

## Distribution and host range of the grapevine plasmodiophorid *Sorosphaera viticola*

L. HUBER<sup>1),2)</sup>, G. EISENBEIS<sup>1)</sup>, E. H. RÜHL<sup>2)</sup>, V. PAGAY<sup>3)</sup> and M. KIRCHMAIR<sup>4)</sup>

<sup>1)</sup> Johannes Gutenberg-University, Institute of Zoology, Soil Ecology, Mainz, Germany

<sup>2)</sup> Geisenheim Research Centre, Grapevine Breeding and Grafting, Geisenheim, Germany

<sup>3)</sup> Cornell University, Department of Horticulture, Ithaca, USA

<sup>4)</sup> Leopold-Franzens-University, Institute of Microbiology, Innsbruck, Austria

### Summary

*Sorosphaera viticola*, an obligate parasite of grapevine, was first detected in 2003 in roots of *Vitis berlandieri* x *V. riparia* rootstocks in a vineyard in the German Rheingau. To estimate the distribution and the abundance of *S. viticola*, other German and Austrian winegrowing areas (Mosel-Saar-Ruwer, Rhineland-Palatinate, Weinviertel) were screened. Vineyards planted with different rootstocks or own-rooted *V. vinifera* vines were chosen to elucidate the host range of this plasmodiophorid within the genus *Vitis*. *S. viticola* was found in different *V. berlandieri* x *V. riparia* hybrids and in roots of *V. vinifera*. Root samples from wild *V. riparia* from the Niagara Peninsula (Canada) were also found to be infested by *S. viticola*. This is the first record of *S. viticola* outside of Europe.

**Key words:** Plant pathogen, plasmodiophorid, *Vitis*.

### Introduction

Plasmodiophorids are obligate endoparasites of plants and straminopiles. Some are well known crop pests. Beyond their parasitic living, plasmodiophorids cause economic loss as vectors for various plant viruses. Most plasmodiophorid species are distributed worldwide but vary considerably in their abundance (KARLING 1968). *S. viticola*, was originally found in only few vines in two commercial vineyards in Germany (HUBER *et al.* 2004, KIRCHMAIR *et al.* 2005). A study conducted in these vineyards indicated, that *S. viticola* is very abundant in infested crops. Up to 44 % of root samples were found to be infested by *S. viticola* and infestation frequencies of examined vines per row ranged from 0 % to 95 % (HUBER *et al.* 2006). The examined vineyards showed differences in growth of vines, ranging from vigorous growth to stunted or dead vines. Growth depressions or death of vines could not be attributed to *S. viticola* because of the presence of additional grapevine pathogens like *Roesleria subterranea* (Weinm.) Redhead or grape phylloxera (*Daktulosphaira vitifoliae* Fitch).

### Material and Methods

To evaluate *S. viticola* infections in commercial vineyards in Germany, roots from the upper 25 cm soil horizon

of 10 randomly chosen vines were sampled. Because of the distribution of grapevine roots in the soil of commercial vineyards, two samples from each vine were taken 10–15 cm to the right or left of the trunk underneath the row (PORTEN and HUBER 2003). Root samples from Austria were provided by grapegrowers. Number of samples of each location was only one to three. Root samples of *V. riparia* in North America were obtained from various locations across the Niagara Peninsula, Ontario, Canada. Sampling was done from the fringe of commercial vineyards planted with *V. vinifera*, general agricultural land planted with tender fruit crops, as well as uncultivated conservation areas. Selected vines were generally healthy in appearance and free of visible signs of disease or pathogenic infection. For the sampling of roots of vines from these locations, soil in the vicinity of vine trunks was removed up to 15–20 cm. Roots were cleaned from soil, transferred to plastic containers, and air dried for 10 d.

For microscopic analysis, roots up to a diameter of 3 mm were cut to pieces of about 5 mm and mixed. Three ml of each sample were put in a plastic Petri-dish and checked for the characteristic auto-fluorescence of sporosori (KIRCHMAIR *et al.* 2005, NEUHAUSER *et al.* 2005) using a Leica MZFLIII Fluorescence-Stereomicroscope (480/40 nm). Fluorescent spots were checked by light microscopic examination. Because auto-fluorescence cannot be observed in all cases (HUBER *et al.* 2006) negative samples were checked by light microscopy. Some of the infected roots were further dehydrated in ascending concentrations of ethanol, transferred to acetone, critical-point dried, mounted on aluminium stubs, and sputtered with gold. Diameters of 31 sporosori from every sample were determined using a PHILIPS ESEM scanning electron microscope.

