicum* spp.). Such diversity of introduced peppers evolved, after hundreds of cycles of selection under specific conditions, to a plethora of landraces, in particular of *Capsicum annuum* L. As a consequence, Spain became an important center of diversity for this crop. At the nutritional level, *Capsicum* peppers are known for their content in antioxidant bioactive compounds. However, systematic studies of the variation for this type of compounds among Spanish landraces have been not been undertaken. As part of our pepper breeding program, we present the results of our studies on the ascorbic acid content (AAC) and total phenolics (TPs) of twelve important Spanish pepper landraces. Several foreign landraces were included as external controls. Given that *Capsicum* peppers can be utilized in both mature (ripe) and green (unripe), all landraces have been analyzed in both stages. A wide range of variation was found for both AAC and TPs at both ripening stages. Thus, AAC ranged between 570 and 2100 mg/kg (on a fresh weight basis) in mature fruits and between 60 and 750 mg/kg in green fruits. All cultivars showed a higher AAC in the mature stage than in the green stage (on average two-fold higher, but up to 10-fold higher in some cases). TPs content ranged between 530 and 2100 mg/kg in ripe fruits and between 350 and 1700 mg/kg in green fruits. Although, on average, ripe fruits had 40% higher TPs content than green fruits, several landraces showed higher TPs content when unripe. This work has allowed us identifying the landraces with a higher content of nutritionally bioactive compounds, as well selecting sources of variation for breeding programs aimed at improving the nutritional value of peppers.

S12.273

Polyclonal Selection in Regional Garlic Clones

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Garlic is one of the most important bulb vegetables grown in Mediterranean regions, used as spice and flavouring agent for foods. In Portugal *Allium sativum* L. is strongly linked to gastronomy and tradition in rural communities. In order to obtain a polyclonal variety well adapted to the country conditions, the Portuguese National Genebank started in 2000, with the collaboration of Instituto Superior de Agronomia, a garlic selection program. A group of 142 clones previously prospected all over the country were compared in 2001 in a field trial at Barcelos. The selection was focused on bulb related traits including bulb weight, number of cloves per compound bulb, clove weight and skin colour. Mixed models were fitted to experimental data in order to estimate broad sense heritability and to predict genetic gain resulting from the selection of the 30 better clones. In the years 2003 and

2004, these selected clones were tested in six different locations: Guimarães, Braga, Esposende, Vila do Conde, Elvas and Faro. According to the data of the above referred traits and also from allicin content and environmental stability, a polyclonal group of 9 clones was selected (the new variety proposed).

S12.274

Evaluation of Portuguese Garlic (*Allium sativum* L.) Clones Based in Morphological Characteristics and Allicin Content

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The garlic (*Allium sativum* L.) assumes great importance in horticultural production and since early civilizations used as a spice and medicinal plant. This species is traditionally multiplied by vegetative propagation, however presents a significant morphological and chemical diversity. The Portuguese National GeneBank has a large collection of *Allium sativum* L. preserved as field collection. The *Allium* collection was morphologically characterized and evaluated for the allicin content, and from the 301 accessions a group of 30 was selected Biodiversity International descriptors for the *Allium* genera were applied for the morphological characterization and for the quantification of allicin HPLC was used. The analysis of variance showed that the garlic clones were significantly different in all evaluated quantitative parameters. Bulb allicin content depended on the garlic ecotype and the mean values ranged from 0.71 to 6.58 mg/g dry weight. Two major groups were originated by UPGMA clustering method analysis on morphological characters. In general it was found that was no relation between phenotypic divergence and geographical origins.

S12.275

Development of Cultivars with High Component of Rebaudioside A in *Stevia* (*Stevia rebaudiana* Bertoni)

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The use of steviol glycosides as an intense sweetener and flavor enhancer at a range of foods is technologically justified in stevia plant. Steviol glycosides are high intensity sweeteners 250-300 times sweeter than sucrose, that also have a flavor enhancing effect when used in association with other flavors. This study was conducted to determine the development of cultivars with 55.2% rebaudioside A's component, and the optimal concentrations of plant hormone for multiple-shooting over twenty-five fold on MS medium contained 0.1 mg/L BA. They can be used in a wide range of foods and beverages that contain sugar, and can either be used in conjunction with sugar or intense sweeteners or as a total sugar or intense sweetener replacement.

S12.276

Ethanobotanic and Genetic Variability Studies in Water Chestnut (*Trapa natans* var. *bispinosa* Roxb.) under Central Uttar Pradesh

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Water chestnut (*Trapa natans* var. *bispinosa* Roxb.) locally known as Singhara & buffalo head fruit, is an important annual aquatic fruit crop belonging to order Trapanaceae, family Trapaceae. Water chestnut is valued throughout the orient as a delicacy and a good source of starch and minerals. It has various nutritive and medicinal properties and is reported to accumulate heavy metals from polluted water bodies. Since waterchest nut is being cultivated on a large scale in U.P., a

preliminary survey was conducted by BB Ambedkar University, Lucknow from 2005-2008 and collection and evaluation of different cultivars of water chestnut for their physical and biochemical parameters was done in order to explore the possibility of genetic variability in water chestnut. The studies showed that considerable variation was found in the leaf length (4.33-6.60cm), leaf breadth (4.10-7.86cm), petiole length (5.96-10.46cm), fruit pedicel length(3.70-5.20cm), fruit length (3.11-4.89cm), fruit breadth(3.38-6.38cm), cheek diameter of fruit(3.26-4.41cm), fruit weight(13.20-66.5g), fruit volume (11.30-27.80ml), specific gravity of fruit (1.01-1.37), kernel length(2.85-4.89cm.), kernel breadth (3.24-4.84cm), cheek diameter of kernel (1.20-4.13cm), kernel weight (7.0-17.50g), kernel volume (4.80-13.68ml), specific gravity of kernel (1.16-1.76), peel weight (4.74-21.57g), thickness of peel (0.14-0.40cm), kernel : peel ratio (0.91-4.73), TSS (4.33-10.55 oB), ascorbic acid (5.05-9.70 mg/100g) and acidity (0.07-0.10%). Water chestnut, grown in the Lucknow region primarily by the poor or marginal farmers, requires very less input for production. It is a potential crop for exploiting the waterlogged and marginal lands and thus, germplasm studies for developing high yielding varieties with superior traits may prove beneficial.

S12.277

Black-Rot Resistance and Clubroot Resistance Are Transferred to the Fertile Somatic Hybrids from *B. nigra* to *B. oleracea*

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Brassica nigra, one of the basal species in the U-triangle of *Brassica* genus, generally is a wild plant, which is reported bearing resistance for black rot and club root diseases popular existed in *Cruciferae* crops. There is a shortage of the resistant germplasm and narrow diversity in the breeding material of *B. oleracea* owing to the long year's artificial selection. Therefore, it could have practical significance to transfer the resistance from *B. nigra* to *B. oleracea* via interspecific somatic hybridization. In this study, symmetrical and asymmetrical somatic hybrids between *B. nigra* and *B. oleracea* were obtained by protoplast fusion. Their progenies were got by selfing or backcrossing with *B. oleracea*. The hybrid nature of the regenerants and their progenies were confirmed and analyzed by molecular marker display, morphological observation and chromosome identification. The fertility varied a lot, generally, it is higher in symmetrical hybrid than in asymmetrical one, and the former has a more well-balanced meiosis behavior as revealed by GISH results. Advanced selfing or backcrossed progenies were got in some fertile or partial fertile hybrids, and subjected to Xcc. and *P. brassicae* resistant assessment with parental lines by artificial inoculation and exposure to the pathogen carrier field. Results showed that the *B. nigra* genotype had resistance to some isolates of Xcc. in China, and had resistance to *P. brassicae* existed in the Yun Nan province. The somatic hybrids obtained the resistances from *B. nigra*. The resistance was kept well in most of the advanced selfed hybrids (S4) but was segregated in the backcrossed progenies. Genome SRAP analysis revealed the existing of new DNA bands, which indicated the possible recombination of chromosomes. The hybrids displayed a middle or cauliflower lean phenotypes. Our researches again proved that somatic hybridization should be a valuable tool for producing pre-breeding material with increased genetic diversity.

S12.278

Morphological Characterization of Bell Pepper (*Capsicum annuum* L. var. *grossum* Sendt.) Genotypes and their Application for Distinctness, Uniformity and Stability Testing

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A study was carried out during 2004-05 on morphological characterization of bell pepper (*Capsicum annuum* L. var *grossum* Sendt.) and to establish distinctness of

the candidate variety from all other varieties. A total of 25 lines of bell pepper were grouped for several agromorphological descriptors. All the genotypes were intermediate/compact in growth habit except genotype Mid Way (prostate) and SKAU-SP-614, SKAU-SP-609, SKAU-SP-633-1, SKAU-SP-609-1, Rajer Collection, EC-464107 and EC 464115 (erect). Wide diversity (31 to 70 cm) has been observed in plant height. All the genotypes have erect flower position except SKAU-SP-633-1, Kandaghat Selection (intermediate) and Rajer Collection (pendent). The blocky fruit shape, which is most desirable for Indian market was observed in genotypes Nishat-1, SKAU-SP-614, SKAU-SP-616-1, SKAU-SP-633-1, SKAU-SP-82-1, SKAU-SP-648, SKAU-SP-601, SKAU-SP-609-1, Capsicum-143570, HC-201, Kandaghat Selection, EC-464107, UHF-494, California Wonder, Mid Way, Local Belgam, EC-464119 and EC-464115. Almost all the varieties are in green group for fruit colour except Russian Yellow and EC 464115 which are in yellow group. Such characterization of released cultivars as well as exotic and indigenous lines will provide valuable information for the strengthening of further breeding programme in bell pepper.

S12.279

Evaluation of Some Tomato Germplasm for Resistance to Tomato Yellow Leaf Curl Virus Disease in Ghana

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The importance of tomato in Ghana cannot be overemphasized. However, it is susceptible to a number of pests and diseases, including viral disease for which the best control strategy is genetic resistance. Tomato leaf curl disease is reported to be widespread in Ghana causing severe yield losses. The disease, caused by a whitefly-transmitted geminivirus, is of economic importance and has become a recent problem on tomato farms in Ghana. Farmers ignorantly use pesticides to control the TYLCV disease but to no avail. The best way to combat this disease is to use plants that are resistant to the disease. Consequently, the objective of this study was to screen fifteen germplasm available from the AVRDC. The germplasm, including known checks, were first screened in greenhouse in a complete randomized design (CRD) with four replications. The tomato plants were exposed to whiteflies infected with tomato leaf curl disease. Scoring was then done when plants were 30 days, 45 days and 60 days after inoculations. These were repeated on a disease hot spot in the field for further evaluation. They were in randomized complete block design (RCB) with three replications. Scoring was done when plants were 30 days, 45 days and 60 days after transplanting. Other horticultural characteristics such fruit weights, plant height, number of fruits per plant were also taken. There was no significant difference between accessions on the incidence of the disease and in terms of their ability to resist the disease. However, tomato accessions A1, A2 and A3 showed mild symptoms of TYLCV infection. Other lines also have lesser incidence and severity of TYLCV than the control and could be used in incorporating TYLCV resistance in breeding work. There were significant differences in plant height and the number of fruits per plant of the tomato accession.

S12.280

Brassica Resource Bank

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The genus *Brassica*, phylogenetically related to *Arabidopsis thaliana*, is one of economically important crops and a botanical model of plant polyploidization and rapid phenotypic evolution. We established the *Brassica* Resource Bank (BRB) in order to supply basic plant materials for structural/functional genomics and breeding of *Brassica*. BRB has served various *Brassica* species, especially, doubled haploid and recombinant inbred lines, and DNA stocks including BAC libraries and cDNA libraries. BRB has collected specific-*Brassica* cultivars as well as subspecies from international plant resource centers. Moreover, all germplasms of BRB have been propagated and maintained. Currently, BRB has collected 9,689 accessions

for *Brassica* species, 91,446 clones for cDNA libraries, and 277,440 clones for BAC libraries, and 1,398 genetic markers. BRB has served more than 621,345 clones, 280 genetic markers and 4,023 lines to researchers in 10 countries since 2003. Information and other requests for genomic resources of *Brassica* are accessible at <http://www.brassica-resource.org>.

S12.281

Portuguese Accessions of *Cucurbita* spp. and *Citrullus lanatus*: Conservation, Evaluation and Breeding

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Cucurbitaceae is one of the most important families of vascular plants. This family includes 118 genera and 825 species. The five major cucurbit crops are *Citrullus lanatus* Thumb Mansf. (watermelon), *Cucurbita maxima* Duchesne (pumpkin), *Cucurbita pepo* L. (squash), *Cucumis sativus* L. (cucumber) and *Cucumis melo* L. (melon). *Citrullus lanatus* and *Cucurbita* spp. are very important in the Portuguese agro-ecosystems, associated with maize, beans and cabbage. Due to the importance of *Citrullus* and *Cucurbita* spp., the Portuguese National Genebank (BPGV), has done systematic collecting missions in Portugal (Mainland and Madeira Island). Since 2001, BPGV in partnership with other National Institutions, Escola Superior Agrária de Santarém, Direcção Regional de Agricultura e Pescas do Algarve and Universidade do Algarve, has been carrying out activities related to preservation, characterization, evaluation and pre-breeding. In Portugal, in BPGV, the Cucurbitaceae collection preserved in *ex situ* conditions (medium and long term) adds up to a total of 573 accessions. The collection of *Citrullus lanatus* and *Cucurbita* spp. has a total of 355 accessions, representing 62% of the whole collection: (37 of *Citrullus lanatus*, 19 of *Cucurbita ficifolia*, 74 of *Cucurbita maxima* and 224 of *Cucurbita pepo*) Based upon the diagnosis of the preserved collection, further germplasm collecting missions were recommended in Algarve Region. AFLP and RAPDs markers were used to check the assignment of accessions to *Cucurbita* species: *C. pepo*, *C. maxima* and *C. moshata*. The morphological characterization followed the *Cucurbita* spp. and *Citrullus* descriptors, elaborated by Bioversity International, integrated in the European Cooperative Program for Genetic Resources, Cucurbits Working Group. Characterization data are reported herein. Departing from the most homogeneous accessions of *Citrullus lanatus*, *Cucurbita maxima* and *C. moschata*, three varieties, one of each species, have already been selected and registered in the National Catalogue of Varieties.

S12.282

Genetic Characterization of "Tomàtiga de Ramellet" Tomato Cultivars from the Balearic Islands

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"Tomàtiga de Ramellet" (TR) are long-storage tomato cultivars from the Balearic Islands. These varieties exhibit dramatically delayed softening due to the delayed fruit deterioration (DFD) phenotype, and they demonstrate good agronomic performance under non-irrigation conditions during the Mediterranean summer. We collected grower-saved seeds from 200 accessions of putative TR varieties from the Balearic Islands. Important differences were observed among TR cultivars for production, fruit quality, morphological, physiological and self-life parameters, suggesting the existence of significant genetic variability. Fruit were evaluated from

a replicated field trial with quantitative morphological data obtained using the Tomato Analyzer Software. Analyses detected distinct fruit-shape classes that were statistically separated at major nodes in the ontology descriptors for tomato. However, several accessions were intermediate or extreme for measured traits. Advances in tomato genetic resources are providing tools to investigate variation in cultivated populations of local tomato varieties. With the aim of achieving genetic characterization of the TR population, sequence variation was investigated using single nucleotide polymorphisms (SNPs), insertion/deletion (InDel), and simple sequence repeats (SSRs). Some accessions were found not to be TR, and clustered with elongated cherry tomatoes spread in Italy. The TR population appears to be a unique genetic resource relative to heirloom tomatoes (vintage varieties) and modern fresh market and processing varieties. The distribution of marker polymorphisms within fruit shape classes combined with the existence of intermediate or extreme shapes suggest that several tomato genotypes were introduced to the Balearic Islands, and occasionally exchanged genes, through pollen, leading to the wide range of variability observed for the Balearic TR population.

