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NEW OR UNUSUAL DISEASE REPORTS

Sorosphaera viticola, a plasmodiophorid parasite of grapevine

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Summary. *Sorosphaera viticola* is a soil-borne, endophytic parasite of grapevine. It is classified within the plasmodiophorids, an enigmatic group of obligate biotrophic parasites of higher plants. *Sorosphaera viticola* has been found abundantly in the roots of *Vitis* spp. in Germany and Canada. This may indicate a global distribution of this root parasite. But its biphasic life-cycle, its soil-borne nature and its co-occurrence with other soil-borne pathogens make an assessment of the disease pattern or a possible yield reduction of this fungus difficult.

Key words: *Vitis*, soil-borne, Cercozoa, clubroot, protist.

In 2003 a new plasmodiophorid plant parasite was discovered in phylloxera-infested root samples from grapevine in a vineyard in the German Rheingau. Although this parasite has since been found repeatedly in phylloxera-induced nodosities, no correlation between phylloxera– and plasmodiophorid – infestation has been found. *Sorosphaera viticola* Kirchm., Neuh. & L. Huber infests the cortical tissue of vine roots, where it forms masses of resting spores which are aggregated to characteristic sporosori (Huber *et al.*, 2004; Kirchmair *et al.*, 2005a; Fig.2, 3, 5). This was the first record of a plasmodiophorid organism in grapevine, although, at the end of the 19th century three "plasmodiophorids" from grapevine were reported: *Plasmodiophora vitis* Viala & Sauvag. (Viala and Sauvageau, 1882a), *P. californica* Viala & Sauvag. (Viala and Sauvageau, 1892b), and *Frankiella viticola* Speschnew (Speschnew, 1901), all of which were observed only in the leaves of *V. vinifera*. However, careful investigations by Massee (1893) indicated that the two *Plasmodiophora* species were phantom species confused with vacuolated tannin vesicles (for the whole story refer to Kirchmair *et al.*, 2005a). The *Frankiella viticola* disease, nowadays known as bitter rot of grapes, is in fact caused by the ascomycete *Greeneria uvicola* (Berk. & M.A. Curtis) Punith. The confusion caused by these parasites was not restricted to grapevine: the whole group of plasmodiophorids has been a challenge to taxonomists since their first discovery. Woronin (1878) described this group as comprising the "most primitive myxomycetes". Traditionally, myxomycetes and therefore plasmodiophorids were treated as primitive fungi (Cook, 1932), but recent phylogenetic studies based on molecular data settled them within the Cercozoa, a group of protists closely related to the Foraminifera and

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Radiolaria (Archibald and Keeling, 2003; Cavalier-Smith, 2003), and which are embedded within the eukaryotic supergroup Rhizaria (Baldauf, 2003; López-García and Moreira, 2008).

Plasmodiophorids are an intriguing group of obligate intercellular parasites of green plants and straminipiles characterised by a multiphasic life-cycle with two main developmental phases (Karling, 1968; Braselton, 1995): (1) primary zoospores encyst and inject their protoplast into the host tissue. These amoebae develop into multinucleate plasmodia and subsequently into thin-walled zoosporangia; (2) secondary zoospores infect host tissues, develop into plasmodia and subsequently into thick-walled resting spores. To date the life-cycle of *S. viticola* has still not been completely elucidated. Secondary plasmodia developing into resting spores were described by Huber *et al*. (2004) and the biflagellate (primary) zoospores were observed by Kirchmair *et al*. (2005a). Until now, the zoosporangial part of the life-cycle was not observed for *S. viticola*, but this is the subject of an ongoing research program (Austrian Science Fund FWF grant T379). Recently, we observed suspicious structures resembling sporangiosori (also referred to as zoosporangia) in the root stem cortical tissue of vines grafted on SO4. As plasmodiophorid zoosporangia lack the characteristic traits whereby species can be distinguished, their identity is currently being tested by PCR-based methods. Nothing is so far known about whether and how *S. viticola* spreads out within the host plant after infection. Direct observation of plant roots indicates that sporosori are crowded within distinct areas but that these sites are unevenly distributed through the root system (Fig.1, insert). Whether *S. viticola* also occurs in other plant parts than the roots is currently being investigated. But until now we have found no evidence that the grapevine plasmodiophorids occur in any other plant tissue than the root cortex. The life-cycle of *S. viticola* seems therefore quite different from that of its probable next relative *S. veronicae,* which forms zoosporangia in the roots of *Veronica* spp. and resting spores in hypertrophies in the stem.

