# Control of phytoplasma vectors in organic viticulture

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Abstract: A comparative study is carried out in 2002 and 2003 in the Mosel viticultural area in order to evaluate the specific risk of infection by Bois noir (Vergilbungskrankheit, VK) in organic viticulture. Occurrence, abundance and infestation of the disease vector *Hyalesthes obsoletus* is monitored in organic vineyards and compared to data from conventional plots. The organic vineyards under investigation are characterized by a wide variety of herbaceous weed species. Immature *H. obsoletus* were found on the roots of known host plants like *Convolvulus arvensis*, *Urtica dioica*, and *Ranunculus bulbosus*. *Calystegia sepium* was identified as a new host plant for the vector. It is, like *C. arvensis*, a widespread and problematic weed in organic viticulture. Different isolates of the VK-phytoplasma were detected in host plants and vectors, but their biological and phytopathological significance is not yet clear. While no significant differences in the abundance of *H. obsoletus* could be observed between organic and conventional vineyards, the infestation of the populations was significantly higher in the latter. Disease incidence was generally lower in organic vineyards compared to adjacent conventional plots. Hibernating larvae of *H. obsoletus* move to a depth of approximately 18 cm during winter. Ploughing brought them up to the surface where they were killed by frost. The efficiency of this method needs to be evaluated during the flight of adult planthoppers in 2003.

Key words: Vector, phytoplasma, grapevine yellows, organic viticulture, control

### Introduction

Two types of grapevine yellows occur in German viticulture. Grapevine Palatinate Yellows (GPY) has no economic significance but Bois noir (Vergilbungskrankheit, VK) is widespread in Germany. Severe damage, however, is restricted to vineyards on the steep slopes of the valleys of Middle-Rhine, Mosel, and Nahe rivers where climate and soils provide favorable conditions for the thermophilic Cixiid planthopper *Hyalesthes obsoletus* Signoret, the only known vector of Bois noir. This species depends on common weeds in vineyards like *Convolvulus arvensis*, *Ranunculus spp.* and *Urtica dioica*, on which it completes its whole life cycle. Larvae and nymphs acquire the VK-phytoplasma by feeding on infected weeds. The pathogen is subsequently transmitted to grapevine by the adult vectors that are active for about ten weeks in June/July.

Due to the biology of *H. obsoletus*, the epidemiology of Bois noir is significantly influenced by soil cultivation and management of green cover. We are therefore interested to evaluate whether the specific conditions in organic viticulture influence infection pressure. Studies are carried out in organic vineyards as well as in adjacent conventional plots. Occurrence and abundance of the planthopper as well as the infestation of the vector populations with the VK phytoplasma and the disease incidence on grapevine were assessed.

Disease incidence could be decreased by two strategies: control of the alternative hosts plants of the phytoplasma or reduction of the population density of the vector. Control strategies have to consider the role of fallow vineyards that provide ideal conditions for both the herbaceous host plants and the vector.

The data presented here are results of the first year of a still ongoing project.

## Material and methods

The study is carried out in organic vineyards of private growers along the river Mosel. Vineyards on slopes were chosen because VK and its vector are associated with those areas. However, no information was available about the incidence of VK and the presence of H. *obsoletus* in those vineyards. In some of the vineyards a more or less permanent green cover has been established, but most plots show a spontaneous herbaceous flora of varying intensity.

The occurrence and relative abundance of *H. obsoletus* was monitored with yellow sticky traps  $(13x26 \text{ cm}^2)$  that were exposed close to the soil surface and in the height of the canopy. Adult planthoppers were captured alive from specific host plants by sweep net or a motorized suction device. To monitor the movement of *H. obsoletus* larvae in the soil, a minimum of 20 larval instars was dug from the roots of host plants in monthly intervals and the average depth was calculated.

The infestation of the vector populations was analyzed by PCR-tests of individual insects using primers specific for the stolbur-group of phytoplasmas to which the VK-phytoplasma belongs (Maixner et al., 1995). For a further characterization of the pathogens detected in insects and plants, amplification products achieved with the primers fTufAy/rTufAy (Schneider et al., 1997) where digested with the restriction enzyme *Hpa*II

Disease incidence was measured in the experimental plots and in adjacent conventional vineyards by visual inspection of each individual grapevine for symptoms of VK.

A section of a fallow field with a high abundance of infected *C. arvensis* was planted with seedlings of *Hieracium pilosella* in 1999. This creeping plant is well adapted to the xerothermic conditions of steep slope vineyards, doesn't require mowing and covers the soil through runners. We tested the ability of this plant to reduce the density of *C. arvensis* by competition. Soil coverage by *C. arvensis* was estimated by repeated estimation of the coverage in a 1m by 1m frame that was randomly thrown to the ground with 25 repeats.

In order to decrease the population density of *H. obsoletus* fields were grubbed in August or ploughed in December during severe frost with the objective to damage the larval instars either mechanically or by freezing.

## **Results and discussion**

### Host plants and phytoplasma isolates

The major host plants of *H. obsoletus* such as *Convolvulus arvensis, Urtica dioica,* and *Ranunculus* spp. were commonly found in the vineyards. *C. arvensis* or *Ranunculus* covered the soil almost completely in some vineyards but they were rare in other plots, mainly where a closed green cover had been established. Another common weed, *Calystegia sepium,* was identified for the first time as a host plant of *H. obsoletus.* We detected both the vector and the phytoplasma in all of these plants. A further characterization revealed differences between phytoplasma isolates from *U. dioica, C. arvensis* and *C. sepium* (data not shown), and corresponding results could be achieved by the analysis of phytoplasma isolates from planthoppers that were caught on these plants. *C. sepium* was found to be infected by a so far unknown isolate of the VK phytoplasma. It is not yet clear whether these differences have a biological significance.

### Relative abundance of H. obsoletus

The trapping results of H. obsoletus are presented in figure 1. The relative abundance of the vector shows a wide variation both in organic and in conventional vineyards that doesn't allow to identify significant differences. In one organic vineyard, however, a ten- to

twentyfold number of planthoppers was caught compared to all other plots. This vineyard was characterized by *Ranunculus sp.* as the predominant weed that was densely colonized by *H. obsoletus*.

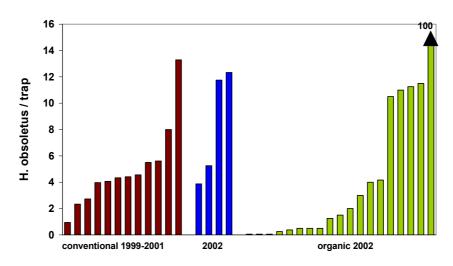


Fig. 1. Trapping of Hyalesthes obsoletus in conventional and organic vineyards

#### Infestation of vector populations

Data on the infestation of the vector populations are presented in figure 2. Compared to data of conventional vineyards from previous years, the infestation of *H. obsoletus* populations in the organic vineyards was significantly lower than in the conventional ones (U-Test, p=0,029). This might be an effect of sampling design, because the organic sites were chosen without information about their VK history, while conventional plots were chosen because of their already known high infection pressure.

The function of host plants is emphasized by the comparison of the first three conventional plots and the first two organic vineyards shown in figure 2. In these vineyards, *Ranunculus* spp. was the predominant host plant of *H. obsoletus*. Previous studies revealed that this weed dies off quickly when infected by the VK phytoplasma. Therefore, infected *Ranunculus* doesn't play a role in VK epidemiology because it doesn't allow the vector to hibernate and acquire the pathogen.

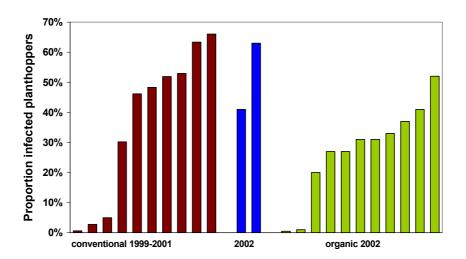


Fig. 2. Infestation of *Hyalesthes obsoletus* populations with the VK phytoplasma.

### Disease incidence

The incidence of VK was assessed in both organic and adjacent conventional plots. Data for vineyards of cv. Riesling are presented as figure 3. The low levels of incidence indicate that our experimental vineyards are not situated in foci of the disease. The proportion of symptomatic vines was usually lower in organic vineyards but no statistical significant difference could be observed. It is not clear yet, whether these differences point towards a lower risk of infection in organic vineyards. Other factors that influence the development and distinctness of VK, e.g. differences in plant nutrition and vitality, could play a role.

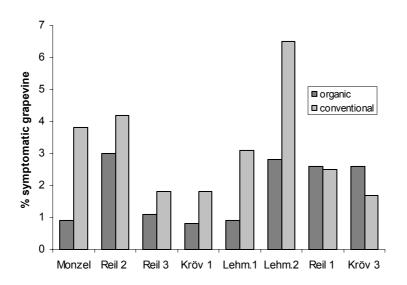


Fig. 3. Incidence of Bois noir in organic and adjacent conventional vineyards of cv. Riesling.

### **Control**

A field trial gave evidence for the ability of *Hieracium pilosella* to suppress *C. arvensis* and to decrease thereby the infestation of the vector population and infestation (figure 4). This plant might be used on fallow fields and other risky areas but, due to its susceptibility to mechanical damage, it is less suitable for vineyards. Another disadvantage is the fact that this plant needs to be planted instead of sown. However, once established, it doesn't need any more care or protection. Additional experiments have been initialized to check the ability of fast growing crops such as *Phacelia tancetifolia*, oil raddish (*Raphanus sativus oleiferus*) and grasses to suppress *C. arvensis* on fallowed vineyards.

Short after hatching the larvae of *H. obsoletus* are still close to the soil level. They move down into the soil during winter, presumably to avoid frost damage (figure 5). The maximum average depth was 18 cm in December (9 to 26 cm). While the mean depth of the larvae remained quite constant until April we recorded a steady increase of the variance which indicates an intensifying mobility of the larvae in early spring.

We tried to damage the young larvae mechanically by grubbing the soil in an abandoned vineyard in August. Furthermore, the hibernating nymphs were brought to the soil by ploughing in December during a period of severe frost. First observations confirmed that this treatment caused a high mortality. However, the efficiency of both treatments in the reduction of *H. obsoletus* populations has to be evaluated by a comparison of the relative abundance of the planthopper on treated and untreated plots during the flight of adult vectors.

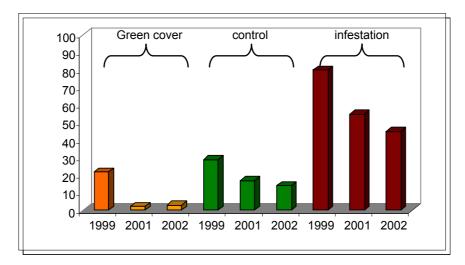


Fig. 4. Influence of green cover by *Hieracium pilosella* on the soil coverage by *Convolvulus arvensis* and the rate of infestation of the *Hyalesthes obsoletus* population in the years 1999-2001.

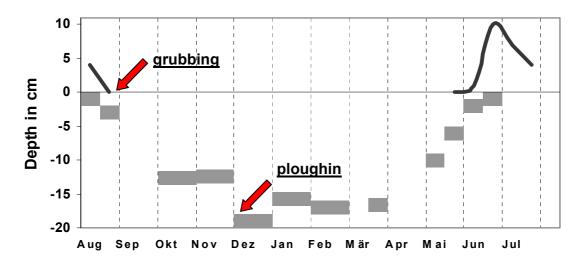


Fig. 5. Movement of *H. obsoletus* larvae in the soil. Average depth was calculated from data of at least 20 insects per date. Arrows indicate the time of soil cultivation.

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