S12.283

Genetic Resources of Jointed Goatgrass (*Aegilops cylindrica* Host) in Hungary

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In order to assess genetic resources of jointed goatgrass in the Hungarian flora Department of Agrobotany in collaboration with Faculty of Horticultural Science has conducted regular collecting missions in different parts of Hungary, where *Aegilops cylindrica* has been expected to occur. The objective of this study was to understand the main distribution areas of jointed goatgrass populations in Hungary and to explore conservation possibilities (*in situ*, *ex situ*) for maintenance genetic resources of this important crop wild relative species. During the period between 1993–2009 the Department of Agrobotany organized several expeditions in the various floristic regions of Hungary to investigate natural habitat of jointed goatgrass. In Hungary jointed goatgrass is found along roadsides, pastures and rarely near the wheat fields. The main area of distribution included the Great Hungarian Plain, the hilly territories of Northern Middle mountain ranges and the Transdanubian Middle mountain ranges. Populations found from low (82–88 m above sea level) to moderate (99–220 m above sea level) altitudes with various densities. A total of 59 habitats were explored from 26 locations and 111 accessions have been collected. All collected samples have been stored and maintained in the active genebank collection at the Department of Agrobotany, Tápiósele. These results should contribute to the knowledge regarding the native distribution and possibilities for realizing conservation of *Aegilops cylindrica* in Hungary.

S12.284

Screening a Diverse Collection of Heirloom Tomato Varieties for Quality and Functional Attributes

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Fruits of cultivated tomato exhibit a high phenotypic diversity, having varied sizes, shapes and colors. This phenotypic variation is particularly apparent in heirloom varieties. In the present study a set of varieties, including many genotypes with fruit described as brown, black and purple were screened for quality and functional attributes. The collection includes mainly heirloom tomato varieties obtained from TomatoFest (<http://www.tomatofest.com>) and some traditional Spanish varieties. Plants were grown in the open air, and their fruits were analyzed for color, titratable acidity and soluble solids content (TSS). In a subset of the collection we have studied functional attributes, as the antioxidant activity and the polyphenols content. Wide

variations were observed in all of the measurements. For example, the soluble solids content varied between 3.91 and 7.21 °Brix, and the titratable acidity between 0.49 and 1.07 g/100g. Very high levels of diversity were also found for the functional attributes analyzed, varying the antioxidant activity between 7.90 and 56.82 mg/100g and the polyphenols content between 26.73 and 43.17 mg/100g. Data obtained have been analyzed for their mutual relationships, and we have found significant correlations among polyphenols content and the titratable acidity, soluble solids content and antioxidant activity. These results could be useful for tomato breeders working on the development of new varieties. The diversity found in heirloom tomato varieties could be a rich source of germplasm for quality and functional attributes.

S12.285

The Gene Pool of Bulbous Plants (*Alliaceae*) in Armenia

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The area of the Republic of Armenia characterized with mountainous vertical zoning is rich in wild species of cultivating plants including representatives from *Alliaceae* family. The species growing in the country's territory are grouped into two genera - *Nectarascordum* Lindl. and *Allium* L. The first genus is represented by only one perennial variety - *N. tripedale* (Trautv.) Grossh. The other 43 varieties are belonged to *Allium* L. genus, which includes cultivating varieties and are grouped into 4 subgenera. The representatives of *Allium* L. genus are perennial plants usually with specific smell of onion or garlic. The flowers in terminal umbrella consist from shawl accreted in 2-4 parts. The peduncles are with bracts on the bottom or without them. The perianth is star-shaped, 6 parts in two circles, fibrils simple or with lateral teeth. Pollen grain distal or lateral-monotonous. Ovary is on the top, three bed with nectars, seed germs are 1-2 or 10 a bed. Fruit is rind boll. Seeds are three-sided, spherical. The most representatives belonging to the genus are widely spread in mountainous, stony areas in alpine and sub alpine meadows. The onion, garlic and leek have been cultivated in Armenia from immemorial times. The shaped local populations of these crops are mainly used in valley parts of the country. In Armenia only leek is spread from biennial deciduous variety. In the first year juicy leaves are harvested, and in the second year plant blooms and gives seed without getting out of the soil.

S12.286

The Role of Home Vegetable Gardening for Conservation of Plant Genetic Resources in Remote Southwest Island in Japan

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Self-sufficient home vegetable gardening used to be practiced all over Japan, but this is now limited to rural areas. In order to ascertain the role of home gardening in the conservation of plant genetic resources, and especially with respect to traditional vegetables, herbs, fruit trees and local varieties, we conducted a study of six remote Southwest islands in Okinawa prefecture. Here, with the exception of changes accompanying improvements in infrastructure, residents continue to live according to their traditional lifestyle. We know from agricultural statistics that the major cash crop in these islands is sugarcane. However, these statistics do not indicate the kinds and numbers of traditional vegetables cultivated in the islands in question. The present survey showed that home vegetable gardens contain numerous traditional vegetables and herbs, including *Crepidiastrum lanceolatum* (Houttuyn) Nakai, *Peucedanum japonicum* Thunb., *Gynura bicolor* (Roxb. ex Willd.) DC., sponge gourd, bitter gourd, white gourd, fennel and Indian mulberry, and local varieties of taro, radish, carrot, cucumber, rakkyo (*Allium chinense* G. Don), leaf mustard, banana, and citrus. Along with traditional vegetables, various common vegetables are cultivated in home vegetable gardens. The results of visual inspections at home gardens and of interviews with 161 gardeners showed that 9 to 96 (mean 38) kinds of vegetables were cultivated in a household's home garden over the course of a year. The above results lead us to conclude that home vegetable gardening plays a role in the conservation of plant genetic resources.

S12.287

Agro-Morphological Characterization of 18 Traditional Tomato (*Lycopersicum esculentum* Mill.) Varieties from Castilla-La Mancha (Central Spain)

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In this study, 18 traditional tomato varieties from Castilla-La Mancha (Central Spain) were agro-morphologically characterized, and relations among the different parameters observed, related to the weight, shape and physical-chemical characteristics of the fruits (dry matter and juice content, firmness, equatorial and shoulder exterior colour, total soluble solids, pH, total acidity), and the germination capacity, colour, weight and size of the seeds, were found. Statistical tools of Multivariate Analysis were applied. First, a Principal Component Analysis (PCA) allowed to reduce the dimension relative to all the variables considered to four principal components, explaining approximately the 70% of the total variability. Through this analysis, the parameters having most weight on this variability were determined, and a first classification of the tomato varieties was done, distinguishing a group with all the local accessions which presented characteristics belonging to the local "Moruno" type, very appreciated in Castilla-La Mancha and characterized by fruits with the presence of dark shoulders, strong-medium-ribbed, high weight, number of locules and dry matter content, flattened predominant shape and irregular fruit cross-sectional shape. From the study of the correlations of the original variables with the extracted components and the corresponding matrix of correlations, such associations among variables were detected and quantified. A multivariate data Analysis-Cluster corroborated the grouping of the "Moruno" varieties, being typified in a simple cluster all the accessions belonging to this group. A strong positive correlation between the variables fruit flattened predominant shape and irregular cross-sectional shape, strong-ribbed, fruit weight and number of locules, slightly flattened predominant shape and slightly-ribbed was found. A strong negative correlation between irregular cross-sectional shape and fruit shoulder lightness, pH and the shoulder a*/b* ratio color, dry matter and juice content, and, in a minor measure, between the seed weight, size and fruit dry matter content, was recorded.

S12.288

Agro-Morphological Characterization of 16 Traditional Pepper (*Capsicum annum* L.) Varieties from Castilla-La Mancha (Central Spain)

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The recovery of local varieties, nowadays suffering an important process of genetic erosion, is an indispensable activity to safeguard the plant genetic resources, patrimony of our people and part of our identity. The excellent organoleptic quality and the high wildness of the traditional varieties would allow their reintroduction on the market, satisfying the demands of a society increasingly demanding and compromised with determine aspects related to the health and the respect to the environment. In this work, 16 traditional pepper varieties from Castilla-La Mancha (Central Spain) were agro-morphologically characterized, attending to aspects related to vegetative plant, fruits or seeds. A multivariate statistical analysis interpreted satisfactorily the information obtained in the characterization of the varieties studied. The Principal Component Analysis (PCA) allowed to establish two well-differentiated groups, the accessions of pepper Grossum group opposite to the rest. An Analysis of Conglomerates corroborated the previous grouping and established a second group which included all the accessions of the pepper Longum group.

- Pepper Grossum group: characterized by fruits with a large number of locules, width, wall thickness and weight and a low dry matter content, and seeds with high weight and size.
- Pepper Longum group: characterized by fruits with a high dry matter content and a low number of locules, width, wall thickness and weight, seeds with a small weight and size, and, in minor measure, fruits longer and more

lightness, and seeds with a lower percentage of germination compared to the previous group. There was an important positive correlation among the fruit weight, width, wall thickness and number of locules. This work reveals the suitability of the multivariate statistical methods for analysis of data in this agronomic field.

S12.289

Pea Landraces and Cultivars Response to Acclimation at High Temperatures and Its Relation to Morphological and Agronomic Characteristics

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Eleven populations of pea (*Pisum sativum* L.) from the ICARDA Pea Germplasm Collection, comprising landraces from Mongolia, Poland, Haiti, Uganda, Spain, Eritrea, Colombia, Turkey, Denmark, Canada and Estonia and five Portuguese commercial cultivars (cv. "Progress", "Television", "Telefone", "Maravilha" and "Rondo") were assessed for the lethal temperatures (LT). These ranged from 42 °C to 44 °C and no significant correlation between climate at the place of origin and LT was found; commercial varieties showed the highest LT. From these populations, seven were tested for acclimation to high temperature; all showed a positive response to acclimation at 39 °C. To establish a relation between LT and both morphological and agronomic characteristics, these populations were further characterized using twenty three IPGRI descriptors, throughout a randomized block design experiment, in Ponte de Lima, NW Portugal. Multivariate (principal components, cluster) analysis was performed on these characters, and four clusters were found. The cluster 1 included the landrace from Poland (acc. 51758) and commercial cultivars Progress and Rondo, that were early season and dwarf type and its LT was 43-44 °C. The same LT of cv. Television (cluster 3) corresponded to a mid-early season and mid-dwarf type. Clusters 2 with landrace Haiti (acc. 124634) and cluster 4 with Uganda (acc. 50223), had LT of 42 °C but different characteristics, early season and dwarf/mid-dwarf type for Haiti and late season and mid-dwarf/tall type for Uganda. The Uganda characteristics were similar to those of Spain (acc. 123192) with the same LT of 42°C, that was not included in the cluster analysis as it was found an outlier through the principal components analysis. Clusters 1, 2 and 3 presented smooth peas and cluster 4 rough peas. Results showed high levels of diversity between populations that may help future use of pea germplasm collections and studies concerning temperature stress tolerance and heat shock processes.

S12.290

Comparative Study of *Phaseolus vulgaris* L. Landraces from Portugal and Angola

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P. vulgaris (common bean) is a species of American origin which was introduced in Portugal during the XVI century. The good adaptation of this crop to the Portuguese environmental conditions resulted in a great number of landraces that are still in use. It is not known when the common bean was introduced in Angola. However, by the end of the XVII century it was already cultivated mainly for local consumption by the European settlers. Twenty five common bean landraces from Portugal and Angola, maintained in the Banco de Germoplasma - Genética (PRT 005) and in the Centro Nacional de Recursos Fitogenéticos (Angola), were characterized with the IPGRI *P. vulgaris* descriptors and their genetic diversity was further assessed using 15 microsatellites (SSRs). One commercial variety was also analysed for comparison purposes. Fragment analysis was carried out using the automated sequencing system CEQ 8000 and fragment lengths were estimated using internal standards. The evaluation of marker polymorphism and the screening for identical

genotypes were performed according to IDENTITY 1.0. The proportion of shared alleles distance was calculated using MICROSAT. Twelve of these microsatellites were polymorphic, with the number of alleles per *locus* ranging from 2 to 7 alleles. Principal Component Analysis of the phenotypic observations showed that the first component separates the accessions from Portugal and Angola according to their seed characteristics. In what concerns the microsatellites results three Portuguese landraces with different names displayed the same SSR allele size, indicating they are synonymous landraces. Concerning the remaining landraces they can be discriminated from each other with the 12 SSRs but no distinction between Angolan and Portuguese accessions can be found. In this study SSRs markers proved to be effective to characterize common bean landraces, what supports the importance of these markers for the management of germplasm collections.

S12.291

The Study of Improvement of Tomato Lines for the Longevity Period of Fruits

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Fruit ripening is a complex, genetically programmed process that culminates in dramatic changes in colour, texture, flavour, and aroma of the fruit flesh. The following research has been done in greenhouse and the laboratory of horticultural science of Ferdowsi University in Iran. In this research 16 tomato lines in a factorial design were crossed to study the combining ability of parental lines for yield and fruit longevity in F1 hybrids. The 16 lines were arranged in two parental groups, that 7 lines were as mother lines and 9 line where as father lines, and the genotypes were planted in a four replication completely randomized design. The main factors that were measured and studied are yield and the longevity period of fruits. Results from analysis of variants show that hybrids, general combining ability and specific combining ability have a significant difference in 1% level. The highest yield was obtained from the Fla × CT6 hybrid that was equal with 10.180 kg per plant. The highest fruit longevity was belonged to LA3770 × Al3012 hybrid with an amount of 151 days. The study of combining abilities of parents shows that male line, Al30, with 27.7 days and female line Fla with 21.5 day storage period are better than other parental lines. The most specific combining ability was found to be related to hybrid Dns × Kalg with 73.1 day period. Also, the biggest amount of Heterosis was shown to be the same Hybrid with 97 day period longer than its parents' average storage period. Finally the lines Al3012, LA1793, Fla and LA3770 were known as the lines with high combining ability for producing the hybrids with long lasting fruits and also the lines 611, Al3013, Al3012 and Fla were known as lines with high combining ability for yield.

S12.292

Evaluation of Genetic Diversity of Common Bean (*Phaseolus vulgaris* L.) Resources from Madeira

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The ISOPlexis Banco de Germoplasma maintains large collection common beans traditionally cultivated on the Madeira Island. This collection represents an important source of genetic diversity, as local crop varieties were cultivated for centuries using traditional practices. The Island of Madeira is a region where common bean hybrids were not introduced so far. In order to achieve a better knowledge of the Madeiran bean diversity, an evaluation of 50 germplasm accessions has been performed, representing a first attempt for understanding crop genetic resources diversity. The criteria for the choice of the analyzed populations took into account a wide range of ecological conditions present on Madeira Island, diversity of common names, and some morphological and reproductive traits. The beans morphological characterization, using 58 characters was con-

ducted during two vegetative cycles. A multivariate analysis of morphological data allowed to identify 15 phenotypically distinct groups. Four of these groups clustered with accessions of determinate growth behaviour (dwarf beans), and 11 groups consisted of accessions with indeterminate growth (climbing beans). The dissimilarity of these groups was confirmed by Tukey tests and discriminant analysis. The racial rank of these groups will be proposed and a brief description of the common bean landraces will be presented. Simultaneously, the biochemical characterization of bean accessions based on phaseolin and total storage protein patterns revealed that 92% of accessions originated from the Andean domestication centre, with C and T phaseolin types representing 56% and 36% of total accessions. The molecular characterization that was performed using nine SSRs, allowed us to determine the genetic variability of accessions and link them and the morphological clusters with some morphological traits. Currently, we are analysing nutritional and anti-nutritional values of these accessions, anticipating that such analyses will allow further valorisation of the local bean varieties (landraces).

