Trioza erytreae Del Guercio and *Diaphorina citri* Kuwayama (*Homoptera*: *Psylloidea*), the two vectors of Citrus Greening Disease: Biological aspects and possible control strategies.

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TRIOZA ERYTREAE DEL GUERCIO ET *DIAPHORINA CITRI* KUWAYAMA (HOMOPTERES, PSYLLIDES), LES DEUX VECTEURS DE LA MALADIE DU GREENING DES AGRUMES. Aspects bioécologiques et stratégies de lutte.
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RESUME - Dans cet article sont décrits les principaux caractères écologiques et éthologiques des deux psylles vecteurs du greening des agrumes : *Trioza erytreae* (DEL GUERCIO) et *Diaphorina citri* KUWAYAMA. Une mise à jour récente de la répartition géographique de ces vecteurs est donnée et une liste détaillée des Rutacées hôtes de l'un et l'autre vecteur est fournie. Sont également passés en revue l'ensemble des ennemis naturels et leur éventuel impact sur l'équilibre des populations. Les stratégies à développer pour promouvoir une lutte aménagée contre ces vecteurs sont évoquées.

INTRODUCTION

The psyllids, or jumping plant lice, belong to the superfamily of *Psylloidea*, and are an important component of the hemipterous fauna. The *Psylloidea* comprise more than 1500 species, feeding on a wide range of host plants (57, 58, 59, 60). They fall in the sternorynchous *Homoptera*, and are adapted for sucking plant vessels, by the mean of ventral elongated mouthparts. The metathoracic legs are highly modified for jumping and the forewings remain folded in roof-like fashion.

Most species of *Psylloidea* are associated with only a few corresponding host plants, and are relatively specific in their food selection.

They breed very actively so long as new buds or foliage are available, and their infestations may occur at sudden, extremely high densities.

Both nymphal and adult stages are phytophagous. The feeding activity of the *Psylloidea* is detrimental to host plants not only due to sap depletion, but also because the nesting of nymphs results in the galling or curling of leaves, as well as honeydew secretion and subsequent development of sooty mould. More important for agricultural production, is the role of some jumping plant lice as vectors of virus-like diseases. Over the last 15 years, seven species of *Psylloidea* have been found to transmit prokariotic organisms thought to be causal agents of plant diseases (Table 1). Among these species, *Trioza erytreae* (Del Guercio) the African Citrus psylla, and *Diaphorina citri* Kuwayama, the Asian Citrus psylla are worth noting as vectors of an endocellular, phloem-restricted bacterium, associated with the *Citrus* greening disease (CGD). CGD is a destructive malady seriously affecting most commercially important Citrus cultivars in Africa and Asia.

Besides *T. erytreae* and *D. citri*, considered to be the true Citrus psyllids and which are presently the only known vectors of CGD, there are six other occasionnal Citrus feeders viz *Mesohomatoma lutheri* Enderlein. *Trioza litseae* Bordage (7), *Diaphorina communis* Mathur (74) and *Diaphorina auberti* Hollis (5, 61). The role of these psyllids as potential vectors of CGD is still unknown. But interestingly the three latter species harbour some chalcidoid

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TABLE 1 - Taxonomic position of the seven psyllas known to transmit prokariotic organisms.

Superfamily	Family	Genus	Species	Diseases transmitted
Psylloidea	Psyllidae	*Psylla*	*Psylla pyri* L. *Psylla pyrisuga* F. *Psylla pyricola*	Pear decline disease (56)
		Diaphorina	*Diaphorina citri* K.	Asian Citrus greening Disease (23) and (34)
	Triozidae	*Trioza*	*Trioza erytreae* D.G.	African Citrus greening disease (72, 76)
			Trioza nigricornis F.	Carrot proliferation disease (48 and 67)
	Carsidaridae	*Paracarsida*	*Paracarsida concolor* C.	Proliferation of Wissadula (40).

parasites insects identical to those attacking either *T. erytreae* or *D. citri*. Two other *Diaphorina* have been mentioned in Swaziland *Diaphorina zebrana* Capener and *Diaphorina punctulata* Pettey (32).

This paper comments on some bionomic aspects of the two known CGD vectors, and mentions their actual geographical distribution. It will also give the list of their respective or common host plants. A short description of the host-parasitoid or predator-prey interactions will also be given, and the possible strategies for monitoring and controlling these insects pests in Citrus orchards will be dealt with.

RELATIONS BETWEEN THE DISEASE AND THE TWO VECTORS

Citrus Greening Disease is known to exist in subsaharan Africa where it is disseminated naturally by the African psylla *Trioza erytreae* (Del Guercio). In Africa, both the insect vectors, and the greening organism, do not tolerate hot and dry climates (18, 50). The disease is thus restricted to the cool elevated areas.

Asian greening which is widely distributed in South East Asia, is a second form of the disease, more severe and heat tolerant, and transmitted by the Asian Citrus psylla *Diaphorina citri* Kuwayama. Both the Asian vector and the Asian strain of greening resist a wide range of climatic conditions, including high temperature and arid weather (18, 19).

Each type of Citrus psylla is not necessarily bound to the strain of greening of its geographical zone, nor does the presence of vectors automatically mean the spread of the disease. In fact, the transmission of the Asian strain was obtained experimentally with *T. erytreae* (73), and that of the African strain with *D. citri* (65). Alternatively, Brazil and Okinawa (Ryu Kyu) harbour the Asian Citrus psylla without the disease (10) and CGD was not reported from Gabon nor from Cape province where *T. erytreae* is established (13).

Although there is little information on the vector-pathogen relationship, it is known that adult psyllas take up the greening organism (GO) from infected plants ; the GO subsequently multiplies within the insect body. Due to the concentration of GO seen in the salivary glands, it is thought that CGD is transmitted through the secretion of salivary ducts (34, 76).

