Phytopathol. Mediterr. (2011) 50, 3–44

REVIEW

Plasmopara viticola: a review of knowledge on downy mildew of grapevine and effective disease management

Cesare GESSLER[1], Ilaria PERTOT[2] and Michele PERAZZOLLI[2]

[1]Plant Pathology, Institute of Integrative Biology, ETH Zürich, 8092, Switzerland
[2]IASMA Research and Innovation Centre, Fondazione Edmund Mach, S. Michele all'Adige 38010, Italy

Summary. The oomycete *Plasmopara viticola* is native to North America and was accidentally introduced into Europe at the end of the 19th century, where it caused widespread damage to the grape industry. Since that time, the damage caused by this plant pathogen has generally been controlled with multiple fungicide applications. Modern fungicides applied as prescribed by weather-based warning systems can effectively prevent any damage that might be caused. Alternatives to chemical treatments, such as the use of biocontrol agents or resistant cultivars, currently play a marginal role in controlling this disease. Until the middle of last century, research efforts were mainly concentrated on optimizing the application of copper fungicides and developing new molecules and formulations for controlling the disease. During the second half of the last century, highly efficient products for control were available, so research efforts moved toward optimizing and minimizing chemical control, mostly through the use of weather-based warning systems based on complex biological models. In the last 20 years, parallel to the development of technologies for genomic and transcriptomic analyses, host-pathogen interactions and population genetics have captured the interest of researchers in this field. Breeding for resistance against downy mildew has always coexisted with chemical control. However, the results of these breeding programmes have traditionally been cultivated only in marginal areas and organic production systems. This review traces the history of European knowledge of *P. viticola*.

Key words: breeding, control, oomycete, resistance, wine.

Introduction

The history of European grape-growing can be divided into three periods. The first (before 1845) was characterized by the absence of major phytosanitary problems. This was followed by a troubled half century during which European grape crops were faced with the arrival of three major problems: first powdery mildew, then phylloxera and, finally, downy mildew. The following years were characterized by a search for solutions for these problems and a period of intensive use of chemical protection lasting until the present. We will follow the accumulation of knowledge concerning the causal agent of downy mildew through until 2010. The number of publications concerning this disease (over 3000, since 1910) is far more than can be discussed in a single review, so in the interest of brevity, we have chosen not to discuss papers that describe fungicide tests, cultivar resistance tests or field tests of warning systems that have been previously described and tested. Up to the 1970s many overlaps can be found with the detailed and precise review on *P. viticola* by Galet (1977) in his book on diseases and parasites of the grape.

Corresponding author: C. Gessler
Fax: +41 44 6321572
E-mail: cesare.gessler@agrl.ethz.ch

Classification

The downy mildew to which this review is dedicated was first noticed and described at its centre of origin north east of the USA. The fact that a microorganism is the cause and not a consequence of this disease was not clear until 1876, when Farlow (Farlow, 1876) correctly described this disease and attributed it to *Peronospora viticola*. Previously, Saunders (1862 cit in Gregory, 1915) and Lippincott (Lippincott, 1866) had stated their view that the presence of injured and/or decaying vine leaves allowed the formation of mildew. They named ozone as the agent of injury. Now, mildew was known to develop in absence of ozone so the primary cause was injuries and not *P. viticola*.

The oomycete that causes downy mildew (Figures 1–5) was first collected by Schweinitz (cit in Gregory, 1915) in the northeastern USA in 1834 and was classified as the fungus *Botrytis cana* Link. In 1848, Berkley and Curtis re-classified it as the new species *B. viticola*. De Bary, who had already successfully studied the life cycle and classification of the potato late blight fungus *Phytophthora infestans*, described the asexual and the sexual stages of the grape pathogen and placed it in the genus *Peronospora*, as *P. viticola* (De Bary, 1863). Only 20 years later, Schröder (Schröter, 1886), using the clear differences between several of the *Peronospora* fungi, separated this genus into *Peronospora* and *Plasmopara*.

Plasmopara sporangiophores (Figures 6, 7 and 8) grow from intercellular mycelium (Figure 9) with monopodial branching, mostly at right angles, with ultimate branches (sterigmata) that are mostly trichotomous. The sterigmata of *Peronospora* are always dichotomous. *Plasmopara* produce sporangia in which zoospores are produced and released; whereas *Peronospora* always germinate by means of a germ tube (conidia). Berlese and deToni (Berlese and de Toni, 1888), using Schröder's classification system, renamed this microorganism *Plasmopara viticola*. A classification earthquake finally moved the oomycetes from the Fungal Kingdom to the heterokonts, acknowledging their close relationship with photosynthetic organisms such as brown algae and diatoms (Van der Auwera *et al.*, 1995). For example, the cell walls of Oomycetes are composed of cellulose rather than chitin and septations are found only in older mycelia, which are coenocytic. In their vegetative state, oomycetes have diploid nuclei, whereas the nuclei of analogous fungi are haploid (Ascomycetes) or dikarontic (Basidiomycetes). Many Oomycetes produce self-motile zoospores with two flagella.

The world-wide spread of this oomycete

Plasmopara viticola is endemic on wild *Vitis* species of North America. It was first observed in Europe in 1878. It was probably introduced into Europe with American grape cuttings used to replant the French vineyards destroyed by phylloxera. Its appearance in Europe was not a real

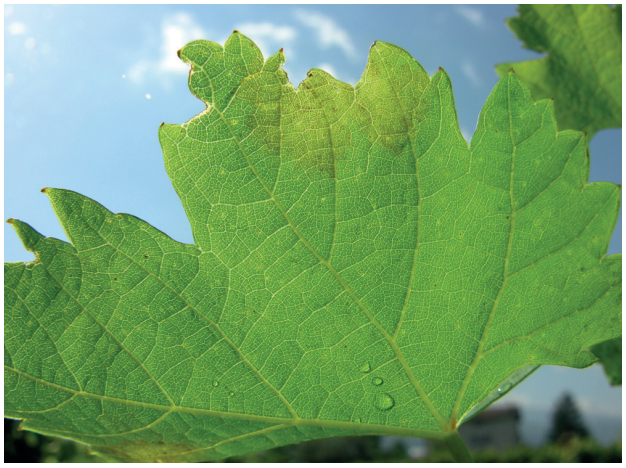

Figure 1. Oilspot caused by *Plasmopara viticola*.

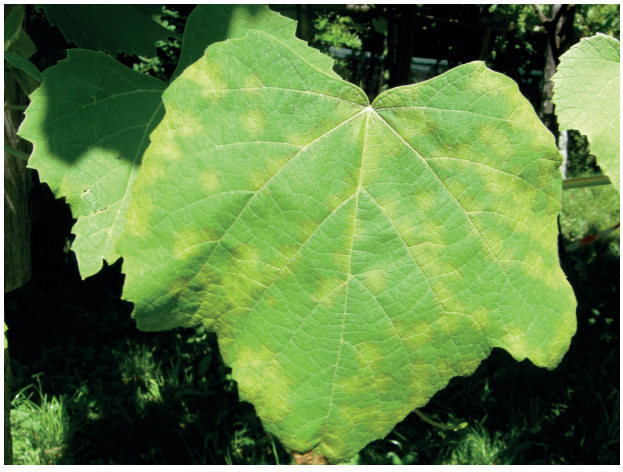

Figure 2. Lesions on the adaxial leaf surface caused by secondary infections in July.

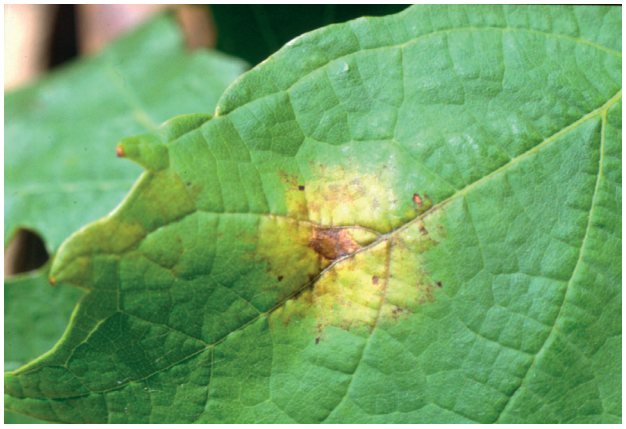

Figure 3. Old oilspot with necrosis on the adaxial leaf surface.

Figure 4. Sporulation of *Plasmopara viticola* on abaxial leaf surface.

Figure 5. Sporulation on a grape cluster before bloom.

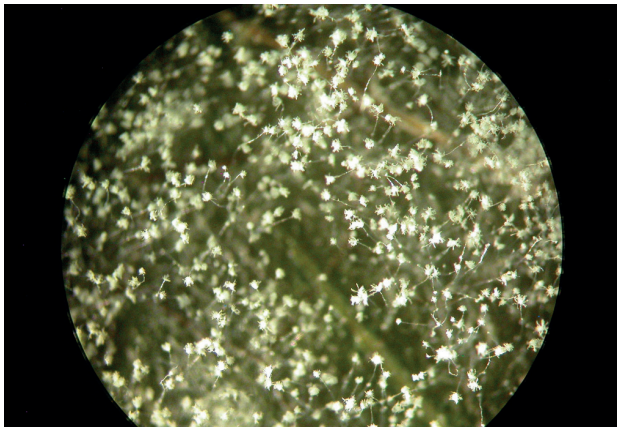

Figure 6. Sporangiophores and sporangia of *Plasmopara viticola*.