### Results and Discussion

**Distribution of *S. viticola*:** HUBER *et al.* (2004) and KIRCHMAIR *et al.* (2005) mentioned records of *S. viticola* from the German Rheingau only. In this study we found additional sites in Germany infested with *S. viticola* (Table). In Rheingau, two out of three examined locations showed root infections with *S. viticola*. In Rhineland-Palatinate, only one vineyard was tested and roots were found to be infested with the plasmodiophorid. In the winegrowing region of Mosel-Saar-Ruwer, *S. viticola* was found in one out of three locations. All examined sites were randomly

T a b l e

Confirmed infections of *S. viticola* (sporosori) in rootlets and roots of *Vitis* from different origins. Statistical significant differences ( $p \leq 0.05$ ; Mann-Whitney U-Test) in diameter (Mean  $\pm$  SD, Min, Max) of resting spores are marked by corresponding letters

| Location/Area                               | Host               | Age of vine (years) | Material examined ( <i>S. viticola</i> in roots)                               | Mean $\pm$ SD<br>Min<br>Max     |               |
|---|--------------------|---------------------|--|---------------------------------|---------------|
| Germany                                     |                    |                     |  |                                 |               |
| Kiedrich/Rheingau                           | SO 4               | 12                  | 06 May 2003, <i>leg. et det.</i><br>HUBER, HAMMES,<br>KIRCHMAIR (IB 2003/0001) | 4.38 $\pm$ 0.26<br>3.94<br>4.85 | b, c, e       |
| Dackenheim/Palatinate                       | 5 BB               | 18                  | 14 Sep 2005, <i>leg et det.</i><br>HUBER, HEMMERLING<br>(MJG-040830)           | 4.69 $\pm$ 0.12<br>4.44<br>4.86 | a, c, d, e, f |
| Wehlen/Mosel-Saar-Ruwer                     | <i>V. vinifera</i> | ~ 36                | 29 Sep 2005, <i>leg. et det.</i><br>HUBER, HEMMERLING<br>(MJG-040831)          | 4.81 $\pm$ 0.10<br>4.63<br>4.96 | a, b, d, e, f |
| Geisenheim/Rheingau                         | 5 C                | 17                  | 14 Sep 2004, <i>leg. et det.</i><br>HUBER, SCHOLZ<br>(IB 2004/0300)            | -<br>-<br>-                     |               |
| Geisenheim/Rheingau                         | 125 AA             | 17                  | 07 Aug 2005,<br>no specimen deposited  | -                               |               |
| Canada                                      |                    |                     |  |                                 |               |
| Lakeshore, Lake Ontario                     | <i>V. riparia</i>  | unknown             | 03 Jul 2005, <i>leg. et det.</i><br>HUBER, HEMMERLING,<br>PAGAY (MJG-040828)   | 4.49 $\pm$ 0.07<br>4.32<br>4.62 | b, c, e       |
| Quarry road, Beamsville                     | <i>V. riparia</i>  | ~ 10                | 03 Jul 2005, <i>leg. et det.</i><br>HUBER, HEMMERLING,<br>PAGAY (MJG-040827)   | 4.55 $\pm$ 0.09<br>4.38<br>4.77 | a, b, c, d, f |
| Woodend Conservation Area,<br>Niagara Falls | <i>V. riparia</i>  | ~ 10                | 22 Aug 2005, <i>leg. et det.</i><br>HUBER, HEMMERLING,<br>PAGAY (MJG-040826)   | 4.45 $\pm$ 0.16<br>4.19<br>4.77 | b, c, e       |
| Balls Falls, Conservation Park,<br>Vineland | <i>V. riparia</i>  | unknown             | 03 Jul 2005, <i>leg. et det.</i><br>HUBER, HEMMERLING,<br>PAGAY (MJG-040829)   | -<br>-<br>-                     |               |

chosen. These results imply a widespread distribution of *S. viticola* in commercial vineyards in Germany.

In root samples from the Austrian Weinviertel no plasmodiophorid resting spores were detected. It is presumed that the absence of *S. viticola* in the Austrian vineyards was most likely due to the small sample size and not due to the absence of *S. viticola* in viticultural soils in Austria.

Populations of wild vines (*V. riparia*) at the Niagara Peninsula (Canada) were screened for *S. viticola* infection as well. Four out of 7 samples were found to be infested. This is the first record of *S. viticola* from North America.

Resting spores and sporosori of *S. viticola* (Figure, A-G) from Canada and Germany had a very similar appearance. There were statistically significant differences ( $p \leq 0.05$ ) between the diameters of resting spores of some of the examined populations. This applied to resting spores from the Canadian and the German populations, as well as between populations of these two countries. All examined resting spore diameters were within the range of 4–5  $\mu$ m as given in KIRCHMAIR *et al.* (2005).

**Host range:** The type specimen of *S. viticola* is described from *V. berlandieri* x *V. riparia* hybrids (KIRCH-

MAIR *et al.* 2005). Within this study *S. viticola* was found also on own-rooted *V. vinifera* 'White Riesling' in the Mosel-Saar-Ruwer region. In Canada *Vitis riparia* was found to be parasitized by the plasmodiophorid. The discovery of *S. viticola* on European *V. vinifera*, as well as American *V. riparia* and *V. berlandieri* x *V. riparia* hybrid vines, implies that many if not all species of the genus *Vitis* may be suitable hosts for *S. viticola*. It is part of future research to examine further *Vitis* species for *Sorosphaera* infestations.

**Soil types:** Vineyard soils of Rheingau, Rhineland-Palatinate, and the infested site in Mosel-Saar-Ruwer contained high proportions of clay and loam and therefore had high water-holding capacities. Soils in the non-infested sites were composed of schist and had only low water-holding capacities. Most soils of the examined Canadian sites had high water-holding capacities and imperfect drainage (KINGSTON and PRESANT 1989, SHAW 2005). Woodend Conservation Area, Niagara Falls, is part of a local conservation area, without agriculture. Due to frequent flooding the soils of the Balls Falls Conservation Park are of limited use to commercial agriculture. The Lakeshore region (Niagara-on-the-Lake, Ontario) is cultivated exten-

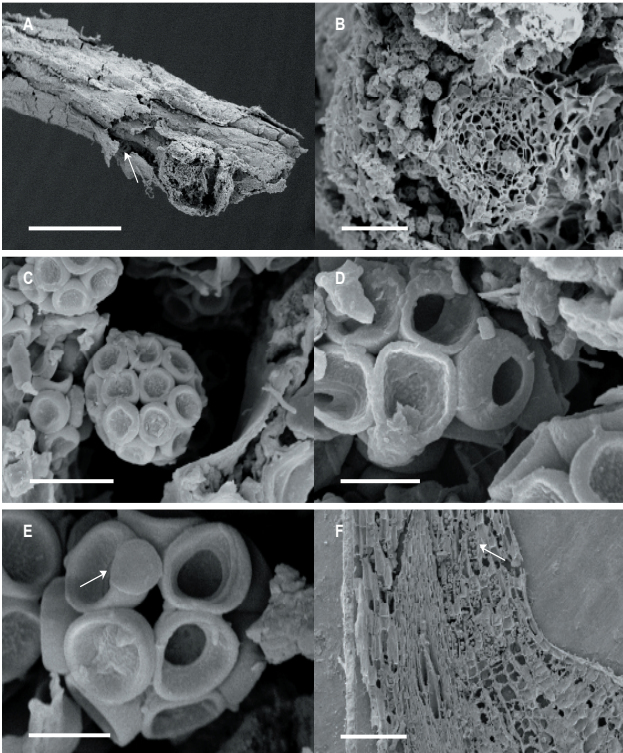


Figure: Electron micrographs of *S. viticola*. **A:** Root of *V. riparia* infected with *S. viticola* (arrow). **B:** Section of **A**. **C-E:** Section of **B**; Closed (**C**) and opened (**D+E**) sporosori, detached and contorted cap of a sporosorus (**E**, arrow). **F:** Cross section of root (*V. berlandieri* x *V. riparia*) with sporosori in parenchyma; many low lying sporosori are hidden from view by cell walls. **G:** Section of **F**. **H+I:** Primary zoospores of *S. viticola*. Bars: **A:** 500  $\mu$ m, **B:** 50  $\mu$ m, **C:** 10  $\mu$ m, **D+E:** 4  $\mu$ m, **F:** 200  $\mu$ m, **G:** 10  $\mu$ m, **H+I:** 5  $\mu$ m.

sively with fruit crops including grapes, peaches, apples and pears. The soils of infested sites in Canada and Germany are perfectly suited for distribution of organisms with free-moving zoospores because of their high water-holding capacities. Given the soils and habitats described above, it can be assumed that the native habitat of this plasmodiophorid consists of sites with wet or marshy soils. Dispersal of plasmodiophorids and infection of a new host requires only a short time (LEGRÈVE *et al.* 2005). Accidentally infested vineyards (e.g. through dispersal of infected roots by cultivating implements) can thus be readily colonized. This is emphasized by the fact that most plasmodiophorids are ubiquitous in traditional agricultural soils.

### Conclusion

Given the above-mentioned findings of the distribution of *S. viticola*, it can be conjectured that the native range of this plasmodiophorid is North America, and that it was

introduced to Europe when it became necessary to graft American rootstocks to *V. vinifera* vines after the phylloxera epidemic at the end of the 19<sup>th</sup> century. This presumption is strengthened by the fact that *S. viticola* was found on the roots of *V. riparia* in the Woodend Conservation Area. Statements about the worldwide distribution and the abundance of *S. viticola* require further investigation. These questions, as well as the molecular characterization of this plasmodiophorid, are the subject of current research. Another focus of interest is the extent of infestation and damage caused by *S. viticola* in commercial vineyards. At present, distinction has to be made between damage caused by *S. viticola* by its parasitizing behavior, and damage caused by *S. viticola* as a vector of plant viruses. Further investigation is also required to determine whether certain environmental stresses such as drought exacerbate *S. viticola* damage to the roots.

### Acknowledgements

The authors wish to thank R. PÖDER, University of Innsbruck, Austria, for providing valuable comments. Thanks also to J. HEMMERLING and M. HOFFMANN of the University of Mainz, Germany. This work was partially supported by the Deutsche Forschungsgemeinschaft (DFG; project EI 87/9-1), the Hessian ministry for environment, the Feldbausch Foundation, Department of Biology, University of Mainz, and the Heinrich-Birk-Gesellschaft e.V. Geisenheim.

### References

- HUBER, L.; HAMMES, M.; EISENBEIS, G.; PÖDER, R.; KIRCHMAIR, M.; 2004: First record of a plasmodiophorid parasite in grapevine. *Vitis* **43**, 187-189.
- HUBER, L.; SCHOLZ, C.; EISENBEIS, G.; RÜHL, E. H.; NEUHAUSER, S.; KIRCHMAIR, M.; 2006: Field distribution of *Sorosphaera viticola* in commercial vineyards in Germany. *FEMS Microbiol. Lett.* **260**, 63-68.
- KARLING, J. S.; 1968: *The Plasmodiophorales*. Hafner, New York.
- KINGSTON, M. S.; PRESANT, E. W.; 1989: *The Soils of the Regional Municipality of Niagara*. Ministry of Agriculture and Food, Toronto, Ontario.
- KIRCHMAIR, M.; NEUHAUSER, S.; HUBER, L.; 2005: *Sorosphaera viticola* spec. nov. (Plasmodiophorids), an intracellular parasite in roots of grape vine. *Sydowia* **57**, 223-232.
- LEGRÈVE, A.; SCHMIT, J. F.; BRAGARD, C.; MARAITE, H.; 2005: The role of climate and alternative hosts in the epidemiology of Rhizomania (Abstract). Proc. VI<sup>th</sup> Meeting of the International Working Group on Plant Viruses with Fungal Vectors (IWGPVVFV), Bologna, Italy.
- NEUHAUSER, S.; HUBER, L.; KIRCHMAIR, M.; 2005: *Sorosphaera veronicae*, neu für Österreich. *Österr. Z. Pilzkunde* **14**, 303-308.
- PORTEN, M.; HUBER, L.; 2003: An assessment method for the quantification of *Daktulosphaira vitifoliae* (Fitch) (Hem., Phylloxeridae) populations in the field. *J. Appl. Ent.* **127**, 157-162.
- SHAW, A. B.; 2005: The Niagara Peninsula viticultural area: A climatic analysis of Canada's largest wine region. *J. Wine Res.* **16**, 85-103.

Received June 2, 2006