The winged adults of both Citrus psyllas breed exclusively on young shoots, the egglaying process being stimulated by the presence of a new flush. The nymphal life goes through 5 instars, and instar duration varies from a minimum of 16-18 days under warm conditions, up to 45 days under cool conditions. None of the two Citrus psyllas undergo diapause. The transmission of CGD was obtained with artificially reared 4th and 5th instar nymphs of *D. citri*, younger stages apparently being unable to transmit the disease (24, 104). No CGD transmission was reported to date with *T. erytreae* nymphs.

On dormant trees *T. erytreae* and *D. citri* adults are forced to feed on mature leaves or twigs. They may live for 80-90 days thus offering a long period of acquisition feeding. It is admitted that the first adults to colonize the spring flush are highly infective and carry the disease to new shoots (27).

The proportion of viruliferous insects in a given psylla populations is variable, and experimental transmissions using wild insects are sometimes erratic. However uniformly high levels of transmission can be obtained by forced acquisition feeding on diseased plants (12, 24, 72). Under natural conditions, the spread of the disease depend on i) the amount of CGD inoculum in the environment, ii) the psylla population density.

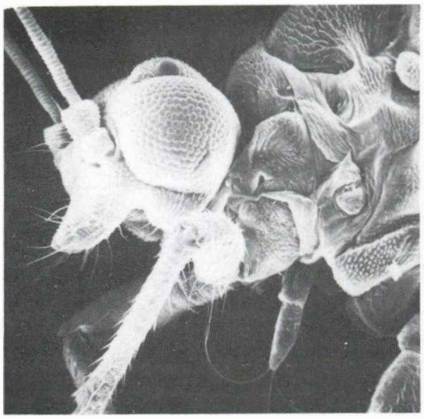
The African and Asian Citrus psyllas are able to exploit their environment in a relatively short period of time because of i) extreme fecundity of the females, ii) flying capacity iii) and ability to build up on wild alternate Rutaceous host plants, thus forming reservoirs. Both insects are therefore efficient vectors in CGD contaminated areas.

THE AFRICAN CITRUS PSYLLA *TRIOZA ERYTREAE* IN SUBSAHARAN EASTERN AND SOUTHERN AFRICA AND ON THE ISLANDS OF THE INDIAN OCEAN

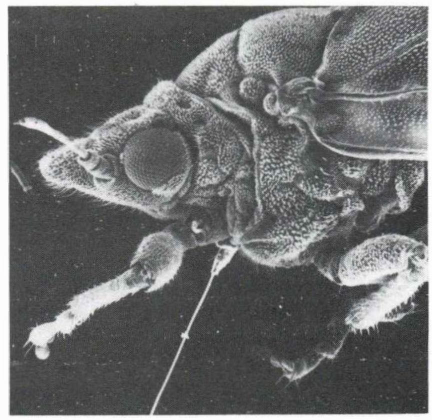
Taxonomic position : (Superfamily : *Psylloidea*, family : *Triozidae*).

Trioza erytreae (Del Guercio) belongs to an African group of 10 species attacking 4 plant families : *Rutaceae, Menispermaceae, Araliaceae* and *Salicaceae*. Three of these

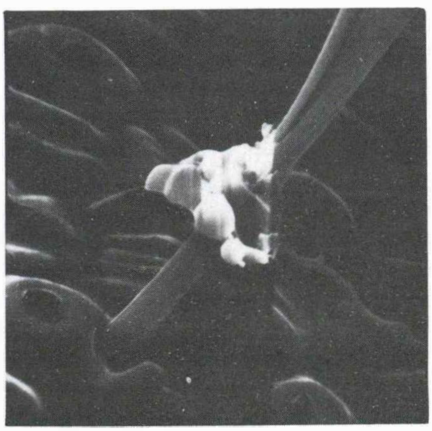
Fig. 1.- Scanning electron photomicrographs of the two Citrus-Greening-Disease vectors (Arrow indicating ventral elongated mouthparts).



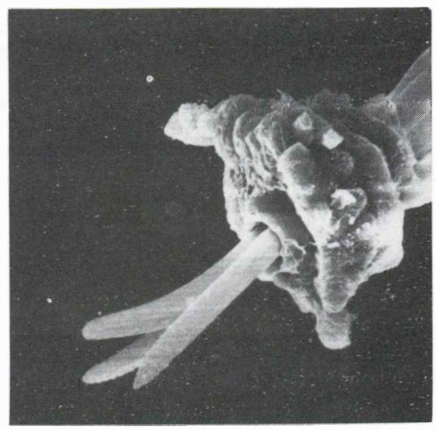
A. Lateral view of the head of *T. erytreae* (x 94).



B. Lateral view of the head of *D. citri* (x 94).



C. Retracting stylet of *D. citri* from the lower side of an orange leaf (x 1560). Note the secretion of saliva from the salivary glands (setal sheath), accumulated near the epidermis of the leave.



D. Distal part of the stylet of *D. citri* (x 3560), showing the three threads of the retracted stylet and an accumulation of saliva from the setal sheath.

10 species are extremely difficult to separate taxonomically i.e. *Trioza erytreae, Trioza catlingi*, Hollis, and *Trioza menispermicola* Hollis (60). Adults and larvae of *T. catlingi* have been found to develop on *Stephania abyssinica*, and *Cesampilos* sp. (*Menispermaceae*) in Kenya, Tanzania, and South Africa, while *T. menispermicola*, develops on *Triclisia macrophylla, T. patens* and *Cissampilos owariensis* in Ghana and Nigeria (60).

In Mascareignes Islands (Mauritius, Reunion and Rodrigues), *Trioza litseae* Bordage is very similar to *Trioza erytreae* apart from a slightly smaller size (7, 60). *T. litseae* breeds preferentially on *Litsea glutinosa* and occasionnally on *Citrus* sp. (7).