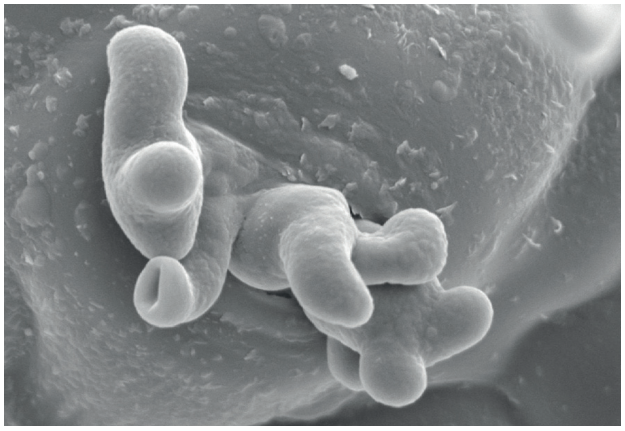

Figure 7. Sporangiophores in formation exiting from a stoma.

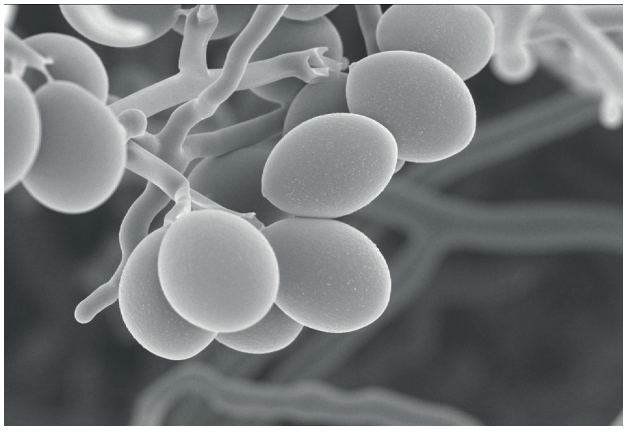

Figure 8. Sporangia on sporangiophores.

surprise, as several voices had expressed concern (Cornu 1872 cited in Müller and Sleumer, 1934) about the danger of the accidental introduction of "*Peronospora viticola*". After Thiemann, Planchon and Farlow identified the causal agent of the new disease as *P. viticola*, it was rapidly identified throughout France, northern Italy, including South Tyrol, and nearby Austrian regions (1879). It was identified a year later in the Alsatian and Mosel region (Germany), advancing in 1881 to Eastern Europe, Turkey and Greece and westward toward Spain and Portugal. It was assumed that, in Europe, the disease would be limited to grapevine foliage, as was the case in the first years (Farlow cit in Müller and Sleumer 1934), but, as early as 1884, berry symptoms were observed that were similar to the symptoms seen in America (brown rot, "Lederbeeren", imbrunimento degli acini, peronospora larvata) (Figure 10). Sporulation of the pathogen, however, was not usually seen, which often led to the assumption that this berry damage was not caused by the same causal agent. However, Pichi (Pichi, 1890) clearly showed that this berry damage could also be attributed to *Peronospora*. Müller and Sleumer (1934) hypothesized that the passage of *Peronospora* from leaves to berries was related to the timing of the infections in the vineyards. In the first years, the disease generally appeared at the end of July or beginning of August (when berries were no longer susceptible). From the 1890s onward, the first signs of disease began to appear at the end of May or early June. By 1905, berry brown rot was generalized and often completely destroyed the affected crops.

From the beginning of the 20th century, the disease was clearly a huge problem for European viticulture (Gäumann, 1927). Epidemics in Europe were sporadic and irregular. In the years in which weather conditions were favourable and sufficient control measures were not yet available or were not applied (1900, 1905. 1906, 1910, 1912, 1913, 1916, 1917, 1930 and 1932), serious damage was caused to viticulture in Germany, France and Switzerland. For example, in 1915, 70% of the French grape crop was lost to *P. viticola* (Cadoret, 1923, 1931). In 1930, 20 million hl of wine were lost in France. From 1907 to 1916, downy mildew was responsible for a 33% reduction in the total vine-growing area in the Baden province of Germany (Müller, 1938). Significant damage was reported in Italy (Baldacci, 1954) in 1889, 1890, 1903, 1910, 1928, 1933 and 1934.

During the Second World War, this disease caused considerable damage. However, the lack of copper contributed to this situation more than unfavourable weather conditions (Hadorn, 1942; Stellwaag, 1943). As an example, Hadorn reported that the normal Swiss copper requirements for agriculture in 1942 would have amounted to 1550 tons but, due to the war situation, the government allocated only 690 tons, of which 300 tons were allocated for the potato crop; 12.5 tons were allocat-

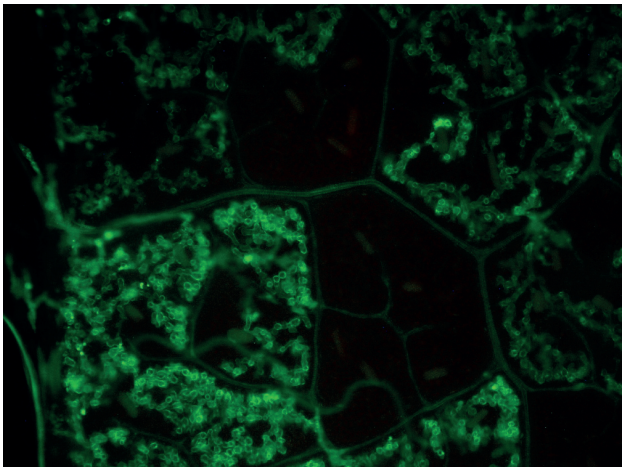

Figure 9. *Plasmopara viticola* hyphae in infected grape leaf tissue after aniline blue staining (Nikon Eclipse80l, Em filter 420–490 nm).

Figure 10. Symptoms caused by *Plasmopara viticola* on a developed cluster of grape in full summer.

ed for beans, tomatoes, celery, and onions; a maximum of 50 tons were allocated for orchards and 320 tons were allocated for viticulture (Hadorn, 1942). Little was published in war-affected countries during those years. However, we must assume that very little copper-based protection was possible. In the years after the Second World War, the grape-producing industry was able to avoid large-scale losses thanks to the widespread use of highly effective chemical control measures.

Description of the life cycle

The sexual cycle

In 1880, Millardet identified oospores of *P. viticola* in leaves of European vines (Millardet, 1883), and Pichi (1890) followed with drawings of the oogonia and antheridia. Ravaz and Verge (Ravaz and Verge, 1913) and Gregory (Gregory, 1915) described the whole life cycle of this organism in great detail and the quantitative and time-related aspects of their work are quite noteworthy. Arens (Arens, 1929a) observed oospores at sites with mosaic symptoms, which he attributed to primary infections occurring as early as late June. This phenomenon was observed not only on the highly susceptible European cultivars, but also on more or less resistant American cultivars. Oospores can be produced at any temperature, but they seem to be preferentially produced under rather dry conditions, when sporulation is impeded, or as the leaves senesce (Grünzel, 1961). Their numbers are generally extremely high, easily reaching 250 oospores/mm^2 in a polygonal fleck, which is usually delimited by the veins of a leaf on which an oil spot is present. These oospores mature over the winter and only mature oospores will rapidly germinate and form sporangia once exposed to a water film.

Attempts to biologically determine the date of oospore germination were made as early as a century ago (Gregory, 1915; Ravaz and Verge, 1921; Arens, 1929b). These early efforts were mostly based on microscopic observations of decaying overwintered leaf material soaked in water and incubated for various periods at selected temperatures. The end of the germination process was determined by the presence of primary zoosporangia (macrosporangia). The presence of actively swarming zoospores in the water surrounding soaked leaf debris was used less often (Müller and Sleumer, 1934). However, coupling the above zoospore assay with counts of zoospores swarming around a floating grape leaf disc, and using the *P. viticola* infection and consequent establishment rates to indicate germination and the production of zoospores rather than microscopic observations, facilitated the determination of the exact date(s) on which germinating (infective) oospores would be present (Hill, 1998). Again, in the older literature, we find the notion that oospores germinate over a longer period. In a particular spring, such germination in the field might be distributed over 2 to 3 months. Some of the oospores might even germinate only the next spring, once they had been brought to the surface as the soil was worked (Gregory, 1915).

In keeping with this line of thinking, Müller and Sleumer (1934) stated "in our region primary infection occurs mostly from the end of April through the whole of July. In any case, several spring infections can be explained once we know that not all oospores germinate at once."

Their explanation for why in some years, including years with favourable infection conditions, the severity of *Peronospora* infection was insignificant is also very interesting. As an example, they cite the year 1932 (Freiburg im Breisgau, Germany), during which a severe first outbreak could have been expected as heavy rainfall was recorded from 15–17 May. However, as the bud burst of the vines did not occur until 7–9 May, little leaf surface area was available for infection and most of the oospores had no effect.

Sung and colleagues (Sung and Clerjeau, 1989; Tran Manh Sung *et al.*, 1990) constructed a model that is currently used at several sites to determine infection risk. Generally, a correlation exists between low rainfall during the winter-spring period and slight epidemics, although an abundance of rain during the same period does not appear to be sufficient to develop an epiphytotic in the absence of at least two primary infection events (Baldacci, 1947). Mature oospores germinate best if their outer walls are ruptured, possibly as a result of a light freeze and sufficient humidity. The germination of oospores requires soil temperatures of 12 to 13°C (occasionally 11°C) and moisture.