S12.293

Conservation of Vegetable Crops Genetic Resources in Armenia: Needs and Priorities

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The territory of Armenia is considered as primary and secondary centers of origin for many vegetable crops with large diversity of indigenous species. Vegetables in Armenian flora are represented by numerous species of the *Solanaceae*, *Brassicaceae*, *Liliaceae*, *Chenopodiaceae*, *Cucurbitaceae*, *Apiaceae*, *Asteraceae*, *Portulacaceae*, *Lamiaceae*, *Malvaceae* families. As per country needs, priority vegetable plants include tomato, cabbage, cucumbers, bulb onion, egg-plant, pepper. Besides these, beet, carrot, garlic, vegetable marrow and green pea are also cultivated. Vegetable crops are grown in almost all regions and their cultivation areas are distributed according to an altitude gradient. Widely distributed vegetable crops such as beet, carrot, melon, onion, garlic, egg plant, asparagus, wild horse radish, artichoke, leafy vegetables (prickly lettuce, spinach), spicy plants are represented by different species, forms and ecotypes of their wild relatives, which are of high importance from the point of view of their high adaptability to agro-climatic conditions and resistance to pests and diseases. Due to over exploitation of pastures and grasslands, untimely and unregulated harvesting of wild edible plants, increased pests and diseases virulence, genetic erosion, climate changes and environmental disasters, plant diversity in general and vegetable crops genetic resources in particular are considered to be in need of conservation attention. Both *in situ* and *ex situ* conservation actions are implemented in the country to conserve vegetable plants genetic resources to ensure their effective utilization in breeding programmes and for keeping environmental balance. Priority needs for better conservation of vegetable plants genetic resources include study (distribution, inventory, mapping, evaluation) and monitoring of vegetable plant populations in nature, development of instructions/guidelines for widely used wild vegetable plants, performance of awareness and educative programs related to on-farm conservation of agrobiodiversity, *ex situ* conservation of entire gene pool of vegetable plants, accessions documentation, characterization and evaluation.

S12.294

Iron (Fe) Deficiency Tolerance in Tomato Recombinant Inbred Lines Obtained from Resistant and Sensitive Genotypes

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The first aim of this study was the determination of iron (Fe) deficiency tolerance levels of the reciprocal recombinant inbred lines (RILs) in tomato. The RILs (F6 generation) have been created using tolerant and sensitive inbred parents for long

time studies since 1994. The second aim was the obtaining data which can be used to map the quantitative trait *loci* (QTL) for iron (Fe) deficiency tolerance in tomato. In this study, totally 224 genotypes have been used. 107 genotypes were contained the cytoplasm of the resistant parent-Roza and 117 genotypes were contained the cytoplasm of the sensitive parent-227/1. The plants were grown in growth chamber in hydroponic medium. Thirty-five days old plants were subjected to Fe deficiency stress by 10 mM NaHCO₃. The experiment repeated 5 times and each experiment was continued for 57 days. At the end of the experiments, leaf chlorosis, active and total Fe concentrations have been determined. The measured parameters of the RILs have been shown the normal distributions. There were significant differences among the RILs for active, total irons ($P \leq 0.05$) and chlorophyll ($P \leq 0.001$) concentrations. It has been concluded that Fe deficiency tolerance in tomato is inherited quantitatively and controlled by multi genes. The findings of this study showed that there were not strong definitive data supporting to the possible cytoplasmic interactions in the inheritance of this trait.

S12.295

Morphological Characterization of Okra Germplasm (*Abelmoschus esculentus* L.) in Ghana

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Okra is the fourth most popular and an extensively cultivated vegetable in Ghana. In spite of the great value of the vegetable, information on characterization and diversity is limited. There is therefore the need to characterize available germplasm as a step to its diversity and improvement. Twenty-five accessions of okra were assessed for morphological traits. The experimental design used was a randomized complete block design with four replications. Data were collected on plant growth habit, general appearance, fruit shape, leaf shape, stem colour/pubescence, immature fruit colour/pubescence and petal colour. All the okra showed erect plant growth habit while more than half (60%) displayed unique orthotrop axis in terms of general appearance. Fruit shape varied among all the okra lines. 'Heart' leaf shape was highest among the lines by less than half of the okra entries. Green stem colour was highest by a little over half (56%) of the okra population while all the entries exhibited little stem pubescence. More than half of the okra entries (70%) displayed green immature fruit colour with about 64% showing smooth fruit pubescence. Petal (flower) colour proved 60% higher for golden yellow. Almost all the okra accessions showed symptoms of Okra Mosaic Virus (OMV) and Okra Leaf Curl Virus (OLCV).

S12.296

Olive (*Olea europea* L.) a Potential Oilseed Crop for India

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Olea europaea L. commonly known as olives belongs to the family *Oleaceae*. It represents a group of evergreen trees and shrubs distributed in the warm temperate and tropical regions of the world. It is a very important oil producing crop of the Mediterranean region. Olive oil is a rich source of polyunsaturated fatty acids (PUFA) and is absolutely free from cholesterol. The consumption of olive oil has been clinically advocated to people suffering from hypertension or coronary heart diseases. Moreover, it is also rich in minerals like iron, calcium and phosphorus besides other potential uses. Confined largely to high end consumers, olive oil is now gaining popularity because of its health benefits. The cultivated olive is an introduced plant in India and is grown in some parts of the country. India meets hundred percent demand of olive oil through imports mainly from Europe. The National Bureau of Plant Genetic Resources, New Delhi, has made efforts to introduce olive germplasm from different countries for oil/table

purpose and stress tolerance. However, most of these either could not be established or the yields were very poor. Promising varieties especially from France, Greece, Israel, Italy, Portugal, Spain and other olive producing countries, suitable to Indian conditions is required to be introduced. The olive cultivation in this country is still in its infancy stage and is restricted to a few provinces only. There is a requirement to identify other sites and suitable cultivars for its cultivation in the country. The paper aims to bring about the world collection of olive germplasm with reference to Indian requirement for large scale cultivation of this important crop in India.

S12.297

Underutilised Crops of Humid Tropics with Potential as Vegetable for Food and Nutritional Security in the Millenium

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Unlike in most other countries, large section of India's population subsists on vegetarian diets which are predominantly based on cereals. A wholesome and nutritious vegetarian diet has to be based on cereals, pulses, vegetables (especially green leafy vegetables), fruits and milk. The vitamins, minerals and antioxidants present in vegetables and fruits maintain health and prevent micronutrient deficiencies and diseases. The spread of exotic introduced vegetable and fruit crops have endangered the existence of many indigenous rare endemic crops which are easy to grow, resistant to pests and diseases, hardy and acceptable to local taste. The leaves of *Alternanthera sessilis*, *Basella* sp., *Boerhaavia diffusa*, *Sesbania grandiflora*, *Trigonella foenum-graecum*, *Pisona alba* are rich in vitamins and minerals. Tender shoots of bamboo (*Bambusa* sp.) are low in fat and calories and good source of fibre are also used as vegetable. Ivy gourd (*Coccinia grandis*), ridge gourd (*Luffa acutangula*), sponge gourd (*L.cylindrica*), parwal (*Trichosanthes dioica*, Roxb.), snake gourd (*T.anguina*) and bitter gourd (*Momordica charantia*) are fruit vegetables used as salads or in curries. The young pods and tender beans of lablab bean (*Dolichos lablab*) and winged bean (*Psophocarpus tetragonolobus*) have high nutritive value. Drumstick (*Moringa oleifera*) is a vitamin rich mineral packed nutritious vegetable grown for immature pods, leaves leaf and flowers. Other potential crops are jack fruit (*Artocarpus heterophyllus*), and bread fruit (*A. altalis*) which are rich source of carbohydrates and can serve as source of energy and nutrients. Research priority should be given for utilization of these crops for alleviating hunger and malnutrition, genetic improvement and molecular characterization to locate genes for nutraceutical properties.

S12.298

Selecting Discriminatory Morphologic and Agronomic Characters to Identify Spanish Tomato Landraces

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Seventy three different Spanish tomato landraces have been grown and characterized during three years, twelve in 2007, thirty seven in 2008 and twenty four in 2009, using the IPGRI tomato descriptors. In 2007 and 2009 sixty, both morphologic and agronomic, characters were measured and in 2008 these same characters, plus eight more morphologic, were measured. Analysis of variance, correlation analysis, and main component analysis were made for the data obtained every year. With the results of the analysis it has been possible to discard non discriminatory characters. The total number of discriminatory characters selected was forty nine: eight of them were discriminatory in every year, ten in two years and the rest in only one. With the discriminatory characters a classification of the studied landraces was made and discussed.

S12.299

Regeneration and Safety Duplication of a Faba Bean Prioritized Collection

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Vicia faba L., known as faba bean, broad bean (or “faveira” in Portugal), is an insect pollinated crop, member of the *Fabaceae* (*Leguminosae*) family which is cultivated primarily for its edible seeds. Thirty seven *V. faba ex situ* collections are known worldwide and one of them is stored in Portugal (INRB/INIA). This collection, mainly composed by Portuguese landraces of local origin, has not been characterized and is very limited safety-duplicated (with no representation in other faba bean international collections). Considering the great importance of the INRB/INIA collection, studies have already started, aiming at its regeneration, characterization and safety duplication. Some accessions were regenerated (in open pollination conditions in order to maintain the level of heterozygosity) in INIA – Elvas, during 2008/2009, and characterized according to the IPGRI/ICARDA Descriptors for faba bean. The field plots were surrounded with a tall rapessed crop (*Brassica napus*), which acted as a barrier from contamination by external pollen. Considering that *V. faba* is also attractive to a large number of arthropod pests, the presence and identification of the pests were registered at all stages of plant development. Data were analysed by Principal Component Analysis (PCA), using NTSYS-pc, version 2.01 and the results suggested that the first component allowed the separation of the accessions according to the seed length, the seed weight, 100 seed weight, pod weight and the number of days to flowering. The second component grouped the accessions according to the flowering period and the number of seeds per pod. The seeds produced will be stored in the INRB germplasm bank and a safety duplicate will be sent to the Svalbard Global Seed Vault and another one to the Centro de Recursos Fitogenéticos (CRF), Madrid, Spain.

S12.300

Genetic Variability of Artichoke “Argentinean Type” Growing in the Chilean Central Valley for Processing Purposes

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Artichoke “Argentinean type” is the main cultivar utilized for processing industry in Chile. Probably, its origin is the Spanish cultivar “Blanca de Tudela”, which it was introduced to South America through Argentina (Mendoza and San Juan Provinces) and lately to Chile. The aptitude of this cultivar for processing is based in its earliness for buds production, high yield, buds shape and compactness of the bracts. In Chile, actually, populations of 20,000 to 25,000 plants are utilized to obtain yields about 200,000 buds·ha⁻¹ (approx. 16 ton·ha⁻¹). A common practice among the Chilean farmers is to use vegetative material without selection for plant health and agronomic characteristics. However, the cultivar variability has been increased due to the exchange of vegetative material between different localities and the environmental natural selection. The aim of this study was to evaluate genetic variability within artichokes “Argentinean type” grown in Chile and to identify outstanding individuals for industry purposes. The main characters looked for were bracts compactness and buds shape. The genetic variability within the material under evaluation was determined through agronomic, phenotypic and genotypic characterization. Phenotypic and agronomic results showed that there was a high variability in buds yield per plant associated to buds shapes and compactness. However, the SSR molecular markers analysis did not show significance within the genetic material under evaluation, except for some specific clones, therefore these results would indicate that variations observed in phenotype and yield are strongly influenced by environment. Also, the best individuals have been evaluated and characterized for cynarin content and some gene expression for this trait.

S12.301

Sugar Gum Trees for Honey Production in Southern Atacama Desert: Bayesian Analysis of Genetic Parameters for Survival and Growth at Age 8

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Eucalyptus cladocalyx (sugar gum) flowers provide a reliable source for the production of honey, complementing the less prolific endemic and native species in Southern Atacama Desert, Chile. The plant breeding is recognized as an important tool for improving of the honey production in arid environments. The objective of this study was to evaluate the genetic control of survival and growth in 49 half-sib families of sugar gum, evaluated in Southern Atacama Desert, Chile, to achieve high biomass productivities by combining tree survival and with high growth rates. A Bayesian variance component estimation model was assumed using the Gibbs sampling algorithm. A threshold model was fitted to survival data, which were considered discrete variables with binary outcomes. Trees were planted during June 2001 in a random complete block design. Mean tree survival (measured 8 years after planting) was high (91.76%) and ranged from 66.7% to 100% (family means). Tree survival and diameter at breast height (DBH; measured at age 8) were moderately heritable with posterior means and credible intervals of $h^2=0.17$ (0.13-0.29) and $h^2=0.34$ (0.19-0.54), respectively. The posterior mean of the genetic correlation between DBH and tree survival was positive ($r=0.46$) and, according to the credible interval (0.06-0.81), it was significantly different from zero, indicating that selection for growth alone would have a positive impact on survival. The high survival rate found in the current study, confirms the potential of *Eucalyptus cladocalyx* to grow under the arid environmental conditions. The presence of a significant genetic variability suggests an opportunity for improving the tree productivity in the Southern part of the Atacama Desert.

S12.302

A Brief Note on Winter Cold Resistance of *Jatropha curcas* One Year After Trasplanting in Open Field in Southern Italy

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Jatropha curcas, L., belonging to the botanical family of *Euphorbiaceae* is a tropical shrub oil species with multiple uses and considerable potential as bioenergy crop. A trial has been conducted from May 2009 to the end of May 2010 to test the winter cold resistance in a location (Restinco, Brindisi, 40° N 39', 11° E 40') of Southern Italy. A latin square experimental design was performed containing 3 cvs. (1. Tais, from Brasil; 2. Karima, from Ghana; 3. Nadir, from Senegal) with size of the elementary plot of 75 m² (10 plants/plot) in row 3.0 m × 2.5 m apart. On May 27/2009 a hand direct seeding, setting 3 seeds in a hole 5 cm deep, was performed. In the same day seeding of the 3 cultivars was effectuated using plastic bags 30 cm × 15 cm with the goal to replace missing plants of the experimental trial. On July 3, 2009 trasplanting of the missing plants was performed using the plants contained in the plastic bags. The direct seeding was unsatisfactory. The plant emergence was 37%, 30%, 16.3% for cv. 2, 3, 1 respectively. The trial was conducted under water irrigation regime from seeding time to the end of August when the plants reached as average, the following height: 102 cm, 98 cm, 94 cm for cv 1, 2, 3 respectively. Plants were checked up every 15 days and until the end of December showed a good vegetation stage. No flowering was observed. However at the end of March 2010, due to the minimum temperature (-2 °C) occurred in February and March 2010, the plants of all cultivars lost all the leaves completely dried. All the cultivars showed 100% of mortality.

S12.303

Chestnut Genetic Landscape Shape in Spain

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Conservation and utilization strategies require fundamental knowledge about levels of genetic diversity, population genetic structure and effective population size, because they are the key elements that determine a species ability to respond to selection, either natural and artificial procedures. Sweet chestnut (*Castanea sativa* Mill.), widely distributed around Europe, is a multipurpose tree species of great economic importance for fruit and timber. Moreover, chestnut contributes to carbon storage, the landscape structure and the biodiversity conservation, that are relevant factors for environment management especially under climate change. Although there is information about chestnut traditional varieties, up to now, there is not complete information on the genetic diversity of natural populations in Spain. In this study, patterns of genetic variation and structure are assessed within and among natural populations of chestnut in Spain. 17 natural populations were evaluated using 7 microsatellite markers and Alleles in Space (AIS) software. This study allowed to describe the level and distribution of the genetic variability in Spanish chestnut populations, showing a high genetic diversity. Moreover, populations displayed a structure that followed a geographical pattern, indicating in a first approach, a separation between populations from West and east of the Iberian Peninsula.

