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Downy mildew is able to carry out its sexual cycle on resistant grape varieties

Sourced from the research article "Evidence for sexual reproduction and fertile oospore production by *Plasmopara viticola* on the leaves of partially resistant grapevine varieties" (ACTA Horticulturae, 2019)[1].

>>> Previous studies have shown that new resistant grape cultivars limit epidemics caused by asexual reproduction of downy mildew. However, until now, there was a lack of knowledge on the sexual phase of the pathogen. This study demonstrates that host resistance has little effect on the sexual cycle of downy mildew. Thus, the pathogen can complete its life cycle (asexual and sexual phases) on partially resistant grapevine varieties. The persistence of pathogen populations on partially resistant varieties from one year to the next represents a major challenge for the sustainability of genetic resistance of grapevines. <<<

Vitis vinifera cultivars are very susceptible to downy mildew[2]. Fungicide treatments are currently the only available method to control this pathogen[3]. However, their systematic use leads to negative effects, including possible impacts on the environment, as well as the emergence of resistances to fungicides[4] which reduce the efficiency of a growing number of products[3]. The use of plant genetic resistance is an effective alternative to fungicides. It makes it possible to limit, delay or prevent the pathogen's infection cycle. The selection of genetic resistance is at the heart of several grapevine varietal improvement programs in Europe, whether in France, Germany, Switzerland or Italy.

Resistant varieties to powdery and downy mildews, with organoleptic characteristics suitable for the production of quality wines, were recently created by cross-breeding. These varieties carry resistance factors derived from wild *Vitis* of American and Asian origin. Recently registered in France, these varieties show partial resistance to downy mildew, which limits mycelium growth and sporulation, thus reducing epidemics due to the asexual multiplication phase of the pathogen during the season. Until now, the host resistance effect has not been assessed on the sexual phase of the downy mildew cycle (Fig. 1). Indeed, *Plasmopara viticola* has a mixed reproduction system based on several cycles of asexual reproduction and a single sexual generation each year that takes place in the fall (Fig. 2)[5]. To answer this question, during two years, this study evaluated the production of winter eggs ("oospores") and the success of subsequent infections ("primary infections"), on resistant varieties, during the sexual generation. The resistant varieties carrying two types of factors (**Rpv1** and **Rpv3**), alone or in combination, were studied.

The approach used

The plant material studied is derived from the descendants of a cross between *Mtp3082-1-42* and *Regent*, as well as Merlot which served as a sensitive control.

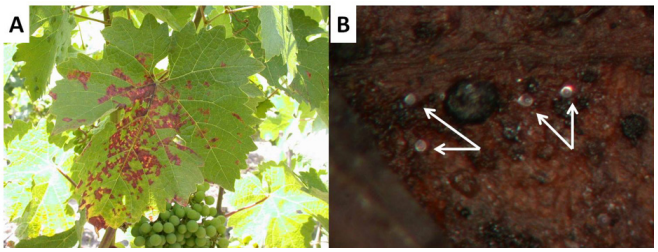


Figure 1. Downy mildew sexual reproduction on vine leaf: A) mosaic form, B) germination of macrosporangia in spring (Pictures: Delbac & Rolle, INRAE).

The parent *Mtp3082-1-42* transmitted the resistance factor **Rpv1** and the parent *Regent* transmitted the factor **Rpv3**. **Rpv1** and **Rpv3** have partial to high resistance[6]. This crossbreeding is at the origin of the first resistant varieties from the INRAE-ResDur programme, recently made available for purchase in France.

The procedure for handling and evaluating the presence of oospores and controlling their germination has been adapted from a method previously published by INRAE[7]. Four possible combinations of resistance factors were studied: **Rpv1**, **Rpv3**, **Rpv1/Rpv3** and lack of resistance. For each genotype studied, the experiment design is as follows:

(**A**) collection of leaves showing "mosaic" symptoms in the fall (Fig. 1A). We assessed the presence of oospores in the necrotic part of the leaf limb and then collected leaf discs which we stored in plaster modeling tubes buried under natural climatic conditions from October (year n-1) to April (year n).

(**B**) evaluation of the number of macrosporangia formed during the spring, April (year n) (Fig. 1B).

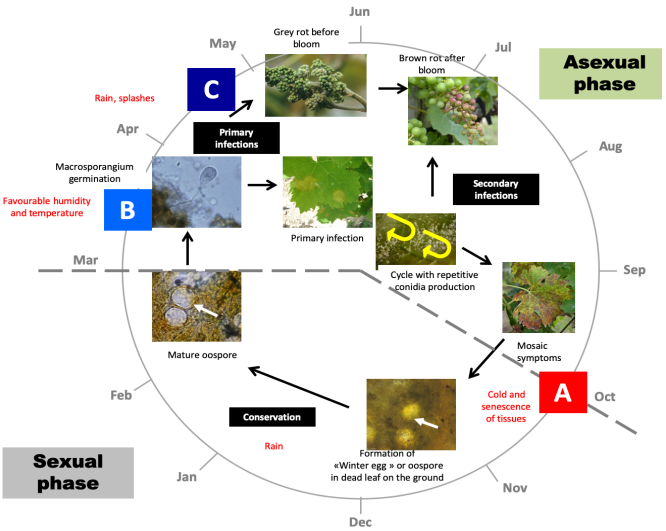


Figura 2. Vine downy mildew cycle and position of the experimental stages carried out (Pictures: Blancard, Bugaret, Delbac & Mazet, INRAE).

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(**C**) inoculation of Cabernet Sauvignon (a susceptible variety) leaves, by the macrosporangia formed in order to obtain primary infections of the pathogen.
These protocols are detailed in Figures 2 and 3.

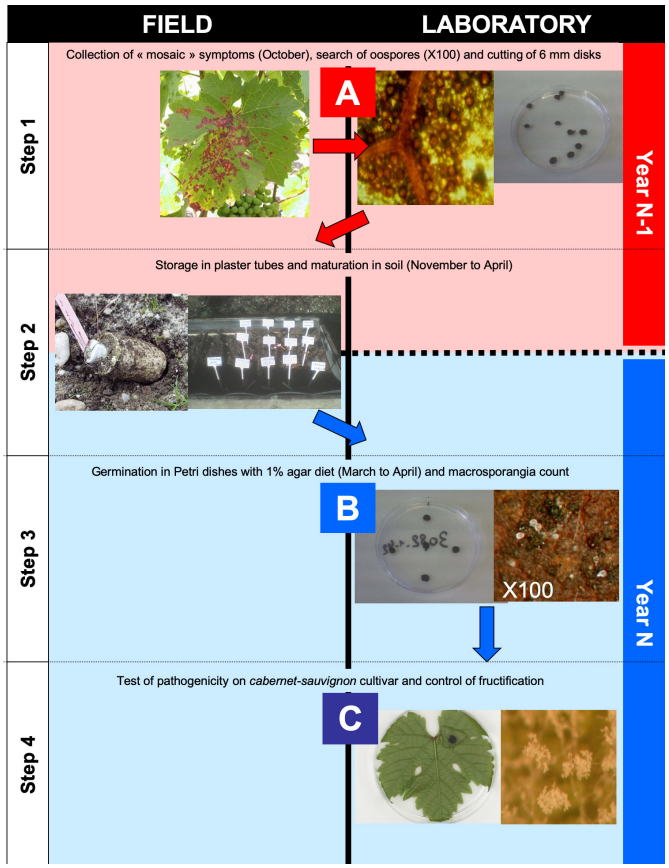


Figure 3. Experimental steps for monitoring the sexual phase of grapevine downy mildew (Pictures: Delbac, Rolle & Tran Mahn Sung, INRAE).

■ Oospores from the sexual cycle on resistant varieties

Downy mildew produces winter eggs on resistant varieties in the fall. The production of oospores is higher on susceptible varieties than on resistant varieties. **Rpv1** and **Rpv3** reduce oospore production of the pathogen, but do not prevent it.
The oospores germinate the following spring. Varieties carrying **Rpv1** factors showed a high production rate of macrosporangia. For all genotypes, we found a high variability in production of macrosporangia related to the date of observation, i.e. more macrosporangia are produced in early spring.

Macrosporangia produce primary infections that are observed for all combinations of resistance factors. However, significant differences between varieties are observed, with one of the genotypes carrying the combination of **Rpv1/Rpv3** having a high success rate of primary infections in contrast to **Rpv3**.

■ What to retain?

Resistant varieties limit the ability of downy mildew to multiply during asexual cycles, thus controlling partially the disease during the growing season. Data from the OSCAR national observatory show that growing resistant varieties can reduce fungicide use in vineyards by more than 80 %[8].
From an evolutionary point of view, there is a risk that strains of downy mildew will appear that are able to bypass the vine's resistance. This is a classic phenomenon when resistance genes are deployed on a large scale. Normally, although the number of downy mildew spores arriving on a plant is significant, it is assumed that the beginning of the deployment of resistant varieties in the vineyard reduces the chances of fixing a favourable mutation during the asexual multiplication cycles of the pathogen. However, the results of this study show that vine resistance does not block the sexual reproduction phase of the pathogen. The achievement of the sexual cycle thus offers the pathogen the possibility of recombining favourable mutations that could eventually allow it to bypass resistance more rapidly.
Complementary epidemic management strategies can be put in place to limit the emergence of virulent strains of downy mildew. These strategies may include the long-term monitoring of the virulence of the pathogen (e.g. National observatory OSCAR)[8], the application of a limited number of fungicide treatments (e.g. 1 or 2 fungicide applications around flowering, which at the same time controls attacks by other pathogens not targeted by resistance genes such as anthracnose and black-rot[8]), the use of plant defense stimulators or the implementation of prophylactic measures (e.g. removal of senescent leaves in the fall)[1].
In all cases, it is the diversity of control methods that will make it possible to create more disease-resilient winegrowing systems. Thus, the use of varietal resistance against downy mildew in vines must be considered as an additional tool to build winegrowing systems that are more efficient in the use of pesticides. ■

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