More recent research indicates that an accumulation of days with favourable temperature conditions is necessary for infection, which Ge-

hmann has defined in Germany as 160°C/days above a threshold of 8°C, from the 1st of January, as measured 2 m above ground (Gehmann *et al.*, 1987). However, this model only predicts the date on which the first oospores will mature and is not quantitative in nature. In Germany, a model for predicting periods of high oospore germination activity was developed based on daily mean temperature, relative humidity and precipitation data, which are used to calculate a daily index to determine the number of incubation days needed to induce germination (Hill, 2000). In a study carried out in the Po Valley of Italy, Rossi *et al.* (Rossi *et al.*, 2008) and Caffi *et al.* (Caffi *et al.*, 2009) used not only accumulated degree-days, but also rain and humidity conditions to accurately predict oospore germination over time. Other oospore-related simulations and models have been focused on producing qualitative predictions of the precise moment that the first oospores will germinate in the spring and modeling the ensuing epidemics based on knowledge of the asexual portion of the pathogen life cycle (Orlandini *et al.*, 1993; Orlandini *et al.*, 2008).

Once the macrosporangia have been spread directly onto leaves by splash or have become airborne with rain splash and turbulence and then been deposited onto the leaves, zoospores are released. It is assumed that the necessary conditions for infection are if not equal then at least similar to the conditions for infection by zoospores released from secondary sporangia. Baldacci's observations (Baldacci, 1947; Baldacci and Refatti, 1956) led him to state that primary infections do not occur unless the minimum atmospheric temperature is about 10°C, at least 10 mm of rain have fallen in the previous 48 h and that the vines have developed leaves measuring 6 to 8 cm^2 (the corresponding shoot measurement being about 10 cm) (see below, Warning systems and models).

Incubation time was a subject of early studies and several publications from that time include graphical representations of incubation time relative to temperature (Müller and Sleumer, 1934; Merjanian and Lipetzkaya, 1936). However, daily variations in other climatic conditions often necessitated adjustments to these models. An alternative to the average daily temperature was found in the sum total of 61 degrees of 'active' or 'effective' average daily temperatures (i.e., temperatures above 13°C and below 24°C). The actual length of the incubation period was obtained by dividing 61 by the average daily 'active' temperature calculated from the daily minimum and maximum (Merjanian and Lipetzkaya, 1936). Further corrections to the original model concerned relative humidity. Sohad proposed considering only those hours in which the relative humidity is over 60% (Sohad, 1943).

Secondary sporulation and infection

The conditions under which *P. viticola* can sporulate in susceptible tissue under controlled conditions were analyzed in detail by Blaeser (Blaeser, 1978; Blaeser and Weltzien, 1978; 1979). Most simulation models still rely on Blaeser's parameters, including a minimum of 98% relative humidity and 4 h of darkness, a minimal temperature of 13°C and an optimal temperature of 19°C. Sporulation proceeds in darkness, but not in the light, and is completed within 7 h. It is inhibited by irradiation with white light (Rumbolz *et al.*, 2002), near-UV light of 310–400 nm or green light (500–560 nm) at intensities >3–3.5 Wm-2 (Brook, 1979). The lifespan of the zoosporangia decreases as the water saturation deficit increases. The zoosprongia are thought to be dispersed by rain splash, as they are detached by water. Zoospore liberation and infection occur in the presence of a water film, following a minimum number of degree-hours of 50°C × h. Lesion productivity (sporangia/lesion) does not decline with lesion age in the absence of weather suitable for sporulation. However, the productivity of all lesions declines rapidly over repeated cycles of sporulation (Kennelly *et al.*, 2007).

A very detailed, literature-based review of the biology and epidemiology of *P. viticola* in Greece was presented by Zachos (Zachos, 1959), complemented with the results of the author's own experiments. That report was prepared in response to the fact that vine growers of the Vello region of Greece were applying Bordeaux mixture to a great extent, but not always seeing the expected level of disease control.

Sites of infection

In his treatise on plant diseases, Prillieux (Prillieux, 1895) described the germination of "conidia" with zoospores and their encystment, the formation of germination tubes and penetration of plant

cuticles. Only 15 years later, Müller-Thurgau (Müller-Thurgau, 1911) showed that the downy mildew pathogen did not enter the leaf when only the upper side of the leaf was inoculated. In contrast, inoculation of the lower side led, in most cases, to the development of symptoms. From microscopic observations, he concluded that the zoospores were able to swim toward the stomata, settle on or near them, geminate, and then penetrate the leaf through the stomata within 3 h at 20°C, forming a swelling in the stomatal cavity with or without light. This work clarified that infection can occur only through stomata. This understanding, as simple and self-evident as it appears to us, was essential for the control of the disease (see below). It also contributed to the understanding of the ontogenic resistance observed in berries and, to a lesser extent, in leaves (Kennelly *et al.*, 2005).

Quantification of oospore germination

Arens (Arens, 1929a) presented the first very thorough study of oospore germination. He determined how long it took oospores in leaf material left outside to germinate. First, he noticed that the germination of the oospores accelerates as spring advances, "the period required for germination being reduced from 8.8 days in material laid in the germinating bed on 21st March, 1926, to 4.4 days on 27th April and 2 days on 10th June".

Secondly, he observed that while germination on overwintered material occurred in the laboratory any time between January and July, no germination was observed in material left outside in late summer. In contrast, the same material gave rise to primary sporangia occasionally in December and very seldom in November. For unknown reasons, some individuals require a second winter in order to complete their dormant period. Germination was found to occur within a temperature range of 13–33°C, with optimal germination observed at 25°C. Low temperatures curtail the dormant period, whereas dry conditions prolong it and may, if protracted, injure the oospores. Arens also suggested that short periods of rain conveyed the primary sporangia or zoospores to the leaves (Arens, 1929a). Several studies have reported that oospore germination is most abundant in the early spring. However, germination on leaves that have passed the winter under natural conditions usually continues until June (Clerjeau, 1989; Jermini *et al.*, 2003; Pertot and Zulini, 2003; Sung and Toffolatti *et al.*, 2006; Kennelly *et al.*, 2007).

Most assays designed to determine the timing of oospore germination have involved the collection of intact or crushed leaves from a specific site and specific, local overwintering conditions. From this collected material, samples are taken at regular intervals and kept at a fixed temperature (20°C) in water or 100% humidity, and the time lag until oospores germinated can be determined, often also quantitatively (Sohad, 1943; Laviola *et al.*, 1986; Sung and Clerjeau, 1988; Burruano *et al.*, 1990; Gherardi *et al.*, 1999). In these experiments, in which many oospores are kept under identical conditions, germination over several months is generally recorded (Jermini *et al.*, 2003; Pertot and Zulini, 2003; Rossi *et al.*, 2008).

It appears reasonable to assume that overwintering conditions are particular for each leaf. Therefore, in each vineyard, a wide variety of conditions are present, suggesting that oospore germination will occur over a long period. Recently, Caffi (Caffi *et al.*, 2011) reported that the germination of oospores collected in a particular vineyard and kept under natural, variable conditions extended over several years. Caffi's findings contradict the widely held assumption that the supply of oospores is depleted shortly after bloom, as well as the previously accepted conclusion that oospores contribute to the downy mildew epidemic only as initial inoculum.

With the development of tools allowing the genetic characterization of single genotypes and identification of the genotype of a single lesion, it became possible to determine whether an epidemic was indeed caused by one or a few clones deriving from a few primary infections, as had been assumed. RAPDs were the first type of markers to be used for this work. Although the power of this type of marker is limited, RAPDs can be used to detect genetic differences between genotypes in a single comparative experiment. However, since *P. viticola* is an obligate biotroph, doubts remain as to whether other genetic material collected with the sporangia may have been the source of the observed differences. In 1998, Kump reported a high level of variability between batches of sporangia collected from individual leaves with single oil spots in a vineyard in northern Switzerland. Attributing a specific genotype and, therefore, a

unique oosporic origin to each specific RAPD banding pattern, they concluded that the ratio between primary and secondary lesions was higher than expected and recommended that the quantitative role of oospores in epidemics of grape downy mildew be reconsidered (Kump *et al.*, 1998). Another step forward came with the development of microsatellite (SSR) markers, which are sufficiently specific to withstand the contamination of the samples with any quantity of foreign DNA and still detect differences between oomycete genotypes (Gobbin *et al.*, 2001; Gobbin *et al.*, 2003a; Delmotte *et al.*, 2006; Scherer and Gisi, 2006). In a study in which four SSRs were used, the majority of the collected isolates had unique allele patterns and, on the basis of this, were considered to be genetically distinct from one another. Over the course of the season, slowly increasing numbers of clonal genotypes were observed, but new genotypes continued to appear as well. This observation proved that new oosporic infections and/or migration processes play an important role in downy mildew epidemics over the course of the growing season (Gobbin *et al.*, 2001). These findings have been confirmed in several vineyards in Europe (Rumbou and Gessler, 2004, 2007; Gobbin *et al.*, 2005; Gobbin *et al.*, 2006; Loskill *et al.*, 2006), South Africa (Koopman *et al.*, 2007) and the northeastern USA (Kennelly *et al.*, 2007). In Western Australia, the genetic variability of this oomycete appears to be so low that oospores were detected in only 1 of 16 surveyed vineyards and it is thought that, in this area, the pathogen may overwinter primarily as mycelia (Killigrew *et al.*, 2005). However, since downy mildew was introduced into this area only in the late 1990s (McKirdy *et al.*, 1999), this absence of oospores may well be caused by a bottleneck situation, due to the presence of only a single genotype, at least at the vineyard level.