S12.304

Genetic Resources of the *Corylus* L. Genus to Ukraine

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The different cultivars and ornamental forms of the *Corylus* L. genus are being studied to National dendrological park Sofiyivka of the NAS of Ukraine in the course of more than 20 years. *Corylus* L. genus is represented by 14 species, 120 cultivars and 15 ornamental forms. Amongst specimens *Corylus* L. genus only *C. avellana* L. is native plant - another are introduced. The most interesting Ukrainian cultivars to breeding are: Bolhrad's'ka novynka, Borov's'kyi, Dar Pavlenka, Klynovydniy, Koronchatyi, Loziv's'kyi sharovydniy, Pirozhok, Raketnyi, Serebrystyi, Stepovyi 83, Shedevr and Shokoladnyi. The exercisable cultivars introduced are: Futcuramy, Romana, Trapesund at alias. The best ornamental forms of *C. avellana* are: 'Aurea', 'Contorta', 'Fuscorubra', 'Heterophylla' and 'Pendula'. The most interesting forms of *C. colurna* L. are: 'Fastigiata', 'Nadija', and 'Poltawska'. On the basis of the studies of the other species introduced of the *Corylus* L. genus (*C. americana* Mill. (Marsh.), *C. chinensis* Franch., *C. cornuta* Marsh., *C. heterophylla* Fisch., *C. lacera* Wall., *C. mandshurica* Maxim., *C. maxima* Mill., *C. pontica* C. Koch, *C. sieboldiana* Blume, *C. tibetica* Batalin) the prospects of utilization of these species to Ukraine are considered.

S12.305

Agromorphological Characterization of Traditional Spanish Almond (*Prunus dulcis* (Mill) D. A. Webb) Cultivars

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Twenty-six traditional almond cultivars from "Arribes del Duero" in Central-Western Spain were surveyed and characterized agromorphologically. A total of 49 descriptors, mainly defined by the International Plant Genetic Resources Institute and the International Union for the Protection of New Varieties of Plants were used to describe flowers, leaves, fruits, one-year-old shoots and the tree itself over a two consecutive years. This made possible the unequivocal identification of 25 cultivars. A dendrogram gave a clear separation between the cultivars and showed

existing synonymies and homonymies. This work is an important step in the conservation of genetic almond resources in the province of Salamanca (Spain), which show distinctive and interesting agronomical characters such as high productivity, early fruit maturity and great rusticity.

S12.306

Investigation of Diversity among Five Populations of Native Persian Walnut in Golestan Province of Iran Using Quantitative Morphological Traits

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In order to understand diversity among five native populations of Persian walnut (*Juglans regia* L.) tree this research has been conducted. 96 tree samples studied for 32 different morphological traits (according to IPGRI and UPOV descriptors) such as leaf traits include length and wide of leaf, length and wide of leaflet and number of leaflet, nut traits and kernel traits include size (length, wide and thickness), volume, weight and nut index (husk, hull and kernel), thickness of hull, husk weight, percentage of husk moisture and kernel percentage). Variance analysis of traits showed significant differences ($p < 0/01$) among all traits existed, expect of thickness of husk, number, length and wide of leaflet that showed high diversity among native walnut traits in Golestan province. Difference of traits among locations used means comparison and Duncan test. There are positive and significant correlation between husk nut weight on kernel weight ($p < 0/01$) and negative correlation between husk nut weight on kernel percentage ($p < 0/05$). For genetic distance identification and genotypes grouping, used cluster analysis that Put on 6 clusters. These Genotypes was studied based on quantitative data and UPGMA algorithm. Classification of genotype based on molecular data did not match with their Geographical situations and existence high diversity among and between populations. Using principal component analysis for identifying each trait on existence diversity among the genotypes. The first six components constituted the 84% of variances.

S12.307

Brazilian Cashew Germplasm Bank

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The Cashew Germplasm Bank, located in Pacajus, Ceará State, Brazil, holds 621 accessions, the majority of them being from the species *Anacardium occidentale*. The introduction of plants in the bank, the main source of adequate materials for the development of commercial products, started in the 1950's. Nowadays, the main goals of the cashew germplasm bank are the enrichment of genetic variability of the specie, agronomic and morphological characterizations of accessions, supporting of breeding programs, and documentation and conservation. The accessions were characterized by means of morphological, agronomic and molecular descriptors. The genetic variability contained in the collection allowed the development of early dwarf cashew clones, recommended for commercial planting in Northeastern Brazil since the 1980's until today, besides 132 other early dwarf cashew clones and 40 common cashews still under evaluation. These results directly impact the cashew agribusiness. Furthermore, it made possible the expansion of the early dwarf cashew genetic basis by natural and artificial hybridization with regular cashew genotypes from the germplasm bank allowing a significant increase in the weight and size of nut and almond. Interspecies' hybrids of *A. occidentale* × *A. othonianum* e *A. occidentale* × *A. microcarpum* were also obtained with the goal of inserting anthracnose resistance alleles and desired quality traits to table cashew as well, which are under evaluation. The first genetic map of cashew was elaborated using AFLP and SSR, containing a total of 238 markers. This map is the basis for identification of QTL for application in the breeding program in order to reduce cost and time needed for cashew germplasm bank development of superior genotypes. The passport data were submitted to SIBRAGEN data bank and is available to the scientific community and breeding programs.

S12.308

Morphological Variability of Cashews from the Brazilian Cerrado

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The cashew Germplasm Bank from Embrapa has accessions collected in Brazil. Among these, there is a collection of accessions from Cerrado, a biome with great biodiversity, with the presence of small, tortuous trees being typical. The goal of this work was to characterize 32 accessions of Cerrado's cashew species. The morphological characterization was performed using the descriptors: plant height, canopy diameter, leaf area, fruit weight, pedicel size and weight, main diseases, tree habit, canopy aspects, peduncle and leaf color, brittleness and leaf shape. The *A. occidentale* accessions presented 60% of tall canopy and upright/open habit; 50% of round peduncle; 75% with orange coloration; 80% of young reddish green leaves; 80% of mature green leaves; 60% of obovates and brittle leaves; and 40% oval and leathery leaves. The *A. othonianum* accessions presented: 66% semitall canopy, 54% upright and compact habit; 41% upright and open habit; 66% reddish green young leaves; 79% green adult leaves; 96% obovates leaves; and 75% leathery leaves. The majority of *A. humile* accessions were constituted of dwarf trees with spreading habit; reddish green young leaves; green and leathery adult leaves. The accessions with highest leaf area values were from *A. othonianum*, 89% higher than those from the *A. occidentale*, common cashew. Despite having leaves larger than the others, *A. othonianum* accessions had small peduncles. Symptoms of anthracnose and black mold were observed for all species. The species showed differences for all characters evaluated and also a high variability within them. There is variability in the peduncle shape and the orange coloration predominated; most of young leaves are reddish green, the adult ones being green and obovates, with the exception of *A. occidentale* in which the oval ones predominate. *A. othonianum* and *A. humile* had more leathery leaves, while *A. occidentale* and *Anacardium* spp. had more brittle ones.

S12.309

Genetic Resources of *Butia* (Arecaceae) in Southern Brazil: A Native Multiple Purpose Palm

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Five native species of *Butia* palms occur in Rio Grande do Sul state, Southern Brazil: *B. capitata*, *B. eriospatha*, *B. lallemantii*, *B. odorata*, and *B. yatay*. Local communities consume the fruits; however, the plant is underused if considered its potential as ornamental or as source of new products for food and cosmetic industries. Over the last century natural populations of *Butia* suffered large reductions due to agriculture and urban expansion. To increase the knowledge related to genetic resources of these palms, research has been conducted on ethnobotany, characterization, and processing by Embrapa Temperate Agriculture. Ethnobotanical survey showed that the interaction of people with *Butia* is established since childhood with a relationship of affection and respect. Fruits are used for production of various kinds of food, beverages, and handicrafts. Juices and liquors produced from the fruit flesh are sources of income for small agroindustries. Morphological characterization showed genetic variability for fruit characters, mainly for color, size, shape and acidity. Analysis of chromosome number showed that the five species assessed are diploid ($2n = 2x = 32$). The karyotypes are symmetrical with small chromosomes. Molecular characterization (ISSR and AFLP markers) demonstrated the largest genetic variability in *B. capitata*, which is distributed within rather than between populations. Functional compounds, especially vitamin C and carotenoids, both

with antioxidant activity, were found in the fruit flesh. The seeds contain high oil levels, which are rich in unsaturated fatty acids (oleic and linoleic acids), and with a high percentage of myristic and lauric oil. These high quality oils can be used in different industrial fields such as food, pharmaceutical or cosmetics, to develop new products. Several processing methods were tested and resulted in products such as filling for chocolates, ice cream, and whole juice with full recovery of the total fiber.

S12.310

Preservation of *Dianthus* Genus Genetic Resources by Micropropagation

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The Southeast of the Iberian Peninsula is phylogenetically rich in species of *Dianthus* genus. There are numerous endemic *Dianthus* species located in the province of Murcia (SE of Spain), some of them registered in the Regional Catalogue of Protected Wild Flora. Plant genetic resources provide the raw material for new and better varieties that are adapted to environmental conditions, resistant to diseases or which have high productivity rates. Since carnation (*Dianthus caryophyllus* L.) is the most widely cultivated ornamental crop in Murcia, it could be highly profitable to obtain new varieties including some characteristics of wild *Dianthus* species. Furthermore, to avoid extinction or to preserve genetic characters that could be used in the future, the conservation of endemic species is nowadays considered the ultimate aim of many researchers. In order to guarantee their survival and preserve their genetic characteristics, four wild species gathered in Murcia (*Dianthus anticariensis* subsp. *Saorinii*, *Dianthus broteri*, *Dianthus subbaeticus* and *Dianthus hispanicus*) are being subjected to different studies concerning sterilization, germination, micropropagation, callus induction and plant regeneration. The results are widening our scientific knowledge and providing techniques for the correct preservation of *Dianthus* genus genetic resources in Murcia. The experiments show that significant differences appear in sterilization sensibility and germination capacity between the four species and subspecies assayed. Thus, *D. anticariensis* subsp. *Saorinii* and *D. broteri* show the higher percentage of germination (50-60%), showing *D. subbaeticus* and *D. hispanicus* a percentage minor than 30%. Regarding the micropropagation and callus induction, the assays with *D. anticariensis* subsp. *Saorinii*, using 2,4-D and kinetin as synthetic fitoregulators in MS media enriched with some vitamins, show an habitual behaviour respect to the hormonal balance.

S12.311

Mass Propagation from Nodular Cluster Cultures of *Vriesea reitzii*, an Endangered Brazilian Bromeliad from Atlantic Forest

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The Brazilian Atlantic biome is a hotspot of biodiversity, and holds high genetic diversity and endemic species. Bromeliads are important components of this biome and among them *V. reitzii*, an endemic and endangered bromeliad. Tissue culture techniques comprise efficient tools for the mass propagation of bromeliads. The regenerative *in vitro* morphogenetic route in bromeliads show specific features associated to the induction and development of nodular cultures (NC). In the present work it was studied factors determining the efficiency of this regenerative route in *V. reitzii*. Seeds were excised from mature capsules and then inoculated on glass flasks (200 ml) containing 30 ml of gelified or liquid BM basal medium supplemented with different types and levels of plant growth regulators. The highest rates of NC induction occurred in response to 4 µM NAA (81.8%), and to 0,1 µM TDZ (80.9%). NC subcultured to BM culture medium supplemented with 4 µM IAA resulted in the highest mean value for microshoot number (1.468 microshoots/g). NC induced in the presence of 4 µM NAA, then subcultured to medium with 2 µM NAA and 2 µM 2-iP presented granular texture and high proliferation. GA3 (10 µM) resulted in the synchronic elongation of microshoots. Histological analy-

ses revealed the features associated with the morphogenetic process leading to the induction multiple meristem associated to NC. The morphogenetic route associated to CN induction and development comprises an efficient tool for the mass and fast propagation of endangered or high ornamental value bromeliads.

S12.312

Current Status of *Davidia involucrata* Baill. Wild Populations in China

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CENTRAL SOUTH UNIVERSITY OF FORESTRY AND TECHNOLOGY, NO 498 SHAOSHAN SOUTH RD., 410004, CHANGSHA, CHINA, *Davidia involucrata* Baill (dove tree) originated from tertiary period and now is a relic plant known to the world. It becomes one of the world famous ornamental tree for gardens and parks in many countries because its flowers look like "flying doves". In recent years, the increase of regional developing projects has led to decrease of this ancient and rare plant in nature, especially its habitats and populations. The plant has been listed in the rare and endangered plants in China. *Davidia involucrata* mainly distributed in very special niche habitats in the mountainous region of Youngzi River in China. After exploring and sampling some natural populations, we summarized that the species became endangered plants because of 1) narrow distribution (niche habitat). Dove tree usually grows in the deep areas of the mountain ranges, with warm summer and abundant moisture. 2) Seed abortion. It is very serious and majority of embryos could not develop normally. 3) Difficulty of natural regeneration. The seed of dove tree has a long period of after-ripening and a hard exterior covering. Germination usually occurred after three years, which leads to the significant loss of seed viability. (4) Human disturbance. Habitat loss (logging) and collection of wild seedlings were one of the important reasons to reduce its populations. Dove tree was very sensitive to the change of habitat. The environment change would cause its reproduction difficulty. The number of individuals of dove tree was declined dramatically. Further studies on its protection and habitat restoration should be our priority.

S12.313

Natural Populations of *Davidia involucrata* Baill in Hupingshan Nature Reserve, China

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CENTRAL SOUTH UNIVERSITY OF FORESTRY AND TECHNOLOGY, NO 498 SHAOSHAN SOUTH RD., 410004, CHANGSHA, CHINA *Davidia involucrata* Baill is the only species in *Davidiaceae* with narrow distribution area of West Hunan, east Guizhou and Sichuan, China. To better understand its natural community and propose management strategy, we investigated 3 populations by sampling 20×30 m² area, respectively. The results concluded that: 1) The community vertical structure was simple, which composed of tree layer, shrub layer, herb layer and interlayer plant. Herb layer was better than tree layer and shrub layer. There were 65 vascular plant species in the community belonging to 37 families and 59 genera. In term of life form, 32.3% phanerophytes were on the top, then 30.8% hemicryptophytes, 24.6% chamaephytes, 7.6% therophytes and 4.6% geophytes. The composition of life form was similar to that in subtropical evergreen and deciduous forest. 2) The importance value of *Davidia involucrata* was 70.3, then was *Tetracentron sinense* (60.8). The entire community richness is 65. The Simpson and Shannon-wiener diversity indices in herb layer were higher than that in tree layer and shrub layer. The *Pielou index* (Jsw and Js) in tree layer were higher than herb layer and shrub layer. 3) The composition of *Davidia involucrata* community was that the East Asia Plants was the largest, reached to 27.1 % (16 genera). Next was North Temperate Zone, about 20.3% (12 genera). The component of species have the typical transition characteristics that from the subtropics to the temperate zone. 4) The community of *Davidia involucrata* was scattered in mountain valley where was warm, abundant rainfall, more mist and few sunshine days. Natural regeneration ability of *Davidia involucrata* was weak and seed germination was low. It's indicated that *Davidia involucrata* community in Hupingshan Nature Reserve had not yet developed into its climax and we should protect this natural treasure plant, especially its habitat.

S12.314

Flower Color and Flavonoid Synthesis Gene in Evergreen Azalea

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Flower color trait is one of the most important breeding objectives in evergreen azalea. Azalea flower colors are due to the presence of flavonols and anthocyanins as the major pigments. To know relationship flower color and flavonoid synthesis gene expression, we compared flower color, pigment composition and gene expression in wild species and cultivars of evergreen azalea. Flower color was mostly dependent on pigment composition in wild species and cultivars. Red flower had cyanidin type pigments and purple flower had both cyanidin and delphinidin type pigments. White flower of azalea had no anthocyanin. A real-time PCR analysis showed that transcripts of chalcone synthase (CHS), flavonoid-3'-hydroxylase (F3'H), flavonoid-3',5'-hydroxylase (F3'5'H), and anthocyanidin synthase (ANS) genes were expressed concomitant with anthocyanin synthesis during petal development of *Rhododendron x pulchrum* 'Oomurasaki', which had purple petal. Also, 'Oomurasaki' red flower sport contained only cyanidin type pigments, and abundant of F3'5'H gene was 0.14-fold those of 'Oomurasaki'. Furthermore, flower color in wild species and their hybrid could be explained by pigment composition and F3'5'H gene expression except partial individuals. These results indicated that F3'5'H gene is strongly correlated to delphinidin synthesis in azalea petals, but expression of other flavonoid genes are unclear for anthocyanin synthesis. We are searching factor or gene which controls floral pigmentation.