The resting spores of *S. viticola* are aggregated into hollow sporosori. A single sporosorus of *S. viticola* consists of fifteen to thirty (sometimes up to 50) thick-walled resting spores (Fig. 2, 5). These resting spores with their typical assembly are the most prominent part of the life-cycle and the only distinctive structure whereby plasmodiophorids can be identified and determined morphologically. The sporosori of *S. viticola* have a further characteristic trait: using epifluorescence microscopy, most of the sporosori exhibit a characteristic yellow to yellowish-green fluorescence at an excitation of 450–490 nm (Neuhauser *et al.,* 2005; Fig.6). Fluorescence was used by Huber *et al*. (2006) to screen root samples for the grapevine plasmodiophorid in two commercial vineyards. Screening of the fine roots demonstrated an abundant occurrence of *S. viticola* in commercial vineyards (Huber *et al.,* 2006) and in the roots of wild North-American *Vitis riparia* (Huber *et al.,* 2007). Resting spores of *S. viticola* were found in the roots of *V. vinifera*, *V. riparia* and the *V. berlandieri*×*V. riparia* hybrids SO4, 5BB and 5C. Random sampling approaches in Germany and Canada suggest a high abundance of *S. viticola* in vines growing in soils with a high water-holding capacity.

Besides the extraordinary life-cycle and their close interaction with host plants, plasmodiophorids are well known to plant pathologists, as some are also important pathogens of (annual) crops such as those of cruciferous plants (club root, fingers and toes disease caused by *P. brassicae* Woron., reviewed in Karling, 1968) and potatoes (powdery scab caused by *Spongospora subterranea* f. sp. *subterranea* J.A. Toml., reviewed by Merz, 2008). Equally important, some plasmodiophorids are vectors of plant viruses: *Polymyxa graminis* Ledingham transmits more than ten viruses to major cereal crops like barley, maize or rice (reviewed by Kanyuka *et al*., 2003), and *Polymyxa betae* Keskin transmits the virus that causes rhizomania in sugar beets (reviewed by Varrelmann, 2007). The discovery of *S. viticola* therefore stimulated our interest to investigate its role as a plant pathogen and a possible vector for viruses (Kirchmair *et al*., 2005b). But so far the only evident disease symptom correlated with *S. viticola* is a diminutive root necrosis which can serve as entrance portals for additional grapevine pests and pathogens (Fig. 4). Observations in the field indicate that an atypical precocious reddening of the scion may be linked to an infestation with this parasite (Fig.1). This symptom would be

in line with the reddening of *Poaceae* described as typical symptom of infestation with *Sorosphaera radicalis* Ivimey Cook (Cook and Schwarz, 1929). In one vineyard the vines infested with *S. viticola* had poorer growth characteristics than plants without proven infestation, but until now growth depression, yield loss or dieback of vines could not be directly attributed to *S. viticola*. More information about the life-cycle, distribution and abundance of *S. viticola* are needed to understand the impact of this enigmatic organism on grapevine and viticulture.