Warning systems and models

Goidànich summed up a very large amount of information in a set of tables showing the incubation periods of grape downy mildew at various temperatures and under two (low and high) humidity regimes. Oospore germination (above 10°C), infection conditions (rain of more than 10 mm) and minimal leaf size (oldest leaf 2.5 to 3 cm) needed to intercept the inoculum were added to this model, and the expanded model was used to develop a set of graphs from which the timing of secondary sporulation could be calculated (Goidànich and Casarini, 1961; Goidànich, 1964).

With the availability of computers, it became possible to integrate this information with meteorological data acquired automatically. With the increasing power of computers, a large range of simulation models were developed that corrected and added factors to Goidànich's model. Some of these models calculated the risk of infection purely on the secondary cycle (MILVIT) (Muckensturm *et al.*, 1990; Magnien *et al.*, 1991), some simulated only particular phases of the epidemic cycle (Lalancette *et al.*, 1988; Gherardi *et al.*, 1999) and some the first date of primary infection followed by the secondary cycle with quantitative outputs (Magarey *et al.*, 1991b; Orlandini *et al.*, 1993). Exploratory simulation models were also constructed using the newly available computing power (Blaise *et al.*, 1999; Orlandini *et al.*, 2003; Spera *et al.*, 2009).

There are many of these models and only some of them have been tested independently of their developers (Maurin, 1983; Egger *et al.*, 1994; Serra *et al.*, 1997; Vercesi *et al.*, 1999; Bugaret and Burosse, 2005; Wissotzky, 2007). Even fewer of these models have had any large-scale impact. Those that have had impact are those that have been integrated into more complex advisory systems, such as Vitimeteo *Plasmopara* (which is used in Switzerland and southern Germany) (Viret *et al.*, 2005; Bleyer *et al.*, 2008), or are used to develop grower recommendations that are transmitted through technical bulletins (i.e., Epi and Milvit in France [http://www.champagne-info.net/avert/; http://www.srpv-aquitaine.com]). There are also devices designed to be used by individual growers that measure temperature, humidity and leaf wetness and provide treatment recommendations based on an algorithm not much different from that used in the Goidànich model. This general algorithm is also defined as the 3–10 spray strategy, as the first treatment is prescribed when the average temperature is above 10°C, more than 10 mm of rain have fallen within 24 h and shoot length in the vineyard is at least 10 cm (http://pessl.metos.at). The following treatments will be scheduled at regular intervals, however, considering rainfall (leaf wetness). Despite evident imprecision due to

the strict parameters, this general model can reliably predict the first risk period and recommend thereafter a treatment schedule that will allow growers to prevent development of severe downy mildew in vineyards.

The weakness of this type of model is that the number of recommended pesticide sprays is usually greater than what is needed to avoid an epidemic, particularly at the beginning of the season (Gherardi *et al.*, 1999). Applying a concept based on a tolerance threshold for downy mildew under the particular climatic conditions of southern Switzerland in a vineyard with cv. Merlot trained in Guyot over several years, Jermini and colleagues (Jermini *et al.*, 2006) were able to eliminate half of the recommended treatments. This paradigm change, from a focus on the pathogen and the disease toward a threshold concept, requires detailed knowledge of the host and its relationship with the environment and human activities, such as pruning and thinning. Plant compensation for foliar damage (Jermini *et al.*, 2010b), and the relation to the reserves in the overwintering wood (Jermini *et al.*, 2010a) may allow treatments which protect only the grape bunches in later season. These are highly complex interactions and there is little available data describing them (Giuntoli and Orlandini, 2000; Orlandini *et al.*, 2001). The information that is available is heavily biased by site, year and cultivar factors, and so cannot be readily used for simulation and modeling activities.

Several authors have attempted to develop more sophisticated downy mildew models (Blaise *et al.*, 1996; 1999; Giuntoli and Orlandini, 2000; Bleyer *et al.*, 2003; Kennelly *et al.*, 2005; Rossi *et al.*, 2009). Blaise considered secondary inoculum to be the driving force of grapevine downy mildew epidemics and was able to simulate the progression of the disease quite accurately. However, he (Blaise and Gessler, 1990) needed a quantitative starting point, in order to calibrate the model with the field epidemic, since the primary infections were not quantifiable (Blaise *et al.*, 1997). This model was not further developed. Currently, the most advanced simulation model considers the oospore inoculum quantitatively over the course of the first 3 months after the first primary infection, and models the specific requirements for germination and infection based on weather parameters (Caffi *et al.*, 2009; Rossi *et al.*, 2009; Caffi *et al.*, 2010). The conditions for secondary infection may be met less frequently than those for primary infection, but they can only be fulfilled when the conditions for primary infection have already been met. Therefore, the advisory output of this model automatically covers any secondary infection.

Infections caused by oospores as compared to those caused by secondary sporangia

The visible parts of *P. viticola* are the sporangiophores (Figure 6) and the sporangia. Large numbers of sporangiophores and sporangia are produced on the lower leaf surfaces, usually at the same time that oil spots are visible on the upper leaf surfaces. As conditions for oosporic and secondary-sporangial infections appear to be similar (duration of leaf wetness and temperature), it was assumed that oosporic infections contribute to the formation of the first oil spots and that the production of primary sporangia was not very important once those oil spots were present. The start of the disease was, therefore, defined by the first fulfilled infection conditions, once the conditions for oospore germination had been fulfilled. Thereafter, the further development of the epidemic was attributed solely to the secondary sporangia. This credo was implemented in all control strategies and warning systems.

With the availability of molecular markers that can be used to differentiate between genotypes, first RAPDs and later the more sophisticated microsatellite markers (SSR), it became possible to differentiate clonal oil spots from oil spots derived from different oospores. Each oospore is derived from a recombination event involving two *P. viticola* genotypes. Therefore, polymorphic markers (consisting of different alleles) are combined in different ways. In contrast, the alleles of all markers will remain identical during asexual reproduction.

One hundred and twenty-five years after its introduction into Europe, *P. viticola* shows a high level of genetic variability and we can see clear distinctions between the populations found in different macro-geographic regions (Germany, French/western Switzerland, southern Alps, Greece) (Gobbin *et al.*, 2005; Gobbin *et al.*, 2006). All Italian single-vineyard populations cluster closely, suggesting frequent gene migration between these populations (Gobbin *et al.*, 2003b). In contrast, in

Greece, individual populations are relatively genetically distant from each other. This difference has been attributed to a large winter-surviving population in Italy that is faced with few geographic barriers. In contrast, in Greece, the pathogen is faced with a bottleneck situation, which has reduced its overwintering (and summer) populations drastically, as well as geographic barriers (Rumbou and Gessler, 2006, 2007). Climatic conditions that are unfavourable for *P. viticola* overwintering and/or the production of massive amounts of oospores lead to limited genetic variation, and epidemics are primarily driven by the multiple clonal infections by one or a few genotypes (Rumbou and Gessler, 2004). In most French, Swiss and Italian vineyards, genetic diversity is high during the first month of an epidemic, due to new oospore-derived primary infections (Gobbin *et al.*, 2003b; Gobbin *et al.*, 2005; Loskill *et al.*, 2006; Matasci *et al.*, 2010).

It is puzzling that, in all of the described cases, the clonal propagation rate was not consistent across all of the genotypes in a single vineyard, even relative to the first appearance of a particular genotype. We might have assumed that a genotype appearing early in the season would have a better chance of producing secondary lesions than one appearing later in the season, and that different genotypes appearing after a particular infection event would have similar clonal reproduction rates. In most cases, one or two genotypes eventually dominate (Pertot *et al.*, 2003). A detailed analysis of individual vine stocks confirmed that most primary infections never lead to secondary infections (Matasci *et al.*, 2010). The few that do produce secondary infections spread in steps of 1–2 m and, sometimes, in jumps (Gobbin *et al.*, 2005). Similar results were reported in South Africa. In that study, researchers noted that epidemics were dominated by one or two genotypes that each contributed between 14 and 67% of all lesions present. The other genotypes mostly appeared only once or with a few lesions. Also, the dominant genotypes that spread around a focal point seldom escape and establish a new focal point at a further distance (Koopman *et al.*, 2007).

The question of races and differences in aggressiveness

Grünzel (1960) in Germany, in one of the most detailed studies, tested and observed *P. viticola* populations and single spore lines from central Europe on 60 grape cultivars and species with a large range of susceptibility/resistance. He could not detect any differences between any of the populations nor single spore isolates. Differences were found in size and branching of the sporangiophores, but variability due to temperature had more influence. So Grünzel concluded that no pathological, morphological or physiological differences are present in *P. viticola* that might justify the establishment of morphological varieties or physiologic races, and no differences are present in aggressiveness. However, he warned that fungicide resistant strains may appear or even races with specific virulence may hamper breeding efforts. Differences in morphological characteristics are inadequate to identify possible biological races, biotypes or forms (Rafaila and David, 1963) and variability of physiological traits was due to temperature and humidity rather than to genetic traits in particular experiment and with selected *P. viticola* material and grape cultivars (Rafaila *et al.*, 1968; Voitovich and Nikolaev 1970). The question of physiological races was again raised with the appearance and description of isolates resistant to fungicides (Gay-Bellile *et al.*, 1983). This can be disputed, however, if strains carrying a gene for resistance against a particular fungicide have to be defined as a particular race in an organism which recombines each year. A race overcoming a particular resistance was described by Kast (1998; 2001), and more recently the cultivar Bianca appears to select for a specific "Bianca" race (Peressotti *et al.*, 2010). So, as expected, *P. viticola* can form physiological races (pathotypes) in relation to single gene resistance of the host and single action-site fungicides. The question of variable aggressiveness still remains open as such differences in aggressiveness would explain the observed differences in frequencies of various genotypes in *P. viticola* populations during the season (Matasci *et al.*, 2010).