Host plants.

The true *T. erytreae* is known to complete its development only on members of the *Rutaceae* family. There are at least 18 species of plants on which the African Citrus psylla can feed. but egg laying and nymphal development are restricted to 15 and 13 species respectively (12, 77). (See table 2).

Clausena anisata and *Vepris undulata* which support more active breeding colonies if compared to other species, are thought to be the indigenous host plants of the African Citrus psylla (77).

Geographical distribution.

A first description of *T. erytreae* was obtained from South Africa in 1897 (71). The insect was later observed in Kenya and Erytreae (42) and then in Zaire and Rwanda (47). In the 1960's *T. erytreae* was also described from the following countries : Sta Helena (97), Zambia (3), Rhodesia Zimbabwe (101), Madagascar (22), Reunion and Mauritius (18, 78), Uganda (68) and Sudan (46). Specimens in the British Museum of Natural History have also been collected from Sao Tome, Swaziland, Malawi, Tanzania, Angola and Cameroon (37). More recently *T. erytreae* was recorded from the South Western part of Saudi Arabia (21) and Yemen (20). Typical leaf galls were seen in Rodrigues (6) and Gabon (13).

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TABLE 2 - Classification of host plants of *T. erytreae*, the African Citrus psylla (12, 77).

		Leaf sucking	Egg laying	Nymphal development
Preferred host plants	*Clausena anisata*	+++	+++	+++
	Vepris lanceolata	+++	+++	+++
	Citrus limon	+++	+++	+++
	Citrus medica	+++	+++	+++
	Citrus aurantifolia	++	++	++
Common host plants	*Citrus sinensis*	++	++	++
	Citrus nobilis	++	++	++
	Citrus reticulata	++	++	++
	Citrus deliciosa	++	++	++
	Citrus paradisi	++	++	++
	Citrus grandis	+	+	+
	Murraya paniculata	+	+	+
	Fagara capense	+	+	+
Occasional host plants	*Toddalia asiatica*	+	+	-
	Fortunella sp.	+	+	-
	Poncirus trifoliata	+	-	-
	Calodendron capense	+	-	-
	Microcitrus australisiaca	+	-	-

+++ Very common ++ Usual + Occasional
- Not observed in natural nor experimental conditions.

Techniques of survey.

T. erytreae can be easily detected on Citrus leaves, owing to the clustering of the nymphs in pitlike galls that snugly accomodate the developing insects. Since Citrus leaves remain on the tree for two or more seasons, a count of the leaves, pitted by psylla nymphs indicates previous *T. erytreae* activities.

Because indigenous plants of the family of Rutaceae host *T. erytreae*, it is essential to detect wild reservoirs of the insect pest in Citrus areas. Monitoring traps can be used for this purpose. Winged adults of *T. erytreae* are positively phototaxic of the responding optimally wavelength 500-550 nanometers (yellow-green) (94). The attractive component of the trap can be a 3M Saturn Yellow adhesive tape (SYAT) (90). Extremely high numbers of *T. erytreae* occur in some rainforest areas of Cameroon, on isolated Citrus orchards and nurseries, and have resulted in catches of 2-3 winged adults per sq cm of SYAT, per day (14). However, psylla populations are generally, much lower in African orchards.

Due to the possible confusion of winged adults of *T. erytreae* with that of other closely related species caught on the same traps, it is advisable to compare the results of trapping with the nymph gall density on Citrus leaves. Gall rating can be assessed by counting the colonies per sq. meter of canopy area. An average of 40 to 50 galls/sq. m. is indicative of a fairly high population. But gall ratings three times higher in magnitude were eventually counted (14).

Influence of extremes climatic conditions on survival.

Eggs and young instars (mostly 1st instar) of *T. erytreae* are extremely vulnerable to dessiccation(29). The two most acceptable predictors of survival are (i) the mean daily maximum temperature, (ii) the mean minimum vapor pressure. Combining these two parameters, one can obtain the air's evaporative power or saturation deficit (SD) (50). Field experiments showed that beyond SD's of 45 mbars, 100 % mortality of eggs and first instar nymphs succumb ; 35 mbars induced an average of 70 % mortality, while at 15 mbars only 10 % of those young organisms died (28, 48).

T. erytreae cannot therefore establish itself in hot and dry areas where midday temperatures and RH's regularly reach 32°C or more combined with 30 % RH. This includes the arid or semi-arid climates with low rainfall and high temperatures (Sahel area and hot dry coastal climates). But on the other hand subtropical climates of highlying areas (between 1000 and 2000 m) which are quite common in Eastern and Southern Africa, or cloudy rainforest areas of middle altitude e.g. Southern Cameroon and Northern Gabon, provide suitable conditions for the development of *T. erytreae*. In such areas there may be seven or eight generations per year with highest population densities during the main flushing periods. In warmer lowlying areas, high summer temperatures can reduce the populations to less than three generations per year (4).

The influence of natural enemies

Several authors have acknoledged that parasitoids and predators may be an important factor limiting populations of the African Citrus psylla (96, 25, 28).

1. **Parasitoids** : In Africa, *T. erytreae* harbours a complex biotic community comprising primary parasites and also hyperparasites i.e. secondary and tertiary parasites. It is generally thought that such a complex community contributes to greater stability within the species population and the ecosystem itself (62). Parasitoids are defined

as entomophagous insects whose development destroys the hosts. They behave as larval parasites only, and few species are highly specific.

Four species of primary chalcidoid parasites are known to attack *T. erytreae* nymphs i.e.

- three eulophid ectoparasites attacking the 3rd, 4th and 5th instars : *Tetrastichus dryi* Waterston and *Tetrastichus sicarius* Waterston both described originally from Kenya (99). *T. sicarius* has not been found neither in Zimbabwe (41) nor in South Africa (84). Some taxonomists are considering *T. sicarius* as a synonymy of *T. dryi*. In numerous South African publications, *T. dryi* has been refered to as *T. ? radiatus*. Recently a new species of *Tetrastichus* has been obtained from *T. erytreae* nymphs in Malawi (11) and Cameroon (14). A taxonomic study of this new species is being implemented.

  A revision of the African *Tetrastichus* parasites of psyllas will probably place this group in the genus *Tamarixia*.

- one encyrtid endoparasite attacking the 2nd, 3rd, 4th and 5th instars : *Psyllaephagus pulvinatus* Waterston, widespread throughout Eastern and Southern Africa.

Psylla nymphs parasitized by the ecto-or the endoparasites are reduced to their exoskeletons. After casting its pupal exuvium, and discharging a meconium, the adult parasitoid wasp will cuts its emergence hole through the thoracic surface (*Tetrastichus* sp.), or the abdominal surface (*Psyllaephagus* sp.) of the nymphal shell (See Fig. 2A, 2B and 2C). Both the exit hole and the meconium are typical of a given parasitic wasp (41).

In Southern Africa primary parasitoids attack a proportion of Citrus psylla nymphs ranging from 30 % to 60 %, but sometimes even up to 90 % (28).

Assessment of parasitism can be made by examining the mummies, (exoskeletons) as these structures remain firmly attached to the leaf (Hyperparasitism can usually be recognized by lateral exit hole (See Fig. 2D). *T. dryi* has proved to be the more effective chalcid in controlling *T. erytreae* outbreaks. However its effectiveness can be reduced by hyperparasites (28). In South Africa and Zimbabwe 12 hyperparasites have been obtained from psylla nymphs harboring *T. dryi*, or *P. pulvinatus* (41) (Tabl. III). The host-parasite relations for *T. sicarius* and other *Tetrastichus* sp. n. have not yet been described.

Aphidencyrtus cassatus Annecke is the more common secondary parasite found on *T. dryi* or *P. pulvinatus*. This hyperparasite is able to severely limit populations of the primary parasites (28, 41, 84). The relative importance of the other eleven hyperparasites is unknown or uncertain (41, 84). In 1974, *T. dryi* and *P. pulvinatus* were introduced without their attendant hyperparasites into Reunion island, where *T. erytreae* was thriving in the absence of any known parasitism (44). Suprinsingly *P. pulvinatus* did not establish itself in the island even after a release of 12.000 insects. But *T. dryi* established easily with an original rate of release of 30 to 50 adults per square kilometer of Citrus aerea (i.e. a total of 33.000 adults in this particular case), and has brought about the complete disappearance of *T. erytreae* in 5 years (7). This is unique in the history of biological control, and highlights the potential of exotic enemies (7, 15).

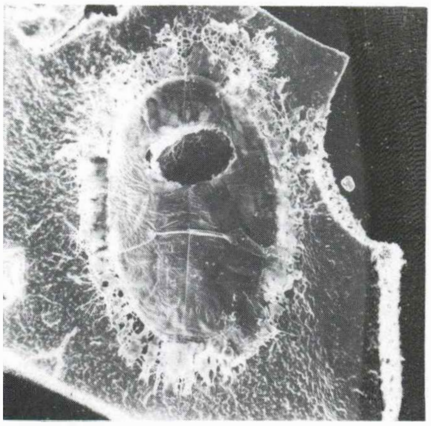
A few adults of *T. dryi* were dispatched from Reunion to Mauritius in 1975. However, only 250 adults were released on this neighbouring island, and it took twice as much time to reduce *T. erytreae* populations significantly (64). On both island *T. dryi* is thought to parasitize *Trioza litseae* (15, 64), but this eulophid is unable to attack

TABLE 3 - Parasitoid complex of *T. erytreae* in Africa (41), Indian Ocean and Middle East.

Geographical	Primary parasites	Secondary and Tertiary parasites
Southern Africa	*Tetrastichus dryi* Waterston (Eulophid) *Psyllaephagus pulvinatus* Waterston (Encyrtid)	*Aphidencyrtus cassatus* Annecke *Cheiloneurus cyanonotus* Waterston *Marietta javensis* Howard *Physcus* sp. *Coccophagus pulvinariae* Compere *Coccophagus eleaphilus* Silvestri *Coccophagus* *Euxanthellus philippiae* *Aphonogmus* Dessart *Aphonogmus incredibilis* Dessart *Pachyneuron* sp. *Tetrastichus* sp.
Eastern Africa	*Tetrastichus sicarius* Waterston (Eulophid)	Not yet described
Western and Eastern Africa	*Tetrastichus* sp. n. (Eulophid)	Not yet described
Reunion - Mauritius	*Tetrastichus dryi* Waterston (imported and established)	,,
Saudi Arabia - Yemen	Not yet described	,,

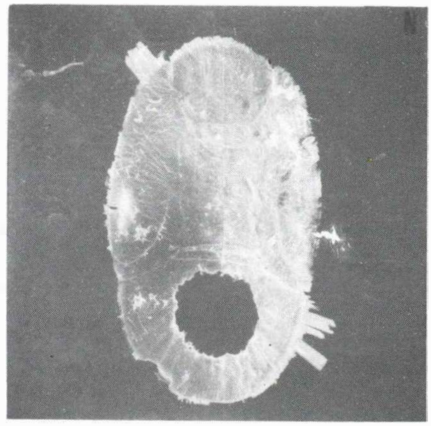
Fig. 2.- Views of parasitized *T. erytreae* or *D. citri* nymphs.

A.



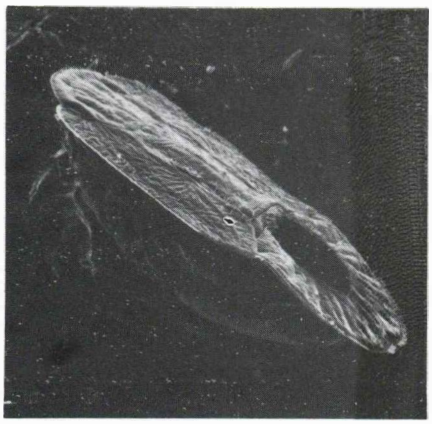
Tetrastichus dryi exit hole on the thorax of a *T. erytreae* nymph (sample from Swaziland).

B.



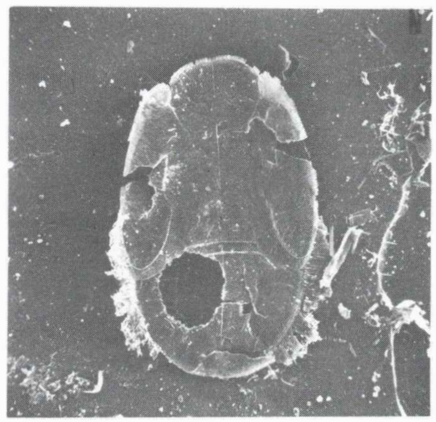
Psyllaephagus pulvinatus exit hole on the abdomen of a *T. erytreae* nymph (sample from Zimbabwe).

C.



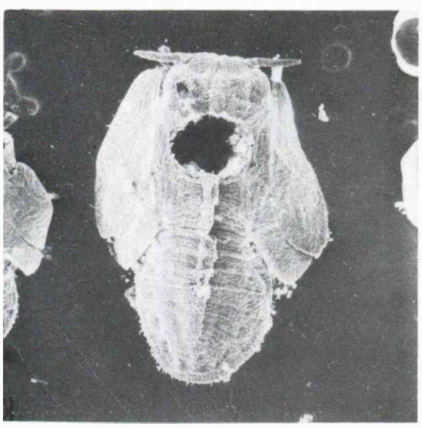
Exoskeleton of a *T. erytreae* nymph after the emergence of the endoparasite *Psyllaephagus pulvinatus* (sample from Rwanda).

D.



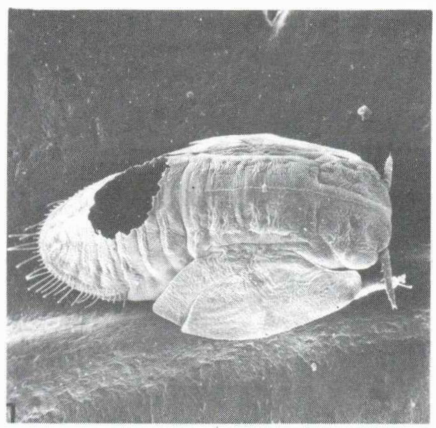
Lateral exit hole on the abdomen of a *T. erytreae* nymph due to an hyperparasite (sample from Rwanda).

E.



Tetrastichus radiatus exit hole on the thorax of *D. citri* nymph (sample from Reunion).

F.



Diaphorencyrtus aligarhensis exit hole on the abdomen of a *D. citri* nymph (sample from Java).

Diaphorina citri (44).

Predators : *Trioza erytreae* is attacked by predators including syrphids, coccinellids, lacewings, spiders and mites. Although predators exhibit less specificity than parasitoids, and generally are poorly synchronized with the upsurge of their psyllid prey, they can reduce the survival of nymphs by as much as 40 % (28), as demonstrated in one experiment (30).

Recently renewed interest has been shown in the 43 different predator species (95). Egg predation deserves particular attention in suppressing population outbreaks. In Southern Oregon, for example, egg predation gave rise to 97 % mortality among pear psylla (100). To date only two predators of *T. erytreae* eggs have been found, i.e. *Anisochrysa* sp. (Chrysopidae) and *Iphiseius degenerans* (Phytoseiidae) (95), but their impact is low, due to poor specificity.

Fungal epizootics : *Cladosporium* sp. nr. *oxysporum* was reportedly causing death and hyphal growth on *T. erytreae* nymphs (49). We found that *Capnodium citri* which induces sooty mould was able to bring about fungal epizootics in artificially reared *T. erytreae* (unpublished data). The effect of the two fungi is however density-dependent, and thus unsuitable for the control of low-level insect populations as found often under natural conditions.

DIAPHORINA CITRI IN ASIA AND SOME ISLANDS OF THE INDIAN OCEAN

Taxonomic position : (Superfamily *Psylloidea*, Family *Psyllidae*).

The genus *Diaphorina* is widespread in the Palaeartic, Ethiopian, and Oriental zoogeographical realms (58). The species *Diaphorina citri* Kuwayama is however restricted to the Oriental realm with the exception of Brazil (59).

As mentionned earlier two other *Diaphorina* species are known to feed and breed on Citrus. They are (i) *Diaphorina communis* Mathur (74) found in India, and (ii) *Diaphorina auberti* recorded from the Comores Island. (Grande Comore, Moheli, Anjouan (5, 61).

The above three psylla species exhibit distinctive morphology of the head, genitalia and wings (61, 12), and only *D. citri* is so far known to transmit CGD.

Host plants.

As with *T. erytreae*, *D. citri* is known to develop only on members of the Rutaceae family. There are at least 21 species of plants on which the Asian Citrus psylla can feed. But eggs laying and nymphal development have not been observed on several of these plants (12, 16, See Table 4).

Geographical distribution of *D. citri*.

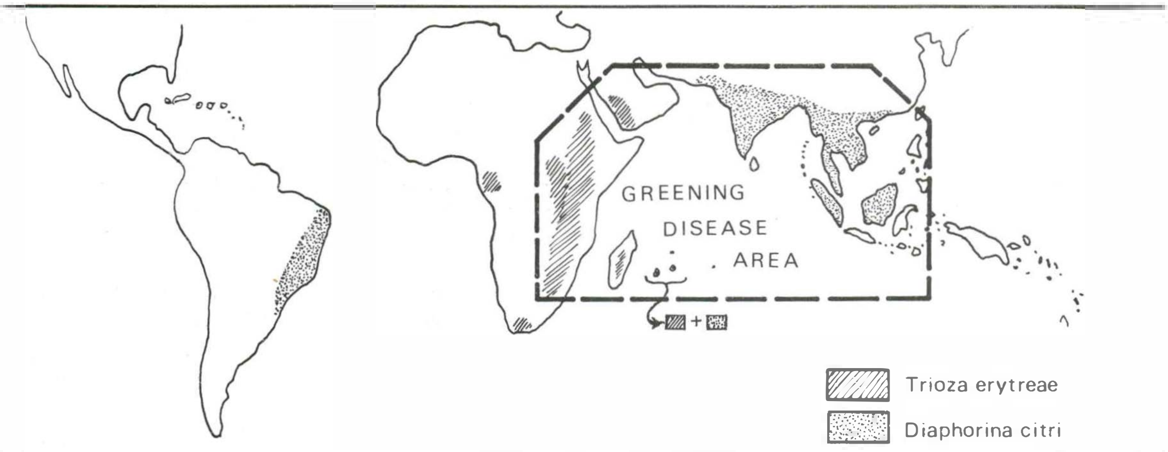
Diaphorina citri (Kuwayama) was first described in the

TABLE 4 - Classification of host plants of *D. citri* the Asian Citrus psylla (12, 16).

		Leaf sucking	Egg laying	Nymphal development
Preferred host plants	*Murraya paniculata*	+++	+++	+++
	Citrus aurantifolia	+++	+++	+++
Common host plants	*Citrus lemon*	++	++	++
	Citrus sinensis	++	++	++
	Citrus medica	++	++	++
	Citrus nobilis	++	++	++
	Citrus reticulata	++	++	++
	Citrus deliciosa	++	++	++
	Microcitrus australisiaca	++	++	++
	Citrus paradisi	++	++	++
Occasional host plants	*Citrus hystrix*	+	+	+
	Citrus grandis	+	+	+
	Triphasia trifoliata	+	+	+
	Fortunella sp.	+	+	+
	Poncirus trifoliata	+	+	-
	Murraya koenigii	+	-	-
	Toddalia asiatica	+	-	-
	Vepris lanceolata	+	-	-
	Coriea sp.	+	unknown	unknown
	Atalantia sp	+		
	Clausena lansium	+		

+++ very common ++ usual + occasionnal - not observed in natural nor experimental conditions.

Fig. 3 : Distribution map of the two CGD vectors.



Philippines (39) India (63) and then in Mainland China and Taiwan (36, 35). It was also found in Burma, Malaysia, Indonesia (43) and later in Sri Lanka Pakistan (1), Bangladesh (2), Nepal (86), Hong-Kong (93), Mauritius (78), Reunion (18), Brazil (69). Recently it was observed in Afganistan (45), Saudi Arabia (102), Viet Nam (80), Singapore (79), Rodrigue Island (6), Cambodia, Laos, Macao, Ryukyu, Singapore (37).

Techniques of survey.

Under prevailing conditions of low relative humidity and rainfall, *D. citri* can reach extremely high population levels in Citrus orchards. Nymphs then completely cover the young leaves and twigs, and visible damage in the form of leaf curling, sooty mould or honeydew secretion, and sometimes wilting of the shoots may result. *D. citri* populations are sensitive to high relative humidity or rainfall, and become much less conspicuous since the nymphs do not induce leaf galls.

Large groves may be surveyed by examining 50-200 flushes with a hand lens *in situ*, or after removal. In small backyard groves or with dormant trees, rapid searches are made for 10-20 minutes for adults or nymphal colonies (31).

Four survey techniques have also been used depending on the psylla population density.

i. Spraying insecticides and collecting winged adults falling on a white sheet of paper under the tree canopy. This method has yielded up to 41.000 *D. citri* per tree (1). This technique can be replaced by sucking the entire tree canopy with a large aspirator. On *Murraya paniculata* hedges for instance, catches of 200 adults *D. citri*, per square meter of aspirated canopy, were obtained in Reunion with a D-VAC machine (15).

Figure 4 - D-VAC machine aspirating *D. citri* on ornamental *Murraya paniculata* hedges Tampon 600 m, Réunion

ii. Collecting winged adults with a mouth aspirator and counting the number of insects caught over given period. Aspirator tubes of 1,5 mm diameter are used, allowing *D. citri* adults to be aspirated singly (7). A count of 150 adults or more caught in 10 minutes reflects a high population density, 10 adults or less in 10 minutes, indicates a low population.

iii. When dealing with young trees of less than 4-5 years, assessment in the orchards can be carried out on a random selection of 40 branches per acre. Every single twig on these branches is carefully examined and counts are made of nymphs and adults *D. citri.* The population is considered high with an average count of 5 adults and 3 nymphs per twig, and low with less than 1 adult or nymph per twig. Counts averaging 8 adults per twig were obtained in Taiwan (98) and up to 18 adults per twig in Nepal (87).

iv. The trapping of winged adults on Saturn Yellow Adhesive tape (SYAT) can also provide a good assessment of the Asian CGD vector. *D. citri* is strongly attracted by the yellow colour, and weekly trappings are able to detect the main outbreak on a spatial and temporal basis (15). Counts of 2 adults caught per cm² per day have been obtained on traps situated near heavily infested *Murraya paniculata* hedges. *D. citri* catches were particularly high at the end of the dry season (15).

The influence of climatic extremes on survival.

Three main factors influence the development of *D. citri* populations i.e. temperature, relative humidity and rainfall. But the Asian CGD vector is far more tolerant to climatic extremes than *T. erytreae.*

i. Temperature

In the South Western Arabian Peninsula, *D. citri* is commonly found from sea level up to an elevation 1500 m (21). Temperatures for such an elevation range from 32-34°C maximum temperatures in summer to 2,5°C minimum in winter. *D. citri* is absent in Citrus orchards above 1700-1800 m where occasional frosts may occur.

Very high temperatures seem also to hinder the development of *D. citri.* For instance, the insect is not found in Medina (21) where maximum temperature can reach 47-48°C during summer, while lowest winter temperatures are in the range of 6-7°C.

In the province of Guangxi, Mainland China, *D. citri* is not found above 1300 m (33). The highest elevations recorded for the Asian CGD vector, was 1100 m in Java (9) 650 m in Reunion Island (7) and 1350 m in Nepal (66). In these countries temperatures at these elevations range from 4°C to 28°C, but accompanied in these particular cases by high relative humidity and rainfall.

ii. Relative humidity.

Relative humidity close to saturation point is invariably associated with severe fungal epizootics in the 2nd, 3rd, 4th and 5th instars. Where minimum daily relative humidities exceed 87-90 % a nymphal mortality of 60-70 % can be expected.

iii. Rainfall.

Monthly rainfall in excess of 150 mm is generally associated with low populations of *D. citri* due to the eggs and young nymphs being washed off plant surfaces. *D. citri* lay eggs on the first primordia of new shoots. But instead of migrating at the lower surface of the young leaves, as do first instars of *T. erytreae, D. citri* nymphs remain fully exposed to rainfall. On the coastal area of Reunion Island for instance, the humid windward side, with 3000 mm of rainfall per year (monthly average of 166 mm), is significantly less prone to *D. citri* infestations than the arid leeward side, with only 500 mm of rainfall per year (falling during the 5 months rainy season (12). Similar observations have been made in the Philippines (26).

In Summary, *D. citri* is more sensitive to high rainfall and relative humidity than extremes of temperatures.

The influence of natural enemies.

Parasitoids and predators adversely affect *D. citri* populations. However, less is known of the general host-parasitoid or predator-prey interactions for *D. citri* than for *T. erytreae.*

Parasitoids.

Only two species of parasitoid wasps have been recorded so far, one ecto and one endoparasite.

• Ectoparasite.

- The eulophid ectoparasite *Tetrastichus radiatus* (Waterston) was described originally from Punjab India (99). This hymenopterous insect has recently been recorded in the Arabian Peninsula (8) but is absent from many Asian countries where *D. citri* is established viz. Philippines (26), Thailand (31), Indonesia (9), Taiwan (92), and probably Mainland China, and Indochina. *T. radiatus* was absent from the Mascareignes archipelago until its recent introduction into Reunion and subsequently Mauritius. In certain parts of northern India *T. radiatus* was known to attack 60 % and even 80 % of the *D. citri* nymphs (63).

T. radiatus was introduced in 1978 from Punjab to Reunion Island without its natural enemies. Here it established easily with an original release rate of 12 adults per square km of Citrus area (44). Six years later *T. radiatus* was sent from Reunion to Mauritius and Taiwan (64, 92). In Reunion and Mauritius *T. radiatus* failed to develop on the African psylla *T.erytreae,* and it only parasitized *D. citri* (44).

• Endoparasite.

- One encyrtid endoparasite *Diaphorencyrtus aligarhensis* (Shaffee *et al.*) was recorded from India (91), and more recently from the Philippines (26), Reunion (44, 7) and Viet Nam (80), This endoparasite was refered to as *Psyllaephagus harrisoni* in Reunion (44), *Psyllaephagus diaphorinae* in Taiwan (70), and *Aphydencyrtus diaphorinae* in Viet Nam (80). Its rate of parasitism does not exceed 20 % of *D. citri* nymphs, and its efficiency is much lower than that of *T. radiatus* (44).

D. aligarhensis was recently introduced into South Africa from Reunion Island but failed to parasitize the African psylla *T. erytreae* (84).

In Uttar Pradesh *D. aligarhensis* is parasitizing *Diaphorina cardiae* Crawford (54, 84), and in Comores Island it is attacking *Diaphorina auberti* Hollis (5).

T. radiatus cuts its emergence hole through the thorax of *D. citri* and *D. aligarhensis* through the abdomen (See Fig. 2E and 2F). Assessment of parasitism can therefore be carried out by examining the exoskeleton of *D. citri* nymphs which remain firmly attached to the host plant after the emergence of the parasite.

The impact of *T. radiatus* and *D. aligarhensis* can be reduced by that of hyperparasites. A common representative of the latter is *Marietta leopardina* Motschulsky (syn. *M. javensis* or *M. exitosia*, or *Perissopterus cheriani* (55). This secondary parasite belongs to the parasitoid complex of several hemipterous insects, and is widely distributed (55).

The influence of *M. leopardina* on the ecosystem of *D. citri*, with regard to especially the two primary parasites *T. radiatus* and *D. aligarhensis* is still unknown.

Other secondary parasites apparently more effective than *M. leopardina* have been found in Taiwan from *D. aligarhensis* on *M. paniculata* (92) viz *Pachyneuron apidis*, *Signiphora* sp., and one *Psyllaephagus* sp. These hyperparasitoids are able to induce up to 18 % parasitism on *D. aligarhensis*. Five other species of minor importance have also been obtained from *D. aligarhensis* and at least one *Pachyneuron* can oviposit and feed on both species *D. aligarhensis* and *T. radiatus* (92). It was found recently that the latter species despatched from Reunion had some difficulties to establish in Taiwan (92).

Predators.
Syrphids of the genus *Allographa* have been found in Reunion and Nepal (unpublished). A voracious coccinellid *Chilocorus nigritus* (F) was described as a predator of *D. citri* many years ago (63). *C. nigritus* attacks immobile or slow moving homoptera, mostly scales (89). Notwithstanding, its lack of specificity, this predator can enhance the existing natural enemy complex against *D. citri*. Other species have been recorded preying on the psyllid in Mainland China *Cheilomenes quadriplagiata* Swartz, *Coeleophora biplagiata* Swartz, *Leis axyridis* Pallas, *Synharmonia octomaculata* (F.), *Chrysopaboninensis okamoto* and *Chrysopa septempunctata* (105).

Fungal epizootics.
Fungal epizootics affecting *D. citri* nymphs are commonly seen when relative humidity is near saturation point. They are closely related to sooty mould induced by *Capnodium citri*. The effect of the fungi is density dependent, and sometimes very severely affects artificially reared colonies.

BIOLOGICAL CONTROL AND INTEGRATED PEST MANAGEMENT AGAINST THE TWO CGD VECTORS

The CGD is a graft transmissible disease, affecting most commercial Citrus, irrespective of species, varieties or scion rootstocks combination. A classical disease free certification programme is not effective for controlling the malady if viruliferous psylla infestations are recurrent.

Owing to the dissemination of vector populations on wild rutaceous plants, i.e. in forest situation where the insects are beyond the reach of chemical treatments, biological control should be encouraged. Attempts to establish host-parasitoid or predator-prey interactions must be directed primarily at forest areas or neglected orchards to reduce the level of the psylla reservoirs. Properly integrated pest management should be developed in nurseries, foundation stocks and orchards.

Biological control.

The experience gained in Reunion on *T. erytreae* and *D. citri* has shown that, as long as active hyperparasites are excluded, successful biological control can be achieved.

This island harbours 9 indigenous rutaceous plants in forest situations and 20 species of *Aurantoidea* imported for ornemental or horticultural purposes, and subsequently naturalized. From this total of 29 species, two indigenous and 16 imported plants are the hosts of *T. erytreae* and/or *D. citri* (12).

Under these environmental conditions, the programme of biological control was achieved by

i. comprehensive pre-release studies demonstrating the lack of efficient primary parasite in the ecosystem, and also the absence of active hyperparasites.

ii. selection and mass rearing of two vigorous exotic eulophids.

iii. proper number of releases in target situations.

iv. monitoring the evolution of adult psylla populations on yellow traps.

The urgency of the greening problem in the island has necessitated a pragmatic approach to transport, mass breeding and choice of the release techniques (44).

T. dryi the most active searching entomophagous parasite of *T. erytreae*, appeared to behave as a general parasite, attacking probably other endemic psylla i.e. *Trioza litseae*. Since its introduction in 1974 it has drastically reduced psylla outbreaks, and played a leading role in exterminating *T. erytreae*. No trace of the latter insect has been detected on the island since 1982 (15).

T. radiatus in Reunion proved to be a specific parasite attacking a single host species : *D. citri*. Its introduction to this island in 1978, has maintained permanently reduced levels of the Asian vector of CGD over the last 6 years.

Interestingly the two eulophid ectoparasites *T. dryi* and *T. radiatus* have completely eclipsed the less active homologous encyrtid endoparasites.

Similar successful parasite introductions have recently been made in Mauritius for both CGD vectors (64).

Integrated pest managment.

The complete biological control of the psylla vectors is probably impossible where hyperparasites decrease the activity of primary parasites. The use of insecticide then becomes necessary.

Adults of *T. erytreae* and *D. citri* are strongly attracted to new flushes, and sometimes invade entire orchards within days (88). However, they frequently congregate on the perifery of orchards where they could be managed on trap trees. These plants must be checked carefully, near the suspected sources of infestation. Another important point is that psylla populations peak during the spring flushes, consequently chemical treatments should depend on regular inspections of the orchards and be applied at critital periods.

Soil applied insecticides have a negligable effect on natural enemies and can provide good protection of nurseries and young orchards. Dimethoate (400 ml of a.i. in 100 liters of water) applied at a rate of 25 liters per 10 sq meters of nursery soil will give six weeks protection against psyllas (75). Granular aldicarb provides four months protection when applied at a rate of 0,15 g. a.i. per 5 liters of nursery soil in containers. Similar protection is obtained on young trees of 1 to 4 years of age by applying 0,75 g a.i. per year of age (17).

Corrective sprays of dimethoate are effective against both eggs and nymphs of *T. erytreae* and *D. citri* at the concentration of 0,01 % a.i. (27, 81). This concentration does not seem to seriously affect the activity of psylla parasites (27).

Three to four weeks protection can be expected with a spray of 0,012 % methyl demeton, 0,05 % phosphamidon or 0,03 % monocrotophos, but these chemicals can interfere with the activity of parasites.

In short, where psylla populations are monitored, the timing of application and the type of insecticide can be correctly chosen resulting in higher efficiency at lower costs.

CONCLUSION

The greening organism is a phloem/hemolymph restricted bacterium. It is entirely limited within plants or insect vectors, therefore, biological control of this prokariotic organism must operate within plant hosts and/or act on the outbreaks of the vector (38).

Certain entomophagous insects with strong searching behaviour, can drastically reduce psylla populations, and thus indirectly prevent an increase of the pathogen biomass. In Reunion island for instance a marked reduction of the African and Asian CGD vectors resulting from the introduction of two chalcid hymenoptera, has brought about a sharp decrease of the greening transmission. Consequently a new generation of healthy Citrus orchards could be established.

The impact on a given psylla population resulting from the introduction of new natural enemies depends not only on the properties of these enemies but also upon the complex of insects which are interacting. Therefore the final strategy to adopt for controlling the CGD vectors must be closely adapted to local ecosystems.

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