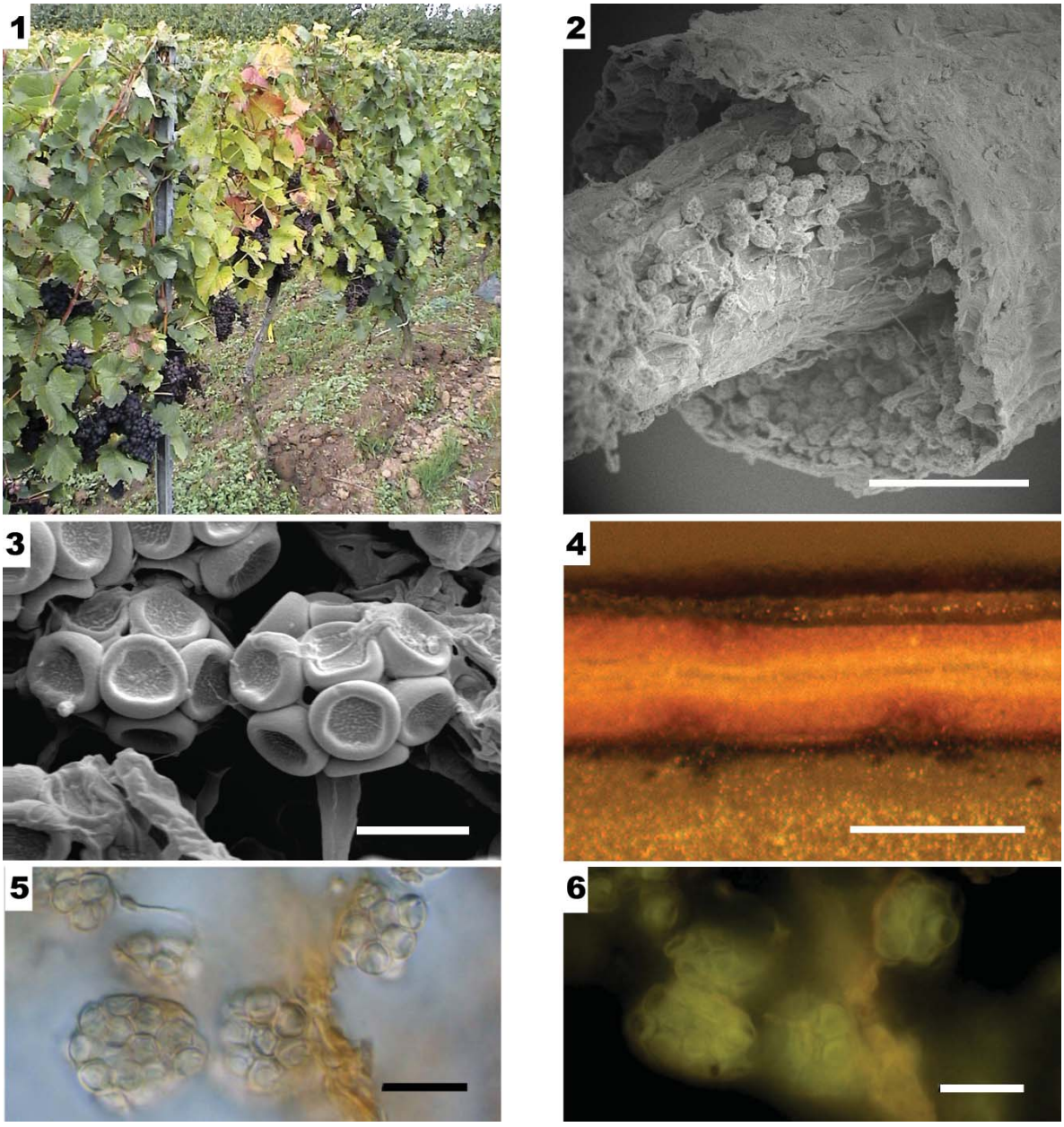

Fig. 1–6. *Sorosphaera viticola* Kirchm., Neuh. & L. Huber. 1. Infested vine exhibiting a precocious reddening. In the insert, arrows mark the infested parts of an excavated rootstock. 2–3. Scanning electron micrographs of sporosori (aggregated resting spores). Bar=100 μm

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