Resistance of grapevine to downy mildew

Genetic traits for grapevine resistance to downy mildew

Although European *V. vinifera* cultivars are highly susceptible to *P. viticola*, *Muscadinia* species and several American and Asian *Vitis* species exhibit varying levels of resistance to the patho-

gen. Resistant accessions of the wild species *V. riparia*, *V. cinerea*, *V. labrusca*, *V. rupestris*, *V. berlandieri*, *V. lincecumii* and *Muscadinia rotundifolia* exhibit variable levels of resistance (Dai *et al.*, 1995; Denzer *et al.*, 1995; Staudt and Kassemeyer, 1995; Brown *et al.*, 1999; Kortekamp and Zyprian, 2003; Unger *et al.*, 2007, Cadle-Davidson, 2008; Diez-Navajas *et al.*, 2008). All resistant accessions of North American *Vitis* species allow *P. viticola* to complete its life cycle, but, on these plants, sporangia are released at lower rates than on susceptible individuals (Bellin *et al.*, 2009). Some resistant genotypes efficiently halt hyphal growth in mesophyll, and neither visible symptoms nor sporulation are observed on these plants (Diez-Navajas *et al.*, 2008). Some Asian grapes can tolerate hyphal growth on the outer sides of the leaf laminae while preventing stomatal penetration (Jürges *et al.*, 2009).

Vitis vinifera can be crossed with resistant grapevine species and usually generates fertile offspring when crossed with almost all of the wild *Vitis* species of North and Central America and western Asia (Alleweldt and Possingham, 1988; Moreira *et al.*, 2011). Wild species are reliable sources of resistance to many of the diseases and environmental stresses that affect cultivated grapevines (Moreira *et al.*, 2011). Attempts to create grapevine cultivars that carry genes for resistance to downy mildew have shown that the resistance trait is quantitatively inherited (Moreira *et al.*, 2011). Quantitative trait loci (QTLs) for resistance to downy mildew have been identified (Fischer *et al.*, 2004; Welter *et al.*, 2007; Bellin *et al.*, 2009; Marguerit *et al.*, 2009; Moreira *et al.*, 2011) and molecular maps based on populations segregating for disease-resistant traits, including resistance to downy mildew, are available (Grando *et al.*, 2003; Di Gaspero *et al.*, 2007; Marguerit *et al.*, 2009). Specifically, QTLs with major downy mildew-resistance effects have been identified in linkage groups (LGs) 9 and 12 in *V. riparia* (Marguerit *et al.*, 2009), LG 14 in *V. amurensis* (Blasi *et al.*, 2011) and in LGs 4 and 18 in the resistant grapevine 'Regent' (Fischer *et al.*, 2004; Welter *et al.*, 2007). A major QTL was also identified on LG 7, together with additional QTLs on LGs 8, 12 and 17 in a segregating population of *V. vinifera* × *V. viparia* (Moreira *et al.*, 2011). Other QTLs were identified on LGs 1, 6 and 7 in a cross between two interspecific hybrids inheriting *V. rotundifoglia* and *V. amurensis* traits (Moreira *et al.*, 2011). QTLs with major effects on downy mildew resistance were identified through comparative genetic mapping (Merdinoglu *et al.*, 2003; Fischer *et al.*, 2004; Welter *et al.*, 2007; Bellin *et al.*, 2009; Marguerit *et al.*, 2009) and were named "*Resistance to P. viticola* (*Rpv*)". *Rpv1* and *Rpv2* are located on chromosome (chr) 12 and chr 18, respectively, and were found to be responsible for the resistance derived from *M. rotundifolia* (Merdinoglu *et al.*, 2003; Peressotti *et al.*, 2010), as a QTL identified in the same region in *V. riparia* (Marguerit *et al.*, 2009). The *Rpv3* locus is found on chr 18, corresponding to QTL peaks for downy mildew resistance in the grapevine 'Bianca', and was found to be responsible for the onset of a hypersensitive response (HR) (Bellin *et al.*, 2009). The resistance durability in 'Bianca' and the emergence of a new isolate of *P. viticola* that has overcome the monogenic resistance of this cultivar demonstrate the importance of stacking different resistance genes in individual resistant varieties (Peressotti *et al.*, 2010).

Efforts to introgress resistant traits into cultivated *V. vinifera* genotypes using conventional breeding techniques (Eibach *et al.*, 2007) have yielded some resistant interspecific hybrids, but further work is needed to couple strong resistance with high quality wine production traits (Burger *et al.*, 2009). Studies of the *V. vinifera* genome have revealed that resistance genes and other genes involved in defence processes tend to be located on chr 5, 7, 9, 12, 13, 18 and 19, in genomic regions associated with *P. viticola* resistance in wild grapevines (Di Gaspero and Cipriani, 2003; Di Gaspero *et al.*, 2007; Velasco *et al.*, 2007; Moroldo *et al.*, 2008). The availability of grapevine genome information (Jaillon *et al.*, 2007; Velasco *et al.*, 2007) has created the opportunity for new breeding efforts. Regions carrying resistance genes can be moved in clusters across the wild and cultivated genomes, through the use of appropriate molecular markers. However, for these findings to have any impact on genome-assisted breeding and viticultural production systems, researchers will first need to identify the genes or genomic regions responsible for important agronomic traits (Martìnez-Zapater *et al.*, 2009). This would allow the breeding of new cultivars with berry compositions that are similar (or better) than those of existing

cultivars, as well as the optimal disease-resistance traits for local environmental and growing conditions.

Mechanisms of grapevine resistance

Mechanisms responsible for resistance to downy mildew have been investigated in wild grapevines over a half century ago, and classes in the level of resistance/susceptibility have been described for several species of the *Vitaceae* (Boubals 1959). While no clear relationship between grapevine leaf characteristics and susceptibility to downy mildew has been observed (Boso *et al.*, 2010), constitutive resistance to *P. viticola* has been linked to high constitutive levels of antimicrobial compounds, such as inositol and caffeic acid, in uninfected leaves of resistant species (Figueiredo *et al.*, 2008). Constitutive resistance is also suggested by the high levels of expression of some genes related to stress and defence in resistant grapevines, as compared to the lower expression levels observed in susceptible uninoculated grapevines (Kortekamp and Zyprian, 2003; Kortekamp, 2006; Fung *et al.*, 2007; Figueiredo *et al.*, 2008). However, comparative gene expression analyses was unable to identify any broad functional category related to defence in which transcript abundance was overall prevalent in the resistant *V. aestivalis* (Fung *et al.*, 2007). Indeed, basal expression levels of defence-related genes did not explain the different infection outcomes observed in the resistant *V. riparia* and the susceptible *V. vinifera*, suggesting the presence of post-infection resistance processes (Polesani *et al.*, 2010).

Microscopic observations have revealed that the first steps in the infection process are essentially the same in susceptible and resistant grapevines (Kortekamp and Zyprian, 2003; Unger *et al.*, 2007; Diez-Navajas *et al.*, 2008; Polesani *et al.*, 2010). This indicates that the restriction of disease development observed in the resistant plants is based on post-infection mechanisms (Polesani *et al.*, 2010). The resistance to *P. viticola* exhibited by American grapevines can be first observed after the first haustoria have established contact with the membranes of mesophyll cells (Diez-Navajas *et al.*, 2008a; Jürges *et al.*, 2009), with the timing and magnitude of infection varying with the host's genotypic features (Bellin *et al.*, 2009). A gene-for-gene mechanism has been proposed for the downy mildew-recognition process observed in the grapevine 'Bianca', since the *Rpv3* resistance locus mapped to a cluster of NBS-LRR receptor genes (Bellin *et al.*, 2009), and the HR-based resistance of this cultivar was overcome by a *P. viticola* isolate which probably carried mutations in its avirulence (*Avr*) gene (Peressotti *et al.*, 2010a).

Mechanisms of post-infection resistance in resistant grapevines include callose deposition in stomata (Gindro *et al.*, 2003), cell wall-associated defence processes (Diez-Navajas *et al.*, 2008; Jürges *et al.*, 2009), the accumulation of reactive oxygen species and increased peroxidase activity (Kortekamp and Zyprian, 2003) and HR activation (Kortekamp and Zyprian, 2003; Kortekamp, 2006; Diez-Navajas *et al.*, 2008; Bellin *et al.*, 2009) in reaction to inoculation with *P. viticola*. Localised necrosis was the earliest phenotypic difference noted between susceptible and resistant individuals, and this was associated with significant reductions in pathogen performance and in symptom development (Bellin *et al.*, 2009).

The results of whole genome gene expression analyses support the view that downy mildew resistance in *V. riparia* (Polesani *et al.*, 2010) and *V. amurensis* (Wu *et al.*, 2010) is based on a post-infection mechanism. Resistant grapevines react to *P. viticola* inoculation by rapidly up-regulating genes coding for pathogenesis-related (PR) proteins (Busam *et al.*, 1997; Kortekamp, 2006) and genes involved in defence-related signal transduction and metabolic processes, including the phenylpropanoid pathway (Kortekamp, 2006; Polesani *et al.*, 2010; Wu *et al.*, 2010). The production of stilbenic phytoalexin is one of the most important responses to fungal infection in grapevine (Jeandet *et al.*, 2002; Schmidlin *et al.*, 2008; Alonso-Villaverde *et al.*, 2011), and accumulation of different antimicrobial compounds, such as resveratrol and viniferins, have been observed following the inoculation of resistant grapevines with this pathogen (Dai *et al.*, 1995; Dercks and Creasy, 1989a; Pezet *et al.*, 2004; Jean-Denis *et al.*, 2006). Microscopic observations of the responses of various *Vitis* species to inoculation with *P. viticola* revealed the presence of three different patterns: "(i) inhibition of pathogen development early after attachment of zoospores; (ii) successful colonization of the mesophyll by the pathogen; and (iii) aberrant development, where the pathogen does not attach to

guard cells, but produces hyphae on the leaf surface without formation of viable sporangiophores" (Jürges *et al*., 2009).

As expected, *V. vinifera* shows Pattern (ii); whereas Pattern (i) is typical of the North American host species and some Siberian species known to be resistant. Other Asian species display Pattern (iii). Evolutionarily differentiated signals passed between the host and the pathogen may regulate these three types of interactions. Detailed knowledge of these signals would open new possibilities for disease control.

Induced resistance to *Plasmopara viticola* in grapevine

Although *V. vinifera* is susceptible to *P. viticola*, it can defend itself against other pathogens, indicating that defence mechanisms are present, but not activated in response to this pathogen (Polesani *et al*., 2010). Gene expression analyses have revealed that susceptible grapevines react to *P. viticola* inoculation by activating the expression of defence-related genes (Busam *et al*., 1997; Hamiduzzaman *et al*., 2005; Kortekamp, 2006; Mzid *et al*., 2007; Chong *et al*., 2008; Trouvelot *et al*., 2008a), but this reaction is not sufficient to prevent or limit the infection. Transcriptional changes associated with early stages of *P. viticola* infection indicate the presence of a weak defence response in susceptible grapevines (Polesani *et al*., 2010). Mechanisms associated with compatibility and disease development are established later, and grapevine transcripts from all major functional categories, including defence processes, were strongly down-regulated at the oil-spot stage (Polesani *et al*., 2008). Moreover, key genes of carbohydrate transport and partitioning were found to be highly induced 10 days after plants were inoculated with *P. viticola*, suggesting pathogen-dependent alteration of leaf carbohydrate metabolism (Hayes *et al*., 2010).

In susceptible grapevines, compatible interaction is probably achieved through a lack of recognition. The failure of susceptible plants to mount an effective defence response is likely due to the fact that their resistance gene alleles do not develop into any *P. viticola*-specific recognition system, since *V. vinifera* does not evolve in the presence of *P. viticola* (Di Gaspero *et al*., 2007). However, applications of substances known to induce resistance increased resistance of susceptible grapevines and significantly reduce downy mildew symptoms. In particular, treatments with resistance inducers such as chitosan (Aziz *et al*., 2006), laminarin (Aziz *et al*., 2003), sulfated laminarin (Trouvelot *et al*., 2008a; Allègre *et al*., 2009), oligogalacturonide (Allègre *et al*., 2009), β-aminobutyric acid (BABA) (Reuveni *et al*., 2001; Hamiduzzaman *et al*., 2005; Dubreuil-Maurizi *et al*., 2010), acibenzolar-S-methyl (BTH) (Perazzolli *et al*., 2008), fosetyl-Al (Dercks and Creasy, 1989b), plant extracts (Godard *et al*., 2009) have been shown to increase resistance. Moreover, plant extracts (Godard *et al*., 2009), aqueous extract of *Penicillium chrysogenum* (Thuerig *et al*., 2006), organic amendments (Thuerig *et al*., 2010), extracts of *Solidago canadensis, Penicillium crysogenum*, *Aureobasidium pullulans* (Harm *et al*., 2011), and the microorganism *Trichoderma harzianum* strain T39 (Perazzolli *et al*., 2008) were all able to increase grapevine resistance to downy mildew.

Mechanisms of resistance to different pathogens and pests (Conrath *et al*., 2006) can be activated in different plant species by eliciting the systemic acquired resistance (SAR) or induced systemic resistance (ISR) pathways (Pieterse *et al*., 2009). Whereas SAR is typically based on direct activation of defence systems (van Hulten *et al*., 2006), ISR usually involves the activation of a priming state for enhanced reaction upon exposure to biotic or abiotic stresses (Conrath *et al*., 2006). In the ISR system, primed plant defences are activated only when they are really needed (Verhagen *et al*., 2004) and this priming provides advantages in terms of energy costs for the plant (van Hulten *et al*., 2006). Priming probably evolved to allow the plant to conserve energy under pathogen-free conditions (Walters and Heil, 2007) and has been identified as a promising low-impact, low-cost strategy for use in modern disease management (Beckers and Conrath, 2007).

Mechanisms of induced resistance against downy mildew of grapevine have been shown to involve stomatal closure (Allègre *et al*., 2009), the expression of defence genes (Hamiduzzaman *et al*., 2005; Trouvelot *et al*., 2008b; Harm *et al*., 2011; Perazzolli *et al*., 2011), increased enzymatic activity (Godard *et al*., 2009; Harm et al., 2011), callose deposits (Hamiduzzaman *et al*., 2005) and the accumulation of phytoalexins (Dercks and Creasy,

1989b; Slaughter *et al*., 2008; Ferri et al., 2009; Godard *et al*., 2009). The priming mechanisms that are involved in resistance to *P. viticola* were described following treatment with BABA (Dubreuil-Maurizi *et al*., 2010; Hamiduzzaman *et al*., 2005), sulfated laminarin (Trouvelot *et al*., 2008a), plant extracts (Godard *et al*., 2009) and fosetyl-Al (Dercks and Creasy, 1989b). Defence reactions were not activated in primed grapevines in the absence of pathogens, but greater accumulation of defence gene products and callose (Hamiduzzaman *et al*., 2005; Trouvelot *et al*., 2008a), reactive oxygen species (Dubreuil-Maurizi *et al*., 2010; (Trouvelot *et al*., 2008a; Dubreuil-Maurizi *et al*., 2010;) and phenolic compounds (Dercks and Creasy, 1989b; Trouvelot *et al*., 2008a; Godard *et al*., 2009) were observed in elicited plants after they were inoculated with *P. viticola*, as compared to uninoculated plants.

Additionally, a dual effect was reported for some resistance inducers. Application of *Rheum palmatum* root extract induced the intense production of phytoalexins in the absence of infection and reinforced the synthesis of these defence compounds following inoculation with *P. viticola* (Godard *et al*., 2009). Treatments of *T. harzianum* T39 caused a direct modulation of defence-related genes and the activation of priming for enhanced expression of these genes after pathogen inoculation (Perazzolli *et al*., 2011). Moreover, the *T. harzianum* T39-induced resistance did not entail apparent energy costs for defence activation under controlled conditions, in contrast to the negative effects of the BTH-activated resistance on grapevine growth (Perazzolli *et al*., 2011). In order to obtain more detailed characterization of the molecular processes involved in resistance to downy mildew that are inducible in grapevine, it will be necessary to identify the cellular pathways and key genes involved in plant self-protection. Since induced resistance is considered to be an enhancement of basal resistance and is affected by plant genotype (Tucci *et al*., 2010), the characterization of self-protection in different cultivars seems to be particularly important. Likewise, more knowledge concerning the influence of physiological state and abiotic stress exposure will be important for the more widespread use of grapevine resistance activation, which, at the moment, is far from being applied under commercial field conditions. Results suggesting a site-factor effect on the susceptibility of 'Chasselas' to *P. viticola* infection, which is correlated with altered expression levels of defence-related genes (Thuerig *et al*., 2010), are quite puzzling. The authors of that study were not able to identify a specific cause for that effect, such as management or induced resistance by organic compost amendments.

Breeding and deploying disease-resistant cultivars

Breeding for grapevine resistance to downy mildew was initiated early in the 19th century. Even when variability in susceptibility toward *P. viticola* was present in the spectrum of the available *V. vinifera* cultivars, breeders focused on hybridization of the *V. vinifera* with selections from American species with high levels of downy mildew resistance, or with the American hybrids that displayed good field disease resistance (Pee-Laby, 1926).

The first American hybrid was the 'Alexander' grape, which was discovered around 1740 near a vineyard in Virginia. It was the product of accidental cross-pollination between a local grapevine and imported vines, which rarely survived more than a year, but in some cases did survive long enough to reproduce with the native grapevines. One typical American grape is the 'Catawba' (with a pronounced musky or "foxy" flavour), which was introduced to wine-growers in the early 1800s by Major John Adlum of Georgetown (Washington, D.C.). Grown predominantly on the East Coast of the United States, the cultivar 'Concord' was selected in 1849 by Ephraim Wales Bull from seeds of the species *Vitis labrusca* (also known as "fox grape"). Some doubts exist as to whether the male parent of this cultivar might not be 'Catawba', which although usually classified as *Vitis labrusca*, may have some *Vitis vinifera* in its background. Other popular early American hybrids included 'Clinton' (*Vitis labrusca* × *Vitis riparia*) and 'Isabella'. 'Isabella' was of *Vitis labrusca* parentage and almost certainly originated from a cross with an unknown *Vitis vinifera*, whose discovery was attributed to a Mrs. Isabella Gibbs of South Carolina in 1816. Large quantities of 'Isabella' vines were imported into Europe in the early 1800s and it is assumed that phylloxera was introduced unknowingly into Europe on the roots of 'Isabella', as 'Isabella' is resistant to phylloxera.

Once the French discovered the possibility of grafting onto phylloxera-resistant rootstocks, they were once again able to grow their centuries-old, traditional varieties. The French government enacted laws (15 January 1935) forbidding commercialization and further planting of hybrids, so that as hybrid vines aged and required re-planting and *V. vinifera* vines were planted in their stead. Advances in the chemical control of downy mildew reduced the demand for cultivars with resistance to *P. viticola*. Resistance breeding was no longer a priority in Italy, France or Spain. Eventually, downy mildew-resistant cultivars of this "first generation" (e.g., 'Marechal Foch', 'Leon Millot'[1] and 'Seyval Blanc'[2]) mostly disappeared from commercial vineyards. Very few vineyards in France still grow hybrid grape varieties, with the notable exception of many vineyards in the Loire valley, where 'Chambourcin' is still a popular variety, and in Armagnac, where 'Baco Blanc' is grown. 'Chambourcin' is a curious hybrid grape variety. Originating in the Loire region of France, this hybrid of American and European vines was developed in the 1880's by Joannes Seyve[3], in an effort to find vines resistant to the phylloxera louse. It has been commercially available only since the 1960's, and is not yet fully accepted in the EU, where wine from this variety cannot be sold under the label 'quality wine' (but only as table wine or *vin ordinaire*). In France, this hybrid currently covers 800 ha (http://www.onivins.fr/EspacePro/Economie/StatistiquesIndex.asp). It is now grown in Australia and is favoured by producers in more humid areas, such as the coastal areas of New South Wales, because it is quite resistant to attacks by different fungi.

The breeding of resistant cultivars became a priority once again in new wine regions or regions in which traditional pesticide applications were intensive and often questioned since the late 1980s. Alternative production systems based on either integrated production or more strict organic production systems had trouble avoiding irregular, but heavy losses. As demand developed for plant resistance, breeders were sometimes able to anticipate the growers' needs (Blattner and Schaller, 1990; Topfer and Eibach, 2002). 'Chambourcin' is one of the parents of a new disease-resistant variety, 'Regent', whose popularity is increasing among German grape growers. 'Regent' is a dark-skinned early ripening, interspecific hybrid variety used for making wine. It has both European (*V. vinifera*) and American vine species in its pedigree and possesses broad resistance to the most significant fungal diseases affecting grapevine, including downy mildew. 'Regent' was developed in 1967 by Gerhardt Alleweldt at the Geilweilerhof Institute for Grape Breeding, Germany by crossing 'Diana' (a Silvaner × Müller-Thurgau cross and thus a *V. vinifera* variety) with the interspecific hybrid 'Chambourcin'. Experimental plantings followed in 1985, and it received varietal protection in 1994 and was first released for cultivation in Germany in 1996 (Topfer and Eibach, 2002). It is currently among the most important new, fungal-resistant quality grape varieties in the world and especially in German wine-producing regions. In 2006, the area planted to this cultivar in Germany was 2,183 ha (5,390 acres) and increasing. This made 'Regent' Germany's 12th most popular variety and its most popular hybrid grape variety. (http://en.wikipedia.org/wiki/Regent_%28grape%29) 'Regent' is also grown in the United Kingdom with some success. However, the durability of its resistance has been questioned. Based on his work with various *P. viticola* isolates, during which an isolate was found that infected and sporulated on these plants, Kast concluded "that resistance of the hybrid variety 'Regent' might be dependent on the biological fitness and the genotype of the *P. viticola* isolate" (Kast, 2001).

The resistance of 'Regent' is due to several different QTLs (one in LG 9 and another in LG 10) (Fischer *et al.*, 2004; Welter *et al.*, 2007) and a third of the main resistance effect is associated with a QTL in LG 18 (Zyprian *et al.*, 2009). 'Bianca', a white grape bred in Hungary, possesses promising wine quality characteristics and excellent *P.*

---

[1] Bred by Eugene Kuhlmann (1858–1932), a breeder from Alsace

[2] Bred in 1919 by Bertille Seyve, Saint-Vallier, Rhone Valley, France (1864–1939)

[3] Albert Seibel was a French physician who was born in the Ardeche in 1844. He founded a school to teach grafting techniques in 1895 and his company produced over 16,000 new hybrids, including nearly 500 varieties that were then grown commercially. His grapes are known as Seibel grapes, the most famous of which are 'Aurore', 'Chancellor', 'Chelois Chambourcin' and 'Vidal Blanc'.

viticola resistance (Blattner and Schaller, 1990; Kozma, 1998). Unfortunately, its resistance is due to a single gene (Bellin *et al.*, 2009) and, in various test vineyards, it already appears to be susceptible to this downy mildew (Peressotti *et al.*, 2010). Several white wine cultivars with good resistance derived from American grapes and backcrossed several times to eliminate any foxy taste are currently being grown by organic growers in northern Europe. In addition, a long list of new disease-resistant cultivars is being tested in northern European wine-producing regions (Ovsepyan, 1983; Spring *et al.*, 1998; Bouquet *et al.*, 2000; Spring, 2001; 2003; Siegfried and Temperli, 2008; Burger *et al.*, 2009). As durability of resistance is a crucial factor for any new cultivar, breeders will have to consider the genetic structure of any resistance that they develop. This will be increasingly possible thanks to genetic linkage maps (Di Gaspero *et al.*, 2007), the availability and annotation of the *V. vinifera* genome sequence (Doddapaneni *et al.*, 2008; Moreira *et al.*, 2011; Moroldo *et al.*, 2008; Velasco *et al.*, 2007) and the identification of QTLs and single genes associated with disease resistance (Di Gaspero *et al.*, 2009) (see the section on Genetic traits for grapevine resistance to downy mildew, above).

The history of downy mildew control

The use of copper compounds

Although there are probably many other unreported observations from the same period, we refer to Millardet, who, in 1882, discovered of the effect of copper against downy mildew. To deter passersby from eating from grapevines close to the road, those vines were sprayed with a mixture of copper sulfate and lime that was both visible and bad-tasting. Millardet noticed that the treated plants remained free of downy mildew symptoms while the rest of the vineyard suffered from the disease. Based on this observation, he began the first studies of the effect of this treatment, the results of which were published in 1885. He recommended this treatment (later called Bordeaux mixture) for the protection of grapevines against downy mildew (Millardet, 1885). Initially, Millardet proposed the use of larger quantities (e.g. higher concentrations) (8:15:100 copper sulfate pentahydrate: hydrated lime: water). Recommended quantities were significantly reduced in later years for economic reasons. The first studies focused mainly on the concentration of copper in the mixture, paying relatively little attention to the volume applied per unit of surface area.

The qualities of Bordeaux mixture received almost immediate worldwide recognition, particularly its strong adhesion to the leaves, its long persistence in the vineyard and its colour, which allowed the applicator to easily verify the distribution of the treatment. Almost immediately after the publication of Millardet's work, many researchers began their own experiments with Bordeaux mixture. In France and Austria at the beginning of the 20th century, two to five treatments with a blue, alkaline Bordeaux mixture containing 2.5–3% copper were considered sufficient, even considering the fact that efficacy varied with latitude. In Switzerland, after a very severe mildew epidemic, five to seven applications (at 1.5% for the first two applications and at 2 to 3.5% for the remainder) were prescribed (Godet, 1923; Hengl, 1923). The use of Bordeaux mixture soon spread beyond Europe, to Australia and the USA (Lyon, 1924).

Bordeaux mixture is based on copper sulfate and was usually applied at a minimum concentration of 1.5%, most often at 2–3% and up to 4% copper sulfate when downy mildew is severe. Initially, growers would prepare homemade Bordeaux mixture by separately dissolving the copper sulfate ($CuSO_4$) and lime [$Ca(OH)_2$] in water and then mixing the two solutions together (Figure 11). The conventional method of describing the mixture's composition is to list the weight of the copper sulfate, the weight of the hydrated lime and the volume of water, in that order. The concentration of the mixture is expressed in terms of the ratio of the weight of the copper sulfate to the weight of the water, meaning that a 1% Bordeaux mixture has the formula 1:1:100. The copper content of a 1% Bordeaux mixture is 0.25%, since cop-

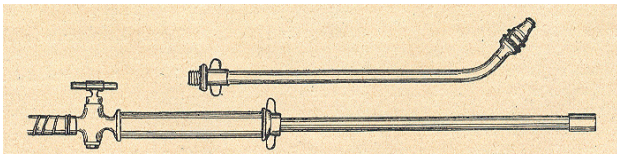

Figure 11. Devices to produce the Bordeaux mixture (drawing from Müller, 1918).

per sulfate contains 25% copper. Over time, proprietary brands of ready-to-use Bordeaux mixture appeared on the market, and nowadays almost nobody prepares the mixture themselves.

Initially, another product called Burgundy mixture was frequently used. Burgundy mixture is a mixture of copper sulfate and sodium carbonate, which was invented in 1887 by Masson (Masson, 1887). It is similar to the Bordeaux mixture in that it contains copper sulfate ($CuSO_4$), but it contains sodium carbonate (Na_2CO_3) instead of lime. The production of the two mixtures is also similar. Burgundy mixture is made by mixing dissolved copper sulfate with dissolved sodium carbonate. Over time, the sodium carbonate will crystallize out of solution and the closer the copper sulfate to sodium carbonate ratio is to 1:1, the faster this process will occur. The fact that Burgundy mixture must be mixed quickly and used almost immediately caused its more discontinuous use. The more difficult preparation and the higher costs associated with this product led to growers abandoning it in favor of the Bordeaux mixture.

In general, acidic Bordeaux mixtures have proved no more effective than neutral or alkaline mixtures. Moreover, acidic mixtures are unsuitable for use in damp regions, where they can cause scorching. For applications to tender foliage, pH-neutral mixtures are recommended. Excessively alkaline mixtures should be avoided as they are liable to cause russeting of the leaves, as a result of the damage they cause to the protective leaf cuticle (Faes and Staehelin, 1930). In warm regions, the recommendation was to make Burgundy mixture very alkaline by adding an excess amount of sodium carbonate (Ravaz, 1927).

Around the same time that other proprietary copper products were being developed, Kurtakol (copper compound in colloidal form manufactured by the firm of Dr Kurt Albert in Biebrich, Germany) debuted on the market. This product was considered to be very easily prepared and did not clog spraying equipment or burn foliage (Lustner, 1922). The copper content of this mixture was about equal to that of Bordeaux mixture, but, in practice, Kurtakol was cheaper due to the smaller quantities required and the absence of lime. Nosperal 1781 and 1782 (Dyeworks; Höchstam-Main, Germany) were other products that contained copper sulfate. They were somewhat cheaper than Bordeaux mixture, on account of the smaller quantities required, namely 1%, as compared with products containing 1.5 and 2% copper sulfate. These mixtures were made by dissolving the commercial powders in water and then adding lime to the solution the next day, in order to neutralize it. Nosperal 1781 was slightly superior to 1782, chiefly in its physiological effects. Foliage treated with this product remained exceptionally luxuriant in color and its development was not negatively affected (Lustner, 1922).

Perocid, which was also prepared by Dr. Albert's firm provided satisfactory disease control (Kramer, 1927). This product consisted of cerium sulfate, the oxides of cerium, neodymium, and lanthanum and small quantities of the oxides of thorium, calcium, and iron, mixed with 900 g of lime in 100 L of water.

Nosperit was a liquid preparation based on copper, which provided very satisfactory control of downy mildew in Germany (Kramer, 1927). Preparations based on mixtures of copper and arsenic, such as Nosprasen, were also developed, in order to control downy mildew and insects at the same time. Both Nosperit and Nosprasen were shown to be just as effective as Bordeaux mixture (Kramer, 1927). The advantages of a colloidal preparation over Bordeaux mixture are mainly related to the savings associated with the use of smaller quantities of such preparations as Perosan and Kurtakol), and the greater fluidity, homogeneity of distribution and adhesiveness provided by the physical structure of these preparations (Guyot, 1927).

A preparation named Kupferpasta-Bosna, manufactured by the Bosnian Electricity Company in Vienna, Austria, was also used and gave very good results after two applications at 1% copper (Lustner, 1922). Other copper based products included Caffaro paste, Protector Ramato and Bouisol (Jacquemain and Gravier, 1932).

Eau Céleste was first applied in 1886 to control downy mildew in France. It was based on the addition of ammonium sulfate to neutralized copper sulfate. The major problem with its use was it caused scorching of vine foliage (Armet, 1934).

The appealing possibility of combining treatments against downy mildew with those against powdery mildew, and the hypothesis that a powder can be more evenly distributed across plant surfaces than a liquid, led to the development of

copper dust. In general, spray treatments with copper always yielded better results that dusting, even though copper dusts protected the grape clusters well. The control provided by the dusts was in general less than satisfactory, since dusts are easily washed off by rain and, therefore, need to be re-applied after each rain event. Sulfur and copper sulfate dustings were specifically recommended for use on fruit clusters, especially to supplement liquid treatments that might fail to reach these fruit clusters. Other recommendations included one regular dust application between each two liquid treatments (Bachala, 1927), or three applications of Bordeaux mixture, followed by three dustings with sulfur-copper sulfate dust (Salomon, 1927). The dust product that provided the best control was made by mixing one part copper sulfate talc powder with two parts sublimed sulfur. This mixture would be used for the first two dustings, followed by an application of a mixture composed of equal parts of copper sulfate talc powder and sulfur (Bachala, 1927). Several dust products were soon placed on the market (De Gibon, 1929).

To summarize, copper compounds can classified as follows: *i*) copper solutions obtained by dissolving a highly soluble copper salt (sulfate or acetate) in water; *ii*) copper mixtures, which are precipitates obtained after the neutralization of a copper salt (e.g. Bordeaux mixture, Burgundy mixture); *iii*) aqueous suspensions of a copper salt that are not very soluble (e.g. copper oxide, basic sulfate, oxychloride, carbonate); and *iv*) dusts.

Experiments were carried out to compare the anti-downy mildew activities of various metals and salts. The activities of metallic elements were examined first and their effects were compared with those of copper oxide and copper carbonate. Magnesium, zinc, aluminium, bismuyh, tungsten and iron were found to be ineffective; mercury, antimony nickel, cadmium, and lead exhibited an intermediate level of activity and gallium and arsenic had the strongest effects. Nitrate, sulfate and chloride salts of a number of metals were also tested, but most of these were not effective. Nickel sulfate and mercuric chloride provided control comparable to that provided by copper sulfate. The next most effective were uranyl nitrate and cadmium sulfate, the latter of which caused damage to grapevines. Zinc sulfate came in next in terms of fungicidal efficacy and its activity was almost equivalent to that of aluminium sulfate and cerium sulfate. Cobalt was not nearly as effective as nickel. A great variety of copper compounds were also examined, but none of them provided better pathogen control than copper sulfate (Kotte, 1924).

In an effort to find an alternative to copper, the fungicidal activities of a number of chemical dyes were tested, including malachite green, brilliant green, rhodainine B, safranin, acridine yellow, auramine and yellow pyocfcanine. Quinosol and sunoxol of the oxyquinoline group of organic substances were also tested (Meyer, 1932b). None of these substances was ever used in practice.

Increased knowledge of the biological cycle of *P. viticola* opened up the possibility of applying the most effective chemical dosage at the most suitable time. An important set of studies were devoted to optimizing the application of copper-based products. Several research stations started from the 1920s to issue instructions for preventive treatment of grapevines as soon as conditions were favourable for the germination of the winter oospores of *P. viticola* and the first infection of the vines, followed by instructions to growers regarding the appropriate treatment for each phase of the attack in accordance with reported rainfall and other meteorological conditions (Voglino, 1922). The relationship between meteorological conditions and downy mildew outbreaks began to receive more attention and was used to identify critical periods for the application of preventive sprays and determine the specific compounds and dosages to be used. In the 1920's, several studies tried to summarize the rules to be used in planning treatments applied against this disease. It was commonly accepted that the efficacy of a treatment did not depend on the use of highly concentrated product, but rather on the thoroughness and timeliness of its application (Cerasoli, 1924). In general, the use of Bordeaux mixtures, including those containing low rates of copper (1–1.5%), were suggested for periods during which the vines were considered to have a low level of susceptibility and there was only a low risk of disease-favoring conditions. Higher rates (3–4%) were recommended for the most risky part of the season. A rate between 1.5 and 2% was considered to be fairly standard (Stiegler, 1923; Kramer, 1925; Faes and Staehelin, 1927; Schellenberg, 1927).

The belief that Bordeaux mixture was the best

protective product and should be applied at the right time further increased scientists' efforts to optimize its application. The necessity of anticipating the active periods of the pathogen by correlating its life cycle with meteorological conditions, so as to be able to apply the treatment at the most appropriate time, was emphasized for several years (Cadoret, 1927). For example, spraying the vines with a Bordeaux mixture containing 1 to 2% copper sulfate as soon as infection was detected and again before flowering (with a 2% copper sulfate mixture) was recommended (Schellenberg, 1927).

Although some researchers recommended the use of low concentrations of copper, in numerous experiments and commercial vineyards the application of proprietary brands of Bordeaux and Burgundy mixtures containing low concentrations of copper sulfate (0.5 to 1%) failed to prevent heavy outbreaks of the disease, resulting in almost total yield losses (Wille, 1927). Therefore, the use of a mixture containing 2% copper sulfate was recommended for wet and cold seasons and generally for susceptible varieties growing in damp soils. In exceptionally humid seasons, it was recommended that vines that had already been attacked be treated with a final application of a 3% Bordeaux mixture. The use of mixtures containing only 1% copper sulfate should be restricted to resistant varieties growing under dry and warm conditions.

Most of the failures experienced by vine-growers in the control of downy mildew have been related to the timing of pesticide applications. According to Musiani (1930), the best time for spraying the vines with copper products is when the characteristic oil spots appear on the leaves. However, during prolonged rainy spells, this criterion was considered invalid and, in those situations, he recommended to spray at the first break in the rain and supplement with applications of cupric sulfur dusts (Musiani, 1930). Moreover, growers were advised to spray their vines after each heavy rain, because when treated parts have been washed by heavy rain, the remaining deposