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Preliminary monitoring of *Citrus tristeza virus* (CTV) vectors in Apulia region

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Abstract. The appearance of the first localized CTV outbreaks in Italy, along the Ionian coast, and the homogeneity within each CTV population, suggested its diffusion by aphid vectors. In the spring 2007, field surveys focused on aphid population took place in three major citrus-growing areas of the Apulia region; the estimation of the number of aphid species landing on citrus trees, their identification and categorization by species were carried out. During the studied period, the population density of each species was evaluated. This research emphasizes the necessity of epidemiological investigations in order to elucidate the spatial-temporal CTV spread patterns and to prevent possible outbreaks in the Mediterranean area.

Keywords. CTV – Aphid – Vector transmission – Apulia – Italy – Citrus.

Suivi préliminaire des vecteurs du CTV dans la region des Pouilles

Résumé. La détection des premiers foyers de CTV en Italie, le long de la côte ionienne, et l'homogénéité au sein de la population de ce virus, nous incitent à supposer que la diffusion soit assurée par des pucerons. Au printemps 2007, des enquêtes ont été réalisées en plein champ dans les trios principales régions agrumicoles des Pouilles pour évaluer la présence des pucerons. On a donc estimé le nombre d'espèces de pucerons qui visitent les agrumes et on les a successivement identifiés et répartis suivant l'espèce. Pendant la période d'étude, la densité de population de chaque espèce a été déterminée. Ce travail a mis en évidence la nécessité de réaliser des recherches épidémiologiques pour éclaircir les modèles de diffusion du CTV dans l'espace et dans le temps et de prévenir d'autres éventuels foyers dans la région méditerranéenne.

Mots-clés. CTV – Puceron – Transmission par vecteur – Pouilles – Italie – Agrumes.

I – Introduction

Worldwide distribution and variable biological behavior of aphids (Homoptera: Aphididae) make them heavily damaging crop pests and efficient vectors for over than 200 plant viruses (Harris, 1989). Hence, the spatio-temporal epidemics of plant viruses are strongly compromised by the abundance and feeding behavior of their vector species (Irwin *et al.*, 2000). Within the Mediterranean Basin, the recent establishment of *Toxoptera citricidus* (Kirkaldy) in Spain and Northern Portugal (Ilharco *et al.*, 2005) represents a serious threat to the Mediterranean citrus industry. Moreover, other aphid species landing on Citrus including *Aphis gossypii* (Glover), *A. spiraecola* (Pagenstecher) and *T. aurantii* (Boyer de Fonscolombe) are endemic and known to be less efficient vectors. In Italy, Citrus tristeza closterovirus outbreaks occurred in 2003 within three major citrus growing areas through illegal importation of infected budwood (Davino *et al.*, 2003 and 2005). But, genomic homogeneity among the viral population and its rapid territorial dissemination provided further evidence of its natural spread by aphids (Yokomi and Garnsey, 1987; Cambra *et al.*, 2000; Saponari *et al.*, 2007). In this work a monitoring of different aphid species was conducted in Apulian citrus groves in the framework of a programme supported by Osservatorio Fitosanitario Regionale of Apulia region, Italy.

II – Materials and methods

Several citrus orchards within three major citrus-growing areas in Apulia, including the provinces of Taranto, Lecce and Foggia, were monitored for aphids.

From each grove, a number of 10 trees across the diagonals were randomly monitored and an average of 4 shoots/ tree were taken at different directions and height levels, then, kept in polyethylene bags in order to be examined in the laboratory.

The total collected aphid population was observed under a compound microscope at 25X to 400X magnification, based on Blackman and Eastop (1984) key of identification. In order to make the species identification easier and more effective, a number of specimens were cleared and mounted on slides following the Heikinheimo procedure (1988).

III – Results and discussion

Once visualized under the microscope, aphid specimens were divided into larval stages, apterous and alate morphs. The overall number of collected and counted aphids from all the investigated groves amounted to around 10 000 individuals. It was composed of around 75 % larvae and 25 % adults. Based on Blackman and Eastop taxonomic key (1984), around 2000 apterous adults were identified. Discrimination between *Aphis* species was mainly based on some common morphological characteristics such as antenna with terminal process less than 3 times the length of basal VI and a helmet shaped cauda. Then, *A. gossypii* was characterised by a pale cauda with 4 to 7 setae, while *A. spiraecola* was recognized owing to its dark cauda with 6 to 12 setae.

Similarly, the differentiation between *Toxoptera* species was founded on their antennae with terminal process more than 3.5 times the length of basal VI and the presence of a cauda with more than 10 hairs. The black citrus aphid *T. aurantii* was known by its antennal striations and cauda with less than 20 hairs, which was not the case for *T. citricidus*.

The identification assays showed that *A. spiraecola* and *A. gossypii* were the most abundant aphid species visiting Apulian citrus groves, representing 45% and 40% respectively of the total population in agreement with previously reported data from Spain (Marroquinet *al.*, 2004).

Nevertheless, the percentage of some species reported as less efficient CTV vectors was noticeable, e.g. *T. aurantii*, while other species represented less than 1% of the total aphid population including *M. persicae*.

As expected, the BrCAT. citricidus has never been found in the framework of this survey.

In the surveyed citrus orchards, aphid colonies were mainly composed of larvae, followed by apterous and winged females. The latter represent the morphs mostly responsible for the establishment of new colonies. A slight decrease in the larvae and apterous adult instars was observed in May, but it was followed by a new increase in June. However, the number of alate landing on citrus trees decreased in the same period, making the migration of the winged morphs to other, often herbaceous, plants (alternative hosts) evident before coming back to citrus during the next autumn. This population dynamics reflects the typical heteroecy or polyphagy of many aphid species.

IV – Concluding remarks

In order to obtain an overall idea about the population dynamics of the different CTV vectors, all the apterous adults were identified and categorized by species and date of collection in the considered period. Thus, *A. gossypii* population was the most important up to June; whereas, the

population of *A. spiraecola* started to decrease in the same period; while, *T. aurantii* appeared later in May but continued to increase up to June, since it is known to be more thermophilic.

As to the distribution of the different aphid species on the surveyed areas, the highest population density of *A. gossypii* was observed in the Nothern Apulian region followed by *T. aurantii*and *A. spiraecola*. In Massafra area, the most important CTV foci in Apulia region, a lower aphid population was scored, with a higher infestation of *A. spiraecola* compared to *A. gossypii* and the total absence of *T. aurantii*. In the Southern part, *A. spiraecola* prevailed over the other two species.

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