S12.315

Phenological and Molecular Approach for Flowering in Evergreen Azalea

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For the purpose to study flowering of evergreen azalea, we have observed plant phenology and investigated flowering-related genes using garden planted 'Oomurasaki' (*Rhododendron x pulchrum*). During the flowering in mid-April to early May, shoots begin to sprout from the base of inflorescence and develop. Shoot development slow down in late June, and in early August, apical bud can observe and develop to grow. Genes related to floral induction and initiation have been reported FLOWER-ING LOCUS T (FT), SUPPRESSOR OF OVER EXPRESSION CONSTANS 1 (SOC1), LEAFY (LFY), APETALA 1 (AP1) and TERMINAL FLOWER 1 (TFL1) in *Arabidopsis*. Floral pathway integrator genes (FT and SOC1-like genes), floral meristem identity genes (LFY and AP1-like genes) and floral inhibitor gene (TFL1-like gene) were isolated from shoot apical part tissues (apical meristems with stem) in 'Oomurasaki'. When the expression of floral pathway integrator genes in stems and leaves was examined, the gene expression was detected in late June. After strong expression of floral inhibitor gene on apical parts in late July, expression of floral meristem identity genes was detected strongly in early August. Expression of homologues related to flowering genes suggest that these genes participate in floral induction and initiation in 'Oomurasaki'.

S12.316

The Effects of Time and IBA on Rooting of Cadaman Cuttings

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Cadaman (*Prunus persica* × *Prunus davidiana*) is a one of the important rootstocks of Peach. In this study, the effects of five IBA levels (0, 1000, 2500, 3500 and 4500 ppm) and two time (December and January) on rooting of Cadaman hardwood

cuttings were evaluated. Results showed that cuttings type, IBA levels and time were significantly affecting on rooting ($P=0/01$). Also, the highest rooting (56.6 %) was related to 4500 ppm IBA in December.

S12.317

Studies on the Stone Cell Content in Pear Fruit

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Stone cell content of 304 pear accessions of 5 species (*Pyrus* L.) preserved in National Germplasm Repository of Apple and Pear were measured by gravimetric method, including 117 white pear varieties (*P. bretschneideri* Rehd.), 89 sand pear varieties (*P. pyrifolia* (Burm.f.) Nakai), 35 ussuriensis varieties (*P. ussuriensis* Maxim.), 8 sinkiang varieties (*P. sinkiangensis* Yü), 16 common pear varieties (*P. communis* L.) and 39 *Pyrus* hybrid varieties. The results showed that the mean value of stone cell content in total varieties is 0.684 g/100g, ranged from 0.010 g/100g to 6.678 g/100g; mean value of stone cell content in *P. bretschneideri* Rehd., *P. communis* L., *P. pyrifolia* (Burm.f.) Nakai., *P. ussuriensis* Maxim. are 0.462 g/100g, 0.524 g/100g, 0.552 g/100g, 1.887 g/100g respectively. Based on the stone cell data, using SAS6.0 cluster analysis, *P. pyrifolia* (Burm.f.) Nakai., *P. communis* L., and *Pyrus* hybrid are similar, whereas, *P. ussuriensis* Maxim is quite different from other species. 8 varieties with very few stone cell content were screened through 3 years' evaluation, they are 'Zaosu', 'Akaho', 'Deshengxiang', 'Saint Maria', 'Siyangqingli', 'Conference', 'Bartlett' and 'Qingkui'.

S12.318

Investigation of Qualitative and Quantitative Traits on Progenies from Control Crosses between Iranian Seeded and Seedless Grapevine

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Grape is a major crop worldwide. Iran has rich grape germplasm and has been known as a part of the earliest domestication region of grapes. Controlled crosses of grapevine are well known and have many advantages for cultivar improvement. Iranian grape breeding program was started with evaluating of 90 native cultivars. Crosses were done in spring of 1999 and 381 progenies originated from 22 cross combinations of seeded × seedless grape cultivars were evaluated during 2006 and 2007, on five year old seedlings. Data were analyzed for simple statistics, principal component (PC) and cluster analysis. PC analysis reduced the original 21 characters to six PCs. PC1 (berry characteristics), explained 31.39% and PC2 (seed characteristics) accounted for 14.34% of the total variation. The dendrogram derived by the average distance of k-means method revealed six clusters. Results showed that there is a high level of morphological diversity for most of the traits. Superior progenies were selected to release new cultivar or to be employed for back crossing. Small coefficients of variation (CVs) were observed for some characters including Total Soluble Solid (TSS), time of ripening. This data would be used to plan new grapevine breeding program.

S12.319

In situ Morphological and Agronomical Characterization of Wild Olives (*Olea europaea* subsp. *europaea* var. *sylvestris*)

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In situ evaluation of morphological and agronomical variability was carried out on 48 wild genotypes from four different sites from two provinces of Andalusia (Cadiz and Jaen), Southern Spain. High correlation values were observed between morphological ($r: 0.56-0.81$) and agronomical measures ($r: 0.50-0.78$) taken in two different prospecting campaigns (2007/08 and 2008/09). For all the characters under study high levels of variability were found, being higher the values within each site than between sites. Only three characters, leaf blade width, fruit length and stone length/width ratio showed significant differences between sites. This is in complete agreement with the results obtained by means of SSR markers where higher levels of genetic variability were also found within wild olive populations than between them. The results of morphological characterization do coincide with previous studies carried out in wild olive trees. As expected, fruit sizes were in average, smaller and with less percentage of olive oil than the values found in fruits of olive cultivars. However, it is worth mentioning that individuals with fruit weights (1.3 g) and olive oil percentage in dry matter (33.8%) comparable to the values found at some olive cultivars, were also found. Molecular data obtained by SSR markers together with *in situ* morphological and agronomical characterization of wild olive trees confirmed the high diversity found within the wild populations. These results indicate the richness of wild olive genetic resources in Andalusia.

S12.320

Genetic Diversity of the Fig (*Ficus carica* L.) Collections in Shandong Province

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Shandong province has a long history for fig (*Ficus carica* L.) cultivation with abundant fig resources. Fig trees are usually distributed by vegetative propagated plants, it is possible that the genetic diversity of figs would result from vegetative mutation and sexual reproduction. There are a lot of accessions of fig in Shandong. However, the genotypes of many of them are still unclear. We collected 58 accessions from 8 areas in Shandong for assessment of genetic identity. RAPD analysis was employed to distinguish genetic difference and the 58 accessions were classified to 23 genotypes. and based on UPGMA (un-weighted pair group method with arithmetic) analysis, the dendrogram of these genotypes was constructed. Additionally, two RAPD markers, S20821640, S21041614, linked to the base cordate/ base non-cordate of leaf shape were selected and the former one was converted into SCAR marker, which could be used for identification of fig resources in dormant season. Furthermore, it was found that plant hardiness and fruit quality varied greatly among these accessions.

S12.321

Genetic Diversity and Phylogeography of *Pyrus calleryana* in Zhejiang Province of China

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Pyrus calleryana, the most ancestral wild resource of *Pyrus* L. and an important rootstock resource, is widely distributed over the South of China. Because of its heat and waterlogging resistance, *P. calleryana* was considered as an important resource to overcome the pear cultivation problem brought by global warming. In this study, we used SSR makers and haplotype analysis to investigate the genetic diversity and phylogeography of *P. calleryana* from 46 individuals over 6 populations in Zhejiang Province, China. It was showed that the six microsatellite *loci* were polymorphic and the number of polymorphic alleles ranged from 4 for KA4 to 21 for BGT with an average of 11.5 per *locus*. UPGMA cluster analysis showed that all of the individuals could be divided into 7 groups in the coefficient of 0.83 and the genetic distance was correlated positively with the geographic distribution among populations. In phylogeographical analysis, 9 haplotypes were detected using the intron of accD. All of the populations had their unique haplotype patterns and the haplotype diversity was not related with the population size. The unique haplotypes in some regions were considered as derived haplotypes because all of them located on the tips of the TCS network.

S12.322

Study on Selection of New Strains of Seedless and Few-Seed 'Gonggan'

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'Gonggan' (*Citrus reticulata* Blanco) was once the cultivar of Guangdong province. Due to its excellent quality, it had developed into the main citrus cultivar in Guangdong in recent years. However, the fruit quality and further development of 'Gonggan' are limited for its excessive seeds (more than 10 seeds per fruit in general). 1 The character and quality of fruit One seedless strain and one few-seed strain of 'Gonggan' were selected through bud mutation for many years. Seedless 'Gonggan' had ≤ 3 seeds per fruit weighted more than 100g, and few-seed one had ≤ 3 seeds per fruit weighted more than 105g. Total soluble solid (TSS) contents of the two new strains were all over 11.0% and SSC/TA (total acid) rates were up to 20. Although the fruit of new strains weighted less than seedy 'Gonggan', they possessed better comprehensive quality and the yield was the same to seedy 'Gonggan'. The demonstration of second-generation is in progress. Seedless Gonggan has been approved by experts through field evaluation of variety. 2 Genetic difference analysis Pollen fertility, development of megaspore and embryo were studied in order to explore the genetic traits of seedless and few-seed 'Gonggan'. The results indicated that germination ability and viability of pollen were significantly lower than that of normal 'Gonggan' since embryo abortion was the main reason that leads to seedless citrus. In addition, nine primer combinations with high polymorphism and distinctiveness were selected from 64 primer combinations and according to AFLP analysis it was conducted that seedless, few-seed and normal 'Gonggan'. The results indicated that 181 specific bands were amplified between seedless and normal 'Gonggan' and 175 specific bands were amplified between few-seed and normal one through nine primer combinations, which showed that there were genetic differences among these three citrus cultivars on molecular level.

S12.323

The Preliminary Study on Pollen Characteristics of Litchi Gemplasm

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Litchi is a typical cross-pollinated plant which can be fertilized through pollination and seed setting. The pollen characteristics of different litchi germplasm resources are the theoretical basis on pollination practice, and the effective number of pollen germination is a prerequisite for the success of fertilization. From the National Fruit Germplasm-Guangzhou Litchi Nursery, Pollen number of 108 litchi germplasm and pollen germination rate of 76 germplasm were measured, and 5 of these were studied on pollen number and germination rate of male flowers in different developmental stages. The results showed that: 1, The pollen number has a significant difference between germplasm, which was a continuous change, 3000-5000 pollen grains in a mature anther can be used as the characteristics of litchi pollen; 2, There is a difference among Litchi germplasm in pollen germination rate, 20% -40% of the pollen germination rate can serve as the litchi pollen germination rate characteristics in a bad year; 3, The pollen number and germination rate were different between different developmental stages of male flower. The development of pollen within the same anther is not synchronized and has a larger difference. There is different in pollen germination rate, but smaller difference. This study found that there has an effective number of the highest pollen germination during the more than half of Filament and before the anther cracking, which is the best time of pollen collection. The study provides a theoretical basis for selecting pollination germplasm of a large quantity of pollen and higher pollen germination rate in cultivation, as well as the best period of pollen collected in artificial breeding.

S12.324

Studies on the Processing Characteristics of Apricot Cultivars Cultivated in Xinjiang

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In order to research the processing characteristics of apricot cultivars cultivated in Xinjiang and to provide basic information for exploring and using apricot resources. The works that we carried on are as follows: (1)The 18 apricot cultivars showed that their average fruit weight were 14.4 g-52 g, juicing ratio 44.6%-62%, soluble solids content (SSC) 13.2%-25.4%, titrable acid content (TAC) 0.67%-1.59%, Vc content 26.7-146.5 mg/kg, the ratio of soluble sugar content was between 6.64% and 15.52%. Based on our evaluated results we would suggested that for juice processing the cultivar should have average fruit weight>40g, juice ratio>58%, SSC>21%, Vc>100mg/kg, The ratio of soluble sugar content to titrable acid content ranged from 8.31 to 12, and soluble sugar content was between 7.81% and 15.11%. (2)The 14 apricot cultivars showed by using headspace solid phase microextraction and GC-MS that 208 aromatic components were identified, including 80 kinds of esters, 25 kinds of aldehydes, 21 kinds of ketones, 39 kinds of alcohols, 27 kinds of alkyl substances, (Z)-; D-limonene; β -Myrcene; ethyl butanoate; β -cis-ocimene. Main volatile components and their relative contents were differences among these cultivars, and there were no significant difference between the Northern China group and the Central Asian group varieties. (3)The 17 apricot cultivars showed by total antioxidant activity, hydroxyl radical scavenging and DPPH• radical scavenging that they had strong antioxidant capacity and significantly difference; All indexes of antioxidant capacity and total polyphenol content were significantly correlated, so polyphenols should be the most important material basis of antioxidant capacity.

S12.325

Evaluation of Pear Varieties in Hungarian Gene Banks

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There are three places in our country, in Hungary, where pear varieties are being observed in gene banks: Újfehértó, Keszthely and Mosonmagyaróvár. One of the genebank was established in 1975 in Újfehértó, which is located in North-east Hungary. There are currently 454 pear varieties tested there. The second gene bank was planted in 1981 in Keszthely with 256 pear varieties. Keszthely is on the West part of the Lake Balaton. A new pear collection was set up in 2000 in Mosonmagyaróvár. The following characteristics are being examined from the beginning till now: - phenophases, phenological features of pear varieties - beginning- main - and the end of the bloom, length of the blooming period, - fertility examination, fruit set from open pollination, seed content of fruit, - ripening (picking) time - fruit morphology characteristics (weight, size, shape, etc.) - quality parameters of the fruits (dry matter of fruit, acidity, etc...) - frost tolerance of buds, - frost damage during the blooming time - sunburn damage on fruit. Correlation was determined on the base of characteristics of pear varieties and the climatic parameters of the different locations to express the genetically determined values of the varieties under different ecological circumstances. The differences and the similarities were noticed between the old and the new foreign and Hungarian origin pear cultivars.

S12.326

Genetic Divergence of Sour Cherry (*Prunus cerasus* L.) Montmorency Clones

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To determine the overall degree of variability and detect similarities among 12 clones of sour cherry cultivar Montmorency, 23 characteristics (phenologic properties, fruit set, fruit, leaf and chemical characteristics) were studied in two years. Clones were selected from commercial orchard of PKB "Voćarske plantaže" Boleč, locality Begaljica, near Belgrade, Serbia. Agronomic evaluation of the clones revealed considerable diversity for almost all characters studied. In order to investigate breeding response and genetic relationships between studied clones and to help the breeder to meet the selection objectives, phenotypic and genetic correlation matrices were calculated. Very significant or significant values of both genetic and phenotypic correlation coefficient were determined among fruit set and fruit harvested, as well as between fruit, leaf and chemical properties. For the assessment of the clones genetic divergence mean values for the most important traits were used. By hierarchy cluster analysis, all Montmorency clones were distinguished into three distinct groups. First cluster consisted of six clones (3, 6, 4, 11, 8 and 10), second one included only two (9 and 14), while the third one had four clones (5, 7, 12 and 13). Despite of the significant differences among clones the number of fruit set and fruit harvested made a clear separation between the clusters.

S12.327

Haplotype Diversity of Iranian Grapevine Cultivars and Wild Genotypes

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This study was conducted to assess the haplotype diversity and genetic relationship between wild and cultivated of Iranian grapevines by using 10 chloroplast microsatellite primers. Results showed that among analyzed cpssr *loci* only *ccmp3* and *ccmp10* were polymorphic within cultivars and only *ccmp3* was polymorphic in wild grape individuals. The size variants of both *loci* combine in a total of 4 different haplotypes. All the 4 haplotypes are displayed in the cultivars while only 2 are presented in wild grapes. Sultani or Keshmeshi Bidaneh cultivar has the haplotype III that there is not this haplotype among the wild grapes of studied regions. Concerning to existence of both haplotypes I and II in the number of Iranian cultivated and wild grapes, it is possible to consider that wild grape are ancestor of some of our native cultivars.

S12.328

Seedless Mechanism on Extraordinary-Late-Maturing Fruits of *Citrus reticulata*

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Citrus productive districts of China locate mainly in the Yangtze River Valley where are rich in varieties and high in yields. Maturity periods of these oranges are basically concentrated between October and December which directly affects the price stability of the citrus market. From this view, looking for and cultivating extraordinary late-maturing and seedless or off-season varieties of citrus become an important research direction in the world. During years of observation we discover an especially late-maturing and seedless tangerine, called JZT(P)-1, belongs to the species of *Citrus reticulata*, which is a promising new resource. Then, a serious experiments of this tangerine are carried by ways of consecutive paraffin sections, scanning electronic microscope and pollen germination, etc. Results show that

JZT(P)-1 blossoms from April to May. There are massive pollens in its anthers. However, the morphology of most pollens is abnormal which can be observed under the SEM. No pollens germinate on the specific media. In nucellus, more than one of archesporial cells develop simultaneously or successively. The normal meiosis of the megaspore mother cell is not observed. The normal seed per carpel is 0.13, and fruit seedless rate arrives at 99.87%. Few seeds which have normal appearance are developed from the apomixis. Fruits of JZT(P)-1 mature during March to April next year, whose are almost seedless but succulent and tasty.

S12.329

Properties of Some Persimmon (*Diospyros kaki* L.) Types Selected from Black Sea Regions

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The study was started in 1996 by collecting 44 selected persimmon types from Blacksea Regions, reproducing them and setting up orchards in the Hazelnut Research Institute. Phenological and pomological analyses were carried out between 2001-2003 in order to put forward the differences of persimmon types in terms of development, yield, and quality; to determine the types for adaptation experiments which will be carried out in different districts of Turkey. Consequently, persimmon types that can be new cultivars were identified. As a result of phenological observations and pomological analyses, weight ranked methods and statically evaluations; 52-TH-08, 08-TH-01, 52-TH-10, 14-TH-01, 53-TH-07, 61-TH-02, 55-TH-01, 08-TH-15, 08-TH-11 and 52-TH-01 types were identified to be new cultivars. In the sensory taste controls made during the harvest period, nearly of the types are determined to be "good", some of them are slightly sour and few of them are sour. The types whose pulps don't have any brown marks have generally a sour taste. The brown marks weren't seen in some of the types; they were seen in a low rate in some types; however, they appeared in a high rate in some types. In the harvest period, it was determined the taste of the hard eatable fruit was generally good and there were brown marks in the pulp.

S12.330

Evaluation of the Natural Virus Infections in Sweet Cherry Genbank at the RIFG, Pitesti, Romania

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The viruses are the diseases that produce the important damages in fruit species of *Prunus* genus. Study conducted at the RIFG, Pitesti shows presence of some virus infections in the Cherry Roumanian National Genbank. The tests have been made on 56 cherry genotypes in natural infection conditions, using leaves as biological material, collected from an orchard of 25 years old. TAS-ELISA, DAS-ELISA and AGRISTRIP have been used as test methods, to mark presence of virus infection with ACLSV, ApMV, ArMV, CLRV, PDV, PNRSV, RpRSV, SLRV, TBRVC and PPV. Tests show the presence of PPV in 33% from sweet cherry genotypes, TBRV in 30%, RpRSV in 44%, CLRV in 4% and ArMV in 2%, respectively. Presence of PPV infection was confirmed by TAS-ELISA and AGRISTRIP test methods, also. Didn't was detected infections with other viruses as ACLSV, ApMV, PDV, PNRSV and SLRV.

S12.331

Pollen Fertility Comparison of Some Sweet Orange (Varieties)

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Some varieties of citrus with male sterility can bear seedless or less seedy fruit due to induced parthenocarpy with abortive pollen. So, it is important to investigate the fertility of citrus in the field of breeding and cultivation. We investigated the vi-

ability and germination rate of 10 seedless citrus varieties (2-1-50, 2-3-26, 3-1-68, 3-2-46, 4-1-30, 4-1-48, 4-2-42, 4-3-23, 4-3-56 and 4-3-57) compared with that of Jincheng and Xianfeng Orange using POX staining method and aqr nutrient medium pollen germination (sucrose 15%). The result showed Jincheng and Xianfeng orange had the highest viability and germination rate which were 34.4%, 12.9% and 28.2%, 14.3%, followed by varieties or stains with partly abortive pollen and the other two seedless varieties (4-3-56 and 4-3-57). Pollen of Xianbai seedless orange had the lowest viability and germination rate which was 5.2% and 0%. This research showed less seedy citrus varieties and abortive pollen had obviously related.

S12.332

Breeding of a New Early Ripening Orange Cultivar 'Yuzao'

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'Yuzao' orange (*Citrus sinensis* L.) was an early-maturing mutation radiation-induced from seeding orange 'Tongzigan' in Chongqing, selected by Fruit Research Institute, Chongqing Academy of Agriculture Sciences through more than 30 years. It's named 'Yuzao' Orange by Chongqing Crop Variety Approval Committee in 9th Nov, 2009. It has excellent characteristics such as: (1) Early maturing. 'Yuzao' Orange matures in first twenty days of November in Jiangjin, 7 days earlier than 'Tongzigan'. (2) High quality. The average fruit mass is 163.5 g. Fruit with palatable favor is from orange-yellow to salmon pink. The fruit has an edible part of 77.35 % and 63.42 % juicy rate. The soluble solid content is 10.9 %. There are 9.45g total sugar, 0.72g acid contents and 45.71mg vitamin per 100ml juice. The soluble solids to acidity ratio is 15.1. (3) Significant Less seeds. The average seed number is 6.5, significantly less than that of 'Tongzigan' 24.3. (4) Early bearing, stable high yield and easy planting. In the second year after top-grafting, the yield is 1487.4 Kg per 667 m² under common manage in Jiangjin, which is 22% higher than 'Tongzigan'. In Sichuan Province, 'Yuzao' Orange tree on trifoliate orange rootstock bearing from 3 years after planting. The yield per 667 m² is 1200 and 2620 Kg after 4 and 6 years respectively. (5) High regional adaptability. The adaptability regional trial showed that the most suitable regions for cultivation are lower than 400 meter above sea level in South Sichuan and Chongqing Three Gorges Reservoir Area, with Jiangan county as the representative. Central Sichuan Province with Zizhong county as the representative is suitable region for cultivation. West Sichuan Province with Nanchong county as the representative is sub-suitable region for cultivation.

S12.333

Genetic Analysis of Resistance to Race 2 France of *P. xanthii* of Wild Melon Material Yuntian-930

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Melon is an important horticultural crop cultivated World wide and has high economic value. Cucurbit powdery mildew is one of the most destructive diseases in all parts of China melon production, both in field and in greenhouse, seriously reduced the quality and yield of melons and cause a great losses. Breeding resistant varieties is an effective way to control powdery mildew of melon. Yuntian-930(P1), is a wild Melon material which was collected and propagated by our research group through many generations and it is resistant to powdery mildew. For find out inheritance of powdery mildew resistance of this wild material, another cultivated melon material "Hualaiishi" (P2), susceptible to powdery mildew, was used to cross with it. All the plant materials of Six generations of P1, P2, F1, BC1, BC2 and F2 were used to study the inheritance of resistance to race 2 France of *P. Xanthii* By joint analysis method of multiple generations. The result showed that the inheri-

tance of resistance to race 2 France of *P. xanthii* of wild melon Yuntian-930 fitted two pairs of additive-dominance-epitasis major genes plus additive-dominant-epitasis polygene model (E model). This result indicated that the powdery mildew resistance of Yuntian-930 was controlled except by major gene and also affected by more minor-gene, was not identical with other report on melon powdery mildew resistance.

S12.334

A Review of the Fruit Germplasm Resources of Iran

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From the point view of fruit genetic resources and diversity, Iran is one of the most significant and unique countries in the world. Many fruit species have been basically originated or domesticated in Iran. Germplasm of those species of greater economic importance, namely *Malus* spp., *Pyrus* spp., *Cydonia* spp., *Crataegus* spp., *Sorbus* spp., *Amygdalus* spp., *Prunus* spp., *Castanea sativa* L., *Pistacia* spp., *Juglans regia* L., and *Punica granatum* L. have been identified as very useful for further improvements of the major fruit trees, e.g. breeding programs (disease resistance, cold and drought resistance), as sources of local foods, or appropriate rootstocks. The fruit trees include *Crataegus* sp., *Cydonia oblonga*, *Diospyros lotus*, *Ficus carica*, *Malus orientalis*, *Mespilus germanica*, *Prunus cerasifera* ssp. *macrocarpa*, *Prunus spinosa*, and *Punica granatum* are existed as wild form in the forests. The areas should be considered as the important evolutionary centers for fruit trees and should be taken into serious consideration for discussions about fruit tree evolution. The conservation of genetic resources of these fruit spp. had been undertaken traditionally by gardeners from long time ago and later several national research institutes started to collect and to conserve the local cultivators in field bank (*ex situ*). Matters regarding their distribution within the provinces, nomenclature, characteristics, uses of, and knowledge about the germplasm of those species of greater economic importance are discussed.

S12.335

A Survey on *Prunus* Subgenous *cerasus* Species Naturally Growing in Iran

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A rich gene pool is prerequisite in fruit breeding programs for obtaining cultivars with appropriate traits such as high yield, good quality, and resistance to stresses, or for dwarfing rootstocks to establish high density commercial orchards. Besides commercial cultivars and autochthonous varieties, relative wild species are valuable gene resources for these purposes. Iran contains significant plant biodiversity for different fruit species, including those from the genus *Prunus*, specially the subgenus *cerasus*. A study has been started on Iranian wild *cerasus* subgenus species in order to evaluate their capacity for introducing into cherry breeding programs. These species wildly existing in Iran include: *Prunus avium* L., *P. cerasus* L., *P. mahaleb* L., *P. incana* Pall., *P. microcarpa* Boiss., *P. brachypetala* Boiss., *P. pseudoprostrata* Pojark., *P. turcomanica* Pojark., *P. chorassanica* Pojark. (Iranian endemic) and *P. yzadiana* Mozaff. (Iranian endemic). These species are various in different traits that can be used as rich gene resources for developing new cultivars or rootstocks. In this report, some main characters of these species as well as their natural distribution in Iran is presented.

S12.336

Uruguayan Native Fruits Provide Antioxidant Phytonutrients and Potential Health Benefits

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Consumer awareness about health beneficial effects of secondary metabolites from

fruits has increased interest in fruits with high levels of these compounds. Globally, non-traditional fruits are generating growing interest for their nutraceutical potential. Uruguay is a primary center of origin of several species of fruits with unique flavor, aroma, appearance and nutritional properties. Those traits have led to their domestication and development as commercial crops. Involved species include those belonging to family: 'Guayabo del Pais' (*Acca selowiana* (Berg.) Burret); 'Pitanga' (*Eugenia uniflora* L.); 'Araza' (*Psidium cattleianum* Wis), Guaviyú (*Myrcianthes pungens* (Berg) Legr.); and 'Quebracho' (*Acanthosyris spinescens* Griseb, *Santalaceae*). The objective of this work was evaluate total polyphenols (TP) content, antocyanins and the free radical scavenging activity (antioxidant capacity) of native fruits in regards of nutraceutical potential. The TP was determined using the Folin-Ciocalteu method. Total anthocyanin content was measured using the pH differential assay. The antioxidant capacity (AA) was evaluated by the reduction of the radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) and expressed as percent of DPPH inhibition. 'Guaviyú' showed a range between 158–349 mg TT/100 g fresh weight; 19–75 of AA; 51–132 mg anthocyanins/ 100 g fresh weight. Values for 'Pitanga' were 252–575 mg TP/100 g; 14–34 AA; 83–110 mg of anthocyanins. 'Araza' gave values ranging between 132–644 mg de TP/100 g; 33–68 de AA and 12–17 mg of anthocyanins. 'Guayabo del Pais' showed a range 129–648 mg de TP/100 g and 24–82 for the AA. This study indicated that Uruguayan native fruits have the potential as valuable sources of antioxidant components. Data illustrated interesting variability both at the intra-species level as well as among different species under study, meaning a significant input to the breeding of commercial varieties derived from native Uruguayan germplasm.

S12.337

Analysis of Morphological Traits of Four Iranian *cerasus* Subgenus Species

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Iran contains significant natural resources of germplasm for different fruit species, especially for those from the genus *Prunus*. Cherries and their wild relatives, classified within the *cerasus* subgenus in *Prunus* species, are among the important plant species distributed in temperate climates in Iran. Quantitative and qualitative traits of vegetative parts and seeds, including 38 traits, were used for categorizing some *cerasus* subgenus genotypes. For this purpose 28 wild *cerasus* subgenus genotypes from eight regions of Iran were studied. These genotypes consisted of *P. avium*, *P. mahaleb*, *P. incana*, and *P. microcarpa* Species. Factor analysis showed that tree height, leaf area, leaf shape, length of petiole, length of seed, seed weight, shape of seed tip and seed thickness were characteristics constituting the main factors. Effective characters were categorized within seven factors and accounted for 85.71 of total variance. Cluster analysis was performed using these seven factors and genotypes in the distance of 19 divided into four groups consisting of the defined species. In the distance of 10 there were eight smaller groups, resulting from intra-species differences between plants collected from different regions, such that genotypes of same species from one region often categorized with each other. *Prunus mahaleb* genotypes showed high variance in their characters, indicating the high biodiversity of these species in Iran.

S12.338

Numerical Classification and Principal Component Analysis of Fruit Characteristics in *Malus crabapple* Germplasm

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18 morphological characters were analyzed using a numerical taxonomic method in the 48 *Malus crabapple* germplasm and. Results from Q cluster analysis revealed that all the genotypes were classified into 8 groups based on the size and the shape

of fruits. The principal component analysis showed that 18 characters were integrated into 6 principal components and their additive contributing rate came up to 87.2 %. This paper suggested that the length of the middle whorl of fruits should be used to judge fruit size. In the classification system, fruit size followed by fruit shape is the most important characters.

S12.339

Fruit Physicochemical Characteristics of Some European Pear (*Pyrus communis* L.) Chance Seedling Genotypes Grown under Tehran, Iran Environmental Conditions

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In order to evaluate genetic diversity of some European pear (*Pyrus communis* L.) chance seedling genotypes this research was conducted during 2009 and 2010 growing seasons. Six mature European pear genotypes were used. Genotypes were grown on their own roots in the Asian pear Collection Orchard at Tarbiat Modares University, Iran. Some physicochemical fruit characteristics were monitored based on the descriptors of International Board for Plant Genetic Resources (IBPGR). Measurements were included fruit length, width, length to width ratio, fruit pedicel length, fruit color, fruit fresh and dry weight, fruit volume, fruit firmness, total soluble solids (TSS), titratable acidity (TA) and pH. Results showed significant differences within the studied genotypes in the most fruit characters ($P < 0.05$). In addition, it was a diversity within the measured characters in fruit length (ranged from 6.99 to 9.82 cm), fruit width (4.69 to 7.17 cm), fruit length to width ratio (1.23 to 1.59 cm), fruit pedicel length (2.2 to 3.73 cm), fruit color (green to yellowish green and green to red with L^* value 60.47 to 74.08 and H^* value 80.06 to 86.95 and C^* value 46.16 to 53.43), fruit fresh weight (80.5 to 308.9 g), fruit dry weight (4.52 to 5.36 g), fruit volume (86.33 to 270.7 cm^3), fruit firmness (1 to 2.37 Kg/cm^2), TSS (14.9 to 17.6 °Brix), TA (0.18 to 0.4 % malic acid), TSS/TA (40.49 to 92.66) and pH (3.9 to 5.3). In conclusion, two genotypes in terms of fruit taste, color and appearance and other measured characters showed better performance than other studied genotypes. These selected genotypes will use in the Tarbiat Modares University pear breeding program for further assessments and selections.

S12.340

A New Line of Valencia: 'Changshou no.1'

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The new line of valencia 'changshou No.1' (*Citrus sinensis* L.), with large fruit and less seeds, selected from grafted-valencia in Changshou, Chongqing. The ovoid spring leaf of the new line is averaged 8.86cm long and 4.68 wide, significantly larger than that of valencia, which is averaged 8.32cm long and 3.70cm wide. The major axis and minor axis of stomata in the new line is 8.95 μm and 7.41 μm , smaller than that of valencia 10.71 μm and 8.35 μm , and there are 6.6 and 4.0 chloroplasts in the stomata of the new line and valencia, respectively. DNA polymorphism of The new line (*C. sinensis* cv.) and valencia were determined by inter simple sequence repeat (ISSR) molecular markers. The results also showed that there were genetic diversity with primer 812, 834 and 841 in genomic DNA between the new line and valencia. The new line has excellent characteristics such as: (1) Large fruit. The fruit averaged 210g is larger than valencia fruit averaged 160g. (2) Less seeds. The average seed number is less than 1.0 per fruit, significantly less than that of valencia. (3) High quality. The total soluble solid content is 10.6 %. There are 9.4g total sugar and 0.94g acid contents per 100ml juice. The total soluble solids to acidity ratio is 11.3. (4) Early maturing. It matures in the last ten days of March or the first ten days of April, 15 days earlier than valencia.

S12.341

Genetic Diversity in Ancient Fruit Tree Germplasm from Southern Italy

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Because of eco-geographical and historical reasons, Italian fruit tree genetic resources patrimony is rich and strongly linked to the traditions of the territory. A loss of local ecotypes could lead to the loss of their social and cultural value, as well as to the erosion of interesting agronomic and adaptive traits, useful for breeding programmes. The aim of this study is to investigate the genetic diversity and the relationship of a set of old *Malus domestica*, *Pyrus communis*, *Prunus domestica* and *Prunus persica* varieties and local ecotypes from Southern Italian Regions (28 from Campania, 9 from Calabria and 27 from Basilicata), chosen within a larger group because potentially suited for re-introduction into niche markets. We also want to expose possible synonyms and homonyms, with the aim to optimize germplasm collection management. Eighteen apple derived SSR couples of primers were used to characterize apple and pear accessions; 17 peach derived SSR to characterize peach and plum accessions. Most of apple derived SSR were successfully transferred to pear, revealing polymorphic SSR *loci* in the pear genotypes analysed, while only 10 on 17 peach derived SSR gave amplification in plum cultivars. The number of alleles per primer pair in the cultivars ranged from 2 to 11 (mean between 3 and 5,375) for diploid species and from 4 to 19 for the hexaploid *P. domestica*. All genotypes showed a unique fingerprint ('Mastrantuono' and 'Agostinella' for pear and 'Ciccio Petrino' and 'Silvette' for peach differed only for one *locus*). While plum and peach UPGMA dendrograms look mostly clustered according to geographic distribution, apple and pear show more complex organization. For all the diploid accessions analysed, the mean observed heterozygosity was comprised between 0.388 (peach) and 0.521 (pear); mean F-index was comprised between 0.245 and 0.398, indicating a high level of inbreeding.

S12.342

The Breeding Programme for Late Flowering, Early Bearing, High Yielding, Blight Tolerant Walnut Varieties (*Juglans regia* L.) Producing High Quality Nuts

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The Turkish walnut populations have important genetic variability consisting of more than 8.5 million natural hybrids on their own roots. These walnut trees are an important source of genetic diversity for *J. regia* L. Turkey climatic conditions is difficult for walnut growing; extremely late spring frost. Therefore nut production is still fluctuant quantitatively and qualitatively. The main problem is almost always early leafing. The environmental adaptations of Turkish national walnut cultivars are often poor due to early bud break and sensitivity to walnut blight and anthracnose. Therefore it is necessary to carry out a breeding programme for developing new cultivars in Turkey. The aim of this national breeding programme was to found new types combining lateral fruitfulness, late leaf break, resistance to pathogens (*Xanthomonas juglandis* and *Gnomonia juglandis*), regular yield, and high nut quality. Franche, Turkish and Californian walnut varieties (Franquette, Şebın, Akça 1, Hartley) with late leafing and high nut quality were used as female parents. These walnut varieties were crossed with Turkish walnut varieties and types (Akça 2, 60 NF 81, Maraş 12, Sütyemez 1, Sütyemez 2, 19 OG 01) that are lateral bears and very productive with early fruiting in this breeding programme. 13 crosses have been made and 1850 hybrids have been observed. First selection was made according to late leafing in selection plots on their own roots. . 22.45 % of this selected seedling ranged for medium late to very late leafing. This selected seedling with late leafing planted in Tokat/Turkey for further observations.

S12.343

Cluster Analysis of Vineyard Peaches Genotypes Suitable for Generative Rootstocks

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During 2007-2008 in 17 vineyard peach genotypes seed germination and some seedling properties are determined with a single aim to select mother plants for generative rootstock production. Most genotypes had good seed germination (over 60%) and satisfactory seedlings viability (over 40%). In the seedlings population differences in vigor as well as the uniformity of properties were established. Based on a mean value of all examined properties, dendrogram of phenotypic differences was constructed by using Ward's procedure. Vineyard genotypes are connected in different ways, which show the existence of numerous hierarchical levels. Two distinct groups were separated in the dendrogram with numerous subgroups. In the first group four subgroups were established, while second group consisted of two subgroups of similar genotypes. Separation between the genotypes in groups was done according to seed germination and seedlings vitality, while seedlings traits influenced forming of subgroups. For the rootstocks production as the most promising can be recommended vineyard peach genotypes with labels II/3, II/10, II/16, II/22 III/27 and XIX/29. Those genotypes had good seed germination, and its seedlings had good vitality and moderate vigor.

S12.344

Manual Cross-Pollination, Fruit Set and Development of Pear Fruits

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The lack of rootstocks compatible to pear in Brazil requires the application of classic genetic techniques. Therefore, for obtaining and selecting of suitable material the seeking for genetic segregation must be continuous, being used as alternative scion cultivars. The work aimed to evaluate fruit set, seeds number, fruit flesh and fruit diameter regarding crosses between pear cultivars. The trial was carried out from September 2008 to February 2009 in a commercial orchard in the city of Vacaria/RS, Brazil. For pollination process, it was used sample of pollens extracted from flowers at pre-flowering stage in the same orchard. The cultivars used were Packham's Triumph and Clapps Favorite. The crossings were defined as: T1 - open pollination; T2 - autopolination of Packham's Triumph and T3 - Packham's Triumph × Clapps Favorite. The pollinations were manually done in flowers at pre-flowering stage, which were opened, emasculated and then pollinated. Each pollinated flower was isolated with a fine nylon bag. Open pollination and cross pollination between Packham's Triumph ↔ Clapps Favorite provided higher fruit set (18 and 16%, respectively). Fruit flesh and diameter did not differ among treatments. Open pollination provided fruits with higher number of seeds, probably due to the presence of bees in the orchard. The lower number of seed observed in the others crossings could be due to parthenocarpy.

S12.345

Berry Trials in the Azores

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The Direcção de Serviços de Agricultura e Pecuária, Ponta Delgada, São Miguel, Açores, Portugal, began a collaboration with the United States Department of Agriculture, Agricultural Research Service, National Clonal Germplasm Repository at Corvallis, Oregon, and The Ohio State University, Department of Entomology in 2004. While previous horticultural production in the Azores included ba-

nanas, pineapples, apples, pears, and grapes, the cultivated temperate-zone berries have not been grown extensively. Our objectives were to establish and evaluate the growth of berry cultivars in the Azores. From 2006-2009, plantings of Southern highbush blueberries, *Vaccinium corymbosum*, 'Emerald', 'Spring High' and 'Jewel', 'Brigitta', 'Duke', 'Elliott', 'Misty' and 'O'Neal' were established in Furnas, and in Ponta Delgada. In 2007 and 2009, plantings of thornless blackberry *Rubus hybrid* 'Triple Crown,' and two locations of red raspberries, *Rubus idaeus* 'Heritage' and 'Taylor,' were established. In Furnas, only the 'Emerald' blueberry produced fruit in 2008 and 2009. The average yield in the first year was 0.4 kg/plant; in 2009 it was 2.6 kg/plant. The average fruit diameter was 13.95 mm, and weight was 2.44 g. The other cultivars did not produce fruit. 'Jewel' was susceptible to leaf rust disease caused by *Pucciniastrum vaccinii*. The blackberries produced fruit in 2009. Bud break occurred very late in the season (beginning of June) and the crop season extended from 26 August to 17 December although during the last month only a few fruit were produced. The production peak occurred on the first week of October. The 'Heritage' red raspberry fruit ripened on 7 August and finished 14 September. The maximum production occurred on 17 August. The average fruit diameter was 15.93 mm, length was 17.50 mm, and weight was 2.60 g/berry. The raspberries became infested with *Spodoptera littoralis* (Boisduval). 'Taylor' was more susceptible than 'Heritage' to this lepidopterous pest. Trials are continuing.

S12.346

Investigations on Bioagronomical Behaviour of Local Sweet Cherry Genotypes of Latium Region *ex situ* Preserved

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Sweet cherry is grown in Latium Region on small hilly areas, normally trained with traditional techniques and utilising local germplasm. The behaviour of nine autochthonous genotypes was evaluated in *ex-situ* field collection planted on 2005 with one-year old plants, grafted on Colt rootstock, at 5 x 3 m spaced and raised at vase training system. Cropping was observed for the first time on 2009, and during this year the following parameters were registered: vigour, time of flowering and harvesting, fruit set, plant yield, morphological and qualitative traits of fruits (weight, shape, sugar content, acidity, phenol content) and compared with two well known cultivar: Mora di Vignola and Napoleon. Concerning flowering time, one group of cultivar, represented by Cellanese, Durone nero and Morona, started blooming at the beginning of April and another group, represented by Core, Ravenna GC and Napoleon, at last decade of April. Regarding harvesting time, Magliolina and Cellanese were ready to be harvested on the second decade of May, while Durone nero, Mora di Vignola and Napoleon on the second decade of June. During this first year of cropping, high differences in plant yield and productive efficiency, among these genotypes were observed. Fruit weight ranged from 3 g of cv. Magliolina to 7 g of cv. Bianca, and high variability was observed for the qualitative aspects of fruit, such as sugar content, acidity and phenols content. The values of basal cross-sectional area indicated the high vigour of the genotypes "Morona" and "Bianca", whereas Core and Ravenna GC the lowest plant vigour. Considering yield and quality aspects of fruits and plant vigour, it seems that some genotypes could be valid for new plantations in some areas of Viterbo province.

S12.347

Pear (*Pyrus* spp.) Genetic Resources from Northern Japan: Evaluation of Threatened Landraces for Morphological and Agronomical Traits

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The number of *Pyrus* spp. endemic in Northern Japan including 'Iwateyamanashi' (*Pyrus ussuriensis* var. *aromatica*) has been decreasing, thus conservation and evaluation are urgently needed. Seven hundred and fifty pear accessions were conserved *ex situ* as wild pear germplasm collection at Kobe University. Thirty landraces in collection were especially maintained by top grafting to evaluate morphological and agronomical traits of their fruits. Flowering date, harvest date, fruit shape, skin color, diameter, height, peduncle length, fresh weight, loculus number, calyx persistency, number of sepals, flavor, firmness, sugar content (Brix), acidity (pH), and astringency were investigated for two years. Sugar content ranged from 9 to 16 %. pH of fruit extract ranged from 2.98 to 4.84. Fruit with high acid content is suitable for cooking or processing. Days from full bloom to harvest was 80 to 200. By sensory evaluation method, eleven accessions were revealed to have sweet and strong flavor not detected in modern Japanese pear cultivars. Early maturing varieties had a significant tendency to have a strong flavor and one of landraces, 'Sanenashi' was seedless. Until the 1940S, 'Sanenashi' fruit had been used as canned preserve. These threatened landraces were revealed to possess such unique agronomical traits those were not found in the modern Japanese pear cultivars. Pear genetic resources collected from Northern Japan will provide new usages not only for breeding but also processing.

S12.348

Effect of Seed Storage Conditions and Gibberellic Acid on *Annona squamosa* L. Seed Germination and Vigour

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In order to evaluate the effect of storage temperature and gibberellic acid (GA3) on germination and vigour of *Annona squamosa* L. seeds, several samples with 21 to 23 % of moisture were placed under room temperature (24 ± 5 °C) and low temperature (4 ± 1 °C) over 2.5 months. After that, the seeds were soaked in GA3 doses of 400, 450, 500, 550 y 600 ppm. A "roll towel" germination test was developed under 38 °C and 80 % of relative humidity. Low temperature affected negatively the germination percent. Gibberellic acid soaking affected positively germination percent and speed of germination. Period of storage had no effect on vigour expression. The best treatments to promote germination were room temperature storage with 400 ppm (83.68 %) and 500 ppm (83.46 %) of GA3 where more than 50 % of germination in each treatment was obtained 10 days after sowing. Low temperature storage reduced germination percent but had no negatively affect neither on speed of germination nor on seedling vigour. The best seedling vigour expression was obtained at low temperature storage with 400 or 450 ppm GA3. The germination began at 5 days after sowing and continued until 18 days, however the maximum germination was observed between 9 and 12 days. These results indicated that the seed storage and seed soaking with GA3 evaluated, allowed the breakdown of dormancy quickly.

S12.349

Investigation of Qualitative Morphological and Geographical Diversity among Native Populations of Persian Walnut (*Juglans regia* L.) in Golestan Province of Iran

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Persian walnut is a plant belongs to *Juglandaceae* family. Among 21 species, of the

Juglans genus, the Persian walnut (*Juglans regia* L.) is recognized as the best species. One of the ways to understand genetic diversity among cultivars or populations is finding genetic similarity using morphological traits. In order to understand diversity among five populations of walnut tree, this research has been conducted. Ninety six tree samples were studied for 49 different Qualitative morphological traits (according to IPGRI and UPOV descriptors). Variance analysis of traits showed significant differences ($p < 0/01$) among all traits existed, except of wide of nut form, bottom of nut form, hard surface structure, deep of narrow on side of, surface structure, skin tissue, fullness of kernel and branch cork. That showed high diversity among native walnut traits in Golestan province. Difference of traits among locations used means comparison and Duncan test. Further results indicated a negative correlation between high altitudes with sensitive of Blight. Reversely there was a positive correlation between altitude with cold resistance and kernel brightness. Other Phenological traits such as seed biophysics and seed morphology (according to IPGRI and UPOV descriptors) were evaluated within this study. This Result showed there was high diversity in native walnut in Golestan province.

S12.350

Seed Characteristics and Seed-Fruit Correlation of Turkish Watermelon Germplasm

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This study was carried out at the Department of Horticulture, Faculty of Agriculture, University of Cukurova in Turkey. Totally, 327 genotypes were used which mostly collected from different regions of Turkey during 1993-2006. Different species of *Citrullus* were also obtained from United States Department of Agriculture (USDA), French National Institute for Agricultural Research (INRA) and Egypt. Seed weight, seed length and seed width were measured for 15 seeds per genotype. The seed weight was between 20 mg and 211 mg, the seed length was between 5.03 mm-16.63 mm and the seed width was between 3.39 mm-9.53 mm among the measured genotypes. Nine genotypes were chosen based on seed size (3 small, 3 medium, 3 big) in order to evaluate correlation between seed characteristics and fruit characteristics. Correlation analysis was done between measured seed parameters and fruit weight, length and diameter in chosen 9 accessions. Measured seed parameters were negatively correlated with fruit weight, fruit length and fruit width while seed weight positively correlated with seed length.

S12.351

Genetic Evidence of Intra-Cultivar Variability within Italian Olive Cultivars

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Cultivar characterization for fruit trees certification requires fast, efficient and reliable techniques. Microsatellite markers (SSR) were used in the molecular characterization of 70 olive trees of *Olea europaea* subsp *europaea* from three cultivars: 'Carolea', 'Coratina' and 'Frantoio'. The DNA from the olive cultivars was analyzed using ten preselected SSR primers (GAPU59, GAPU71A, GAPU71B, GAPU103A, UDO99-01, UDO99-12, UDO99-28, UDO99-39 DCA9 and DCA18) and revealed 44 alleles, which allowed each genotype to be identified. In the dendrogram, the ten primers allowed the 70 olive trees to be grouped into subgroups corresponding to the same cultivar denominations. SSR markers proved to be efficient and reliable for the molecular characterization of Italian olive cultivars.

S12.352

Response to Postharvest Storage and Fruit Characterization of Wild Plum (*Ximenia americana* L.) in Mexico

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Wild plum (*Ximenia americana* L.) is distributed in several states in the Central and South of Mexico. Although, in some African countries, there are reports showing the potential use of the fruit of this species, in Mexico only fresh consumption, as well as the use of leaves for treating chest pain have been reported. One important location of trees of this species is Tepexi de Rodríguez, Puebla, in Central Mexico. There, in the first year, we characterized the fruit of 15 trees. Fruit weight was between 6.5 and 4.2 g; with a flesh percentage around 60%. Total sugar content was between 9 and 20%, with a titratable acidity of 1 g·100 g⁻¹ F.W. Total soluble solids (TSS) was between 11 and 17 °B. In the second year the fruit of one selected tree were stored at 5 °C and room temperature. Eight days after harvest (DAH) fruit stored at room temperature lost 20% of their initial weight 8 days after harvest (DAH); whereas those fruit stored at 5 °C lost only 5% at 12 DAH. The TSS increased at the end of the postharvest storage in the fruit at room temperature but they were stable at 5 °C, with an average of 14 °B; similarly, at the end of the storage, the content of total sugars increased in the fruit at room temperature. Titratable acidity was different between the two treatments, at room temperature the fruit increased their acid content 3 DAH, but the fruit stored at 5 °C were stable. These results show a good response of wild plum fruit to cold storage, increasing four days their postharvest life.

S12.353

Management of Fruit and Grapevine Germplasm in Georgia

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Georgia, as one of the ancient country, is very rich with autochthonous varieties of fruit and grapevine, still playing important role in the horticulture of the country. And as a country of the South Caucasus Georgia is also rich with crop wild relatives of fruits and grape. That is why management of local germplasm is very significant for sustainable development of future agriculture, started since the XIX centuries and having the best period in the XX century, while ample number of field collections with native and introduced varieties has been established in the country. Consequently, the germplasm became an object of wide scientific investigation of various disciplines, created the basis of the recommended lists of cultivated varieties. In the XXI centuries - in spite of financial difficulties of the country due to transmission economic period - some new field collections have been established and ample number of native varieties as well as their wild relatives has been investigated using traditional ampelographic and pomological methods of their study and modern tools of DNA fingerprinting - especially on the grapevine - mostly within activities of international collaboration on conservation and investigation for genetic resources in Georgia.

S12.354

European *Corylus avellana* Germplasm Collections

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The main aim of this work is to gather the maximum information on hazelnut germplasm existing in different European collections. Each Research Centre keeps

its hazelnut material according to its own interest: native varieties, suitable material from abroad and promoting the exchange of plant material among research centres from all over the world. At first a complete list of 264 hazelnut accessions existing in 13 different collections was elaborated: one collection in France and Greece, two in Slovenia and Spain, three in Portugal and four in Italy. The material of each collection was accurately observed, morphologically characterized and also identified by molecular markers (SSR). Some mistakes on cultivar spelling have been noticed. Characterization of hazelnut collections allowed to detect some synonyms in the germplasm studied and correct the spelling mistakes. A final list of 209 cultivars and 40 selections, growing in 13 European hazelnut collections, was elaborated. Cultivars were originated from the following countries: Albania (1 cultivar), Balkan area (2 cvs.), Belgium (1 cv.), England (12 cvs.), USA (7 cvs.), France (7 cvs.), Germany (5 cvs.), Greece (1 cv.), Hungary (1 cv.), Italy (61 cvs.), Portugal (3 cvs.), Romania (4 cvs.), Slovenia (3 cvs.), Spain (83 cvs.) and Turkey (7 cvs.). Eleven cultivars from unknown origin were listed.

S12.355

Morphological Evaluation of Some Mango Genotypes in Iran

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The relationship among 48 quantitative and qualitative traits of leaf, flower, fruit, seed and internal components of 48 Mango genotypes evaluated. The traits include leaf length, leaf width, petiole length, leaf shape, fruit weight, fruit length, fruit width, fruit shape, seed weight, length, and width, quantity of seed fiber, fruit vitamin C, juice TSS, acidity, fruit juice pH and yield. Analysis of variance showed that these genotypes have significant difference in most of the investigated traits. Significant positive and negative correlations among some important traits were found. The effective characters are divided in 16 groups that covered 83.7% of total variances. Eigen value more than 0.6 considered as significant Eigen value. Cluster analysis by these factors and using Ward Method based on Euclidean distance divided genotypes in to 2 main this clustering pattern separated genotypes from each other. Group 1 Pakistan genotypes and group 2 Indian genotypes. Biplot analysis by using the 2 first factors distributed genotypes and separated Pakistani and Indian genotypes.

S12.356

Responses of Melon Genotypes to Drought Stress

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The objectives of this work were (1) to screen some melon genotypes (31 accessions) for drought tolerance and (2) to identify the best traits for screening the melon germplasm in tolerance to drought stress. Drought condition was performed when plantlets reach 3 to 4 leaves stages. Responses of the genotypes to drought were evaluated visually by using 0-5 scale as well as fresh and dry plant weights, leaf number, leaf area, plant height, plant diameter, RWC (Relative Water Content), membrane injury, water use efficiency (WUE), K and Ca ions contents. At the end of the study, melon genotypes classified as tolerant, mild tolerant and susceptible. WUE and membrane injury index were most reliable traits for screening melon genotypes to drought stress. Among the screened accessions CU 159 and CU 196 showed most tolerant and CU 40 and CU 252 were most sensitive genotypes to drought conditions.

S12.357

Determination of Salt Tolerance Levels of Melon Genotypes

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Salinity is a major abiotic stress reducing the yield of a wide variety of crops all over

the world. One screening study was performed in order to determine the genotypical differences of 31 melon genotypes for salt stress. Seeds were germinated in a mixture of peat: perlite of 2:1 ratio. After 21 days of sowing, seedlings were transferred to plastic pots containing mixture of peat: perlite of 2:1 ratio. Two week later, salt treatment started and NaCl concentration was increased by increments of 50 mM per day until a final concentration of 200 mM was achieved. Stress responses of the melon genotypes were evaluated in early plant development stage (seedling stage). Genotypes were classified according to the severity of leaf damage symptoms by using a 1-5 symptoms scores. Dry plant and root weights, leaf number, leaf area, plant height and plant diameter, relative water content (RWC) and membrane injury index, Na, K, Ca and Cl ions content were also investigated. In conclusion, the melon genotypes showed large variation in their sensitivity to salt tolerance. Reliable and effective screening parameters for determining salt tolerance level of the melon genotypes were discussed.

S12.358

Genetic Diversity in Some Turkish Melon Accessions

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Melon (*Cucumis melo*) is considered the most diversified species in the genus *Cucumis*. It can be divided into seven distinct types based on the previously discussed variation in the species. Asia Minor is situated in the secondary diversification center of the melon. Present study was carried out to characterize a group of melon selected from our germplasm in which more than 400 genotypes exists. A total of seventy-seven accessions, sixty from different part of Turkey and seventeen as control from abroad, were characterized according to their morphological characteristics. A total of eighteen morphological properties (related plant growth habit, leaf, flower, fruit and seed) of the studied genotypes were observed and measured. Collected data were evaluated and UPGMA similarity tree plot dendrogram was generated by correlation matrix. Similarity coefficient values were measured between -0.12 and 0.98 (average 0.43) and genotypes were distributed in to four clusters. French genotypes Vedrantaïs and Isabelle were more closely related ones (0.98) according to the morphological characteristics measured and the genotypes Hasanbey and Ipsala-Yuva were followed French genotypes by 0.95 similarity coefficient. Mantel test was applied to compare correlation matrixes and r value was obtained as 0.64.

S12.359

Ovule Dimensions and How this Relate with Fruit and Seed Size in Pear

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This study was conducted to analysis and compare pear ovule dimensions and its relation with fruit and seed size in couples of pear species and cultivars. Pear flowers at the anthesis were collected from *P. boissierana*, *P. betulifolia*, *P. pyraeaster*, *P. koroshinsky*, *P. cossoni*, *P. ussriensis maxim*, *P. longipes* and *p. species*. Cultivars were included; japanese pear "Nijisseiki", "Sensation" and "Lemon Bergomot. Petal, sepals, anthers were discarded and ovule were fixed in 3% glutaraldehyde in 0.025 M phosphate buffer (pH 7.0) or in FAA (formaline: acetic acid:50% ethanol, 5:5:90). Sections at 3 or 4 mm of thickness were stained with periodic acid-Schiff's reagent and toulidine blue and were studied using light microscopy. Length and diameter of ovary and ovules of pear were measured and analysed under a complete randomized design. Comparison of means using Duncan's test at 5%. Results showed that the highest length of pear ovule belonged to *P. cossoni* species and lowest length belonged to the species of *P. ussriensis maxim*.

The highest diameter belongs to; *P.cossoni*, *P.boissierana*, *P.longipes* and the lowest diameter belonged to *P. ussriensis maxim*. For length of ovary no significant differences were observed among pear species and cultivars. The lowest diameter of ovule belonged to *P. ussriensis. maxim*. Nashi (Nijisseiki) cultivar owned the highest diameter of ovule. There was a significant correlation between seed length and width with fruit length and diameter ($P < 0.01$). Furthermore, ovary width showed a positive correlation with fruit diameter, fruit length, seed length and ovary length ($P < 0.01$)

S12.360

Preliminary Evaluation of Apple Seedlings Obtained from Controlled Cross between 'Golden Smoothee' and 'Shafi Abadi'

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In order to produce and introduce new early and mid ripening apple cultivars, seedlings derived from controlled cross between 'Golden Smoothee' as late ripening maternity parent and 'Shafi Abadi' an Iranian cultivar, as early ripening paternity parent were evaluated. This program was started since 2004 at Department of Horticulture Science, University of Tehran and this evaluation was performed at 2009 when some seedlings came into bearing. Some important fruit qualitative and quantitative characters such as fruit weight and dimensions, firmness, color, Brix, and acid content were recorded along with morphological and phenological characters. According to preliminary results, progenies were divided into four categories of early, mid, semi late and late ripening with frequency of 23, 43, 16 and 19 percent respectively. Some progenies had desirable characters for Brix, fruit size, firmness and also other characters such as tree size and height. Simple correlation analysis showed the existence of significant, positive and negative correlations among some important characters. For example significant correlations were observed between Brix and canopy width ($r=0.471$), firmness and time of leaf abscission ($r=0.299$), between length of fruit growth period and time of leaf abscission ($r=0.339$) and internode length with time of fruit ripening ($r=-0.480$).

S12.361

Variation and Correlation of Some Morphological and Phenological Characteristics in Four Apple Seedling Populations

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The variation and correlation of some phenological (seed chilling requirement, leaf abscission time and time of leaf bud break) and morphological (leaf, main axis and branches) characteristics in four populations (more than 100 plant from each population) of crosses between two Iranian apples of 'Golab-e Kohanz' and 'Shafi Abadi' (as pollen parents) and two foreign cultivars of 'Golden Smoothee' and 'Red Spur' (as seed parents) were investigated. Seeds chilling requirements were determined during stratification process, and derived plants, after four months of growth in greenhouse were transferred to the orchard in early May. All of morphological traits and leaf abscission time were measured at the end of first growth season and bud break time at the beginning of next season. SPSS software was used for data analysis. Results showed that, most of the measured characteristics had wide range of variations among populations and trends of mean were logical and justifiable based on characteristics of parent cultivars of the populations. There were high correlations between most of the leaf characteristics and also some intermediate to high

correlations between some of the leaf characteristics and some of the main axis, branches and phenological characteristics. There were intermediate to high correlations between most of the main axis and branches characteristics and between these characteristics and some of the leaf and phenological characteristics as well. All of the phenological characteristics also had intermediate correlations with each other. Result of factor analysis showed that six main and independent factors describe most of the variations, and characteristics in each of the factors were the highly correlated ones. Cluster analysis of the four populations based on six main factors showed that, two populations of each seed parents were located in separate groups.

S12.362

Maturity Stage at Harvest Influence Red Blush Development, and Quality of Apple Fruit

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Poor fruit colour development in 'Cripp's Pink' apple causes serious economic losses to apple growers. We investigated the effects of harvest maturity on the development of red blush and quality of 'Cripp's Pink' apple fruit. The fruit were harvested randomly around the tree canopy commencing from commercial harvest (CH) on April 26, and on May 10, May 24, and June 7 during two consecutive years from Perth Hills, Western Australia. The fruit were obtained from apple trees grafted on M. M. 109 rootstock, and trained on a central leader system. The experiment was laid out by following randomised block design, included four harvest dates as treatments and three replications, and single tree was treated as an experimental unit. Delaying harvesting (2 - 6 weeks) during both the seasons significantly increased red blush on the fruit surface and augmented chroma values, and lowered hue angle and lightness of ground colour of the fruit, regardless of the fruit side. Concentrations of internal ethylene were as low as $0.2 \mu\text{L}\cdot\text{L}^{-1}$, at CH and increased significantly in next two weeks during both seasons. Concentrations of total chlorophyll (chl), chl a and chl b started to decline following 2-4 weeks CH. Delaying fruit harvesting by 2 - 6 weeks following CH affected the concentrations of total anthocyanins, cyanidin 3-galactoside, quercetin glycosides, chlorogenic acid, proanthocyanidins, and phloridzin in apple fruit skin at harvest. Fruit firmness was significantly reduced and soluble solids concentration/acid ratio increased with delayed harvest (2-6 weeks following CH). Delaying fruit harvesting by 2 weeks following CH does not adversely affect fruit quality at harvest and even after 90 days of cold storage, whereas delayed harvesting (> 2 weeks following CH) adversely affects cold storage life and fruit quality.

S12.363

Cryopreservation of Genetic Resources of Tropical Underutilized Fruits in India

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Tropical part of the World has been designated as the vast store of species which one can look for new potential crops. More than 500 species of fruits are estimated to be found in Southeast Asia while the Hindustani region of diversity represents 344 species of fruits. This area has been centre of origin of a number of tropical fruits, most of which are still growing as wild or semi-cultivated. Vast genetic diversity in many of these fruits is available, however only fruits like mango, banana, citrus and guava have gained in the productivity and acceptability. Many of the tropical indigenous fruits are still remained underexploited due to the lack of awareness of their potential, market demand and low and erratic bearing in many cases. Genetic resources of such fruits are facing threat of extinction due to climate change, large-scale urbanization and developmental projects. To safeguard the existing diversity of underutilized fruits for future generations and to achieve sustainable development based on use of available genetic wealth, promotion and conservation of these species is of immense importance. Collection, characterization and conservation of genetic resources of tropical fruits species such as *Aegle marmelos* (Bel), *Artocarpus heterophyllus*, *Buchnanian lanzan* (Chironjee), *Capparis decidua* (Ket), *Carissa carandus* (Karonda), *Citrus* species, *Cordia myxa* (La-

soora), *Embilica officinalis* (Aonla), *Garcinia* species, *Grewia asiatica* (Phalsa), *Manilkara hexandra* (Khirni), *Phoenix sylvestris* (Date sugar palm), *Salvadora oleoides* (Pilu), *Syzygium cumini* (Jamun) and *Ziziphus* species (Ber) has been undertaken. Several of these underutilized fruit species are propagated through seeds and no commercial cultivars are available. Presently several *ex situ* conservation approaches have been suggested depending on propagation method and storage behaviour of species. Successful protocols were developed for cryopreservation of seeds, embryos, and embryonic axes with variable percentage survival and more than 1800 accessions have been successfully cryostored at National Cryogenebank.