Biology of *Plasmopara viticola* – Approach to a biological control of grapevine downy mildew

Untersuchungen zur Biologie von *Plasmopara viticola* – Ein Ansatz zur biologischen Bekämpfung des Peronosporapilzes der Rebe

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Introduction

Plasmopara viticola, the causal agent of grapevine downy mildew is very well adapted to its host plant. Therefore it is very difficult to develop methods for the biological control which meet the requests of ecological viticulture. One of the possibilities for a biological control of grapevine downy mildew is the usage of the natural resistance potential of the European cultivars of *Vitis vinifera*, more exactly the induced resistance. Another way is the use of specific inhibitors which either stop the infection process or have an effect on the interaction of the pathogen with the host plant. Whereas general information about the resistance reactions in plants is available and the first results were also won for vine, the knowledge about inhibitors is very scanty.

The basic prerequisite to develop methods and strategies for the biological control of grapevine downy mildew is a comprehensive knowledge about the biology of *P. viticola*, especially so with respect to the interaction between pathogen and host.

Since *P. viticola* is well adapted to the host plant, the development of biological methods is a scientific and technical challenge. *P. viticola* is indigenous in the south-eastern part of North America with a damp warm climate where it can be found on different autochtonous *Vitis* species. Due to the coevolution of the pathogen and the host a balanced relationship exists between the pathogen's virulence and the host's resistance. In contrast the European *Vitis vinifera* cultivars are highly susceptible for *P. viticola*; they have got in contact with the pathogen only 120 years ago and no adaptation has been developed in this short period of time. According to our hypothesis, a lack of a fast recognition of the pathogen results in a delayed resistance response of *V. vinifera* cultivars. The growth and development of *P. viticola* is very fast on the host plant and reproduction as well as propagation potential is high. In addition, the growing conditions of grapevine, e.g. regional coverage of viticulture, dense canopy and humid climate, promote epidemics

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Two different propagation strategies, sexual and asexual, occur in the life cycle of *P. viticola*. The former serves the recombination and leads to the formation of perennial oospores. The latter is responsible for the fast propagation during the vegetation period and ensures a quick spreading by a large-scale production of sporangias. While the oospores are leading to primary infections and therefore to the start of the epidemic the asexual sporangia cause the secondary infections and by this means influence the course of the epidemic.
The infection cycle of *P. viticola* is the key process for an epidemic. A cycle can be subdivided into different sections representing the single developmental steps of the pathogen. In recent studies we showed the developmental steps of *P. viticola* by means of epifluorescence microscopy (FM), confocal laser scanning microscopy (CLSM) and low temperature scanning electrone microscopy (LTSEM). In addition we carried out investigations on the cytology of encystation and the formation of the penetration peg and the first growing stage s of the hypha too. In these studies we also characterised the temporal and spatial course of the infection cycle.

Studies on the developmental biology of *Plasmopara viticola*

The infection process *sensu strictu* is the first step of the infection cycle. It starts with the attachment of the sporangia to the surface of the host plant and ends with the formation of a penetration peg. Thereafter the infection process only continues if the humidity conditions permit the release of zoospores out of the sporangia and if it is possible for the zoospores to reach the stomata. At the stomata the zoospores accumulate specifically around the porus and encystate by forming a cell wall. The encystated spores form an infection peg which penetrates the host. After this, the incubation period follows, in which the pathogen colonises the intercellular space of the host tissue and parasites the host cells by the formation of haustoria. The penetration peg forms a substomatal vesicle of which a hypha grows out. Hyphal growth is slow within the first 24h but after that they branch and colonise the intercellular space of the mesophyll very fast. The first symptoms of the disease are macroscopically visible at the end of the incubation period when the host tissue is completely filled up with the pathogen's mycelium. The latency is the period between infection and sporulation. Towards the end of the latency period, *P. viticola* forms a secondary substomatal vesicle from which sporangiophores emerge during the night and favourable humidity conditions. Within 7 hours the sporangiophores form third order branchings with sporangia developing at the tips. The sporangia are taken off and are spread by wind or water drops. After the end of the latency, repeated sporulation can take place until the host tissue necroses due to the parasitation. Our studies revealed that the complete infection cycle of *P. viticola* is a highly co-ordinated temporal and spatial process, in which host plant and pathogen interact with each other. It is particularly interesting that the growth pattern of the mycelium depends on the type of the plant tissue. In leaves the hypha develop mainly in the mesophyll and the expansion of the mycelium is limited to the veins. In the rachis of the inflorescence and the bunch, however, the mycelium of *P. viticola* grows along the parenchyma and is not impeded by the veins.

An epidemic progresses only if several repeated infection cycles take place. This sequence of infections, the so-called infection chain, ensures a sufficient propagation and spreading of the pathogen. Therefore the most appropriate method to control the epidemic is to interrupt the infection chain. Some of the events during the infection process mentioned above represent very sensitive stages of the development. These stages offer an approach for the specific control by means of biological methods, such as treatments with inhibitors. Due to our basic work a method with which the inhibitory effect of natural compounds can be tested is now available. As a future prospect, we are going to characterise natural inhibitors, especially compounds of the secondary metabolism of plants. In addition, we are strengthening our efforts to clear up more details of the development of *P. viticola*, particularly physiological and cytological processes during the infection stages and the host-pathogen-interactions.

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