FIRST DESCRIPTION OF THE OCCURRENCE OF THE LEAFHOPPER *PHLOGOTETTIX CYCLOPS* IN A BORDEAUX VINEYARD

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Abstract

Aim: The aim of this study was to investigate the occurrence of new leafhopper species, which could be potential vectors of plant pathogens, in the vineyards of the Bordeaux region.

Methods and results: Grapevine woody canes were collected in a vineyard without insecticide treatments during the winter of 2008. The unidentified leafhoppers that hatched from the dormant canes were collected and individually reared to adults on grapevine cuttings in a cage. Species identification was performed with males. The survey led to the detection of a new leafhopper species in Bordeaux vineyards, *Phlogotettix cyclops*.

Conclusions: *P. cyclops* is an invasive species whose larvae look like the ones of *Scaphoideus titanus*. The presence of this species in a Bordeaux vineyard suggests that it is spreading northward across the southwest of France. Furthermore, this leafhopper could become a vine pest in the future.

Significance and impact of the study: This study provided useful information for viticultural researchers and professionals, particularly in France, but also elsewhere in Europe.

Keywords: Leafhopper, grapevine, *Phlogotettix cyclops, Scaphoideus titanus*, vection

Résumé

Objectifs: Cette étude vise à rechercher l'apparition de nouvelles espèces de cicadelles potentiellement vectrice dans le vignoble bordelais.

Méthodes et résultats: Des bois de vigne ont été collectés dans un vignoble sans traitement insecticide durant l'hiver 2008. Les cicadelles inconnues qui ont éclos des bois ont été collectées puis élevées sur des boutures de vigne jusqu'à ce qu'elles deviennent adultes. L'identification de l'espèce a été réalisée sur les mâles. Cette étude a conduit à la détection d'une nouvelle espèce de cicadelle pour le vignoble bordelais, *Phlogotettix cyclops*.

Conclusions: *P. cyclops* est une espèce invasive dont les larves ressemblent à celles de *Scaphoideus titanus*. Cette espèce s'étend au sud-ouest de la France et pourrait devenir un ravageur de la vigne dans le futur.

Impact scientifique de cette étude: Cette étude apporte de précieuses informations pour les professionnels de la viticulture en France, et plus généralement en Europe.

Mots clés: Cicadelle, vigne, *Phlogottetix cyclops, Scaphoideus titanus*, vection

manuscript received the 23rd March - revised manuscript received the 6th September

INTRODUCTION

Growing international trade increases the invasion of alien species, causing important crop damages and environmental and/or economic problems (PIMENTEL et al., 2005; MEYERSON and MOONEY, 2007). Insects, as pests or as vectors, are a major concern. For example, in the last 10 years, 50 new species of Hemiptera were introduced in Italy (JUCKER et al., 2008). Climate change could promote the success of invasions and establishments of alien species but could also create new host plantpathogen-vector interactions and thus promote new diseases (TOMAS et al., 2009). For these reasons, the regular monitoring of agrosystems for possible new pests is essential for early detection of new species that could become important pests. The earlier we can detect a new potential pest, the faster we can attempt to prevent its permanent establishment. Even if an alien species seems to be harmless, the introduction of a new species in an ecosystem could induce new interactions and lead to unpredictable new problems.

A vineyard, like any other type of agrosystem, is open to colonization by alien species with sometimes very harmful consequences (e.g. the introduction during the last century of the leafhopper Scaphoideus titanus, vector of the Flavescence dorée disease (FD)). Still, this leafhopper has become an important pest of European vineyards as a vector of the Flavescence dorée phytoplasma: by allowing a phytoplasma present outside the vineyard to shift from the wild compartment to a new host plant, the grapevine, and by promoting the rapid spread of FD within the vineyards, S. titanus is responsible for one of the most current harmful diseases of the vineyard (ARNAUD et al., 2007; MALEMBIC-MAHER et al., 2007). Thus, the monitoring of new potential vectors, including leafhoppers, can prevent the introduction of new vector-borne pathogens (bacteria, viruses, and phytoplasmas) in vineyards by the early detection of invading vector species, such as the detection of potential vectors of the bacterium Xylella fastidiosa (Pierce's disease agent) in Californian vineyards (REDACK et al., 2004).

MATERIALS AND METHODS

In order to sample egg populations of leafhoppers, twenty four kg of two-year-old grapevine woody canes (20-25 cm long) were collected in a vineyard without insecticide treatments in Barsac (Sauternes, south of Bordeaux) where important populations of *S. titanus* have been observed for successive years. The woody canes were randomly collected over an area of about 10,000 square meters by pruning the grapevine stocks at the end of October 2008 and stored in a climatic chamber at 5 °C (simulating winter conditions) for three months before hatching.

The woody canes were then pooled together and randomly split into 12 hatching cages with 2 kg of canes in each cage (see a description in CHUCHE and THIÉRY, 2009). To prevent the eggs from desiccating, a 1 cm layer of vermiculite (Efisol, France) was placed in the bottom of the plastic hatching cages (50 x 38 x 36 cm) and was humidified by the addition of distilled water every week. The hatching cages were placed in a climatic chamber under a 16:8 (L: D) photoperiod, a temperature of $23 \pm$ 1 °C and a relative humidity of 65 - 70 %. In order to harvest neonate larvae, six detached grape leaves of Cabernet-Sauvignon cultivar, maintained in a glass tube with water, were added to each hatching cages 20 days after woody canes bearing the eggs were removed from 5 °C (CHUCHE and THIÉRY, 2009). Grape leaves were replaced when they began to wither (ca. every 15 days).

Besides *S. titanus* larvae, unidentified leafhopper larvae that hatched from the dormant canes were collected and individually reared on Cabernet-Sauvignon grapevine cuttings in a cage until adult emergence. By collecting dormant canes that contained eggs we insured that the leafhoppers that we observed had oviposited on the grape canes, and by rearing emerging larvae on grape leaves we insured that they could complete their development on *Vitis vinifera*. Males were used for identification because the main criterion for identifying species of leafhoppers (family Cicadellidae) is the morphology of the male aedeagus (reproductive intromittent organ). The identification followed the taxonomy handbook for leafhoppers (RIBAUT, 1952).

RESULTS

1) Leafhopper species was determined without ambiguity after the aedeagus dissection in potassium hydroxide 10 %. The analysis of the morphological characteristics of whole body and aedeagus confirmed that the species observed was *Phlogotettix cyclops*. Indeed, the shape of some aedeagus elements is outstanding in the *Phlogotettix* genus (RIBAUT, 1952).

2) Ten *P. cyclops* larvae were recorded as hatching from the 24 kg of collected canes and only two individuals completed their development to adults on grape leaves.

DISCUSSION AND BIOLOGICAL NOTES

Phlogotettix cyclops is a polyphagous species (table 1) native to an area including Asia and Russia but now present in Middle East, Central and Southern East Europe and France (ANUFRIEV and EMELJANOV, 1988; Hoch, 2009). In France, *P. cyclops* was observed until 1987 only in Southeast France and Corsica (DELLA GIUSTINA, 1989). In 2001, this leafhopper was found

	Country	Collection site	Abundance Reference
Austria	vineyard	_	Riedle-Bauer, 2006
Bulgaria		rare	Gueorguiev et al., 1998
China	Ulmus sp.		Zhen et al., 2006
France	Apricot orchard		Jaraush et al., 200
Hungary	Apple orchards	rare	Bleicher, 2007
	Pear orchad		Bleicher, 2007
	Raspberries	high number	Dér, 2005
	Apricot orchard	high number	Dér, 2005
Italy	vineyard	frequently captured	Braccini and Pavan, 2000
Japan	Persicaria perfoliata		Miura et al., 2008
Russia	Salixaceae dominant forest	—	Gnezlidovi, 2000
Turky C	Castanea sp., Rhododendron sp.	occasionally	Lodos and Kalkandelen, 1986

Table 1 - Occurrence, abundance and host plants of *P. cyclops* in some countries.

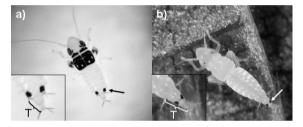


Figure 1 - First larval instar of *P. cyclops* (a) and S. *titanus* (b).

Arrows indicate characteristic black spot on the distal extremity of the larvae. The spots are on the antepenultimate segment of *P. cyclops* and on the telson of *S. titanus*. T: telson.

further west, in the French Catalonia (Eastern Pyrenees) (JARAUSCH *et al.*, 2001) (table 1). Our discovery of this leafhopper in a Bordeaux vineyard close to Sauternes represents, to our knowledge, its most recently reported Northern location in France. This suggests that after extending westward, *P. cyclops* continued to spread northward at least as far north as Gironde. Because very scarce data are available on the biology of this leafhopper, and because there are no record of its past distribution over time, we could not link this phenomenon to global warming, as has been observed for many popularly collected insects such as butterflies (PARMESAN *et al.*, 1999; THIÉRY and CHUCHE, 2007).

Like *S. titanus, P. cyclops* seems to be univoltine. Indeed, in the Eastern Pyrenees, a survey from May to November of potential phytoplasma vectors occurring in an apricot orchard yielded adults (about 40 individuals) only from early August to late September (DANET, unpublished personal observations), which is typical of insects with a single annual generation.

The P. cyclops species has never been described as a pest or vector, and during the epidemiological study on European stone fruit yellows in France, no phytoplasma could be detected in any of the tested P. cyclops individuals (JARAUSCH et al., 2001). However, this leafhopper could represent a potential vector of phytoplasma because it belongs to the family Cicadellidae which includes most of the phytoplasma vectors (WEINTRAUB and BEANLAND, 2006). Even if P. cyclops does not yet represent a problem in viticulture, its occurrence in vineyards could be confounded with S. titanus and thus complicate the monitoring of this vector species. Indeed, for vine-growers the morphological traits that distinguishes S. titanus among other grape leafhoppers, like Empoasca vitis or Zygina ramni, is the presence of two black spots on the posterior extremity of the larvae (figure 1a). But P. cyclops also has two black spots (figure 1b), making confusion possible between these two species when monitoring S. titanus. However, even if the two species look similar, the black spots are located on a different segment: on the last abdominal segment for S. titanus and on the penultimate one for P. cyclops (figure 1). Furthermore, the thorax and the mid legs of the larvae of P. cyclops are melanised (figure 1).

Because the presence of Flavescence dorée vectors in a vineyard is currently the main element that triggers the decision to apply insecticides, the accurate identification of the vector is essential, especially when only a few *S. titanus* individuals are detected and when there is a risk of misidentification. If an *S. titanus* -free vineyard is falsely diagnosed as *S. titanus* -positive (*P. cyclops* misidentified as *S. titanus*), the mandatory treatments against Flavescence dorée vector would be launched with adverse economical and ecological consequences, especially for organic or minimal insecticide wine-growers. Even though confusion is

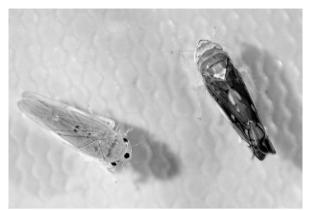


Figure 2 - Adultes of *P. cyclops* (left) and *S. titanus* (right).

possible between larvae, adults are easier to tell apart (figure 2). In our study, very few individuals of *P. cyclops* were observed compared to *S. titanus* (10 *P. cyclops* against 15,000 *S. titanus*), which would not lead to significant underestimates of *P. cyclops*. However, with high *P. cyclops* populations, like the ones observed in Italian vineyards (BRACCINI and PAVAN, 2000), misidentification could be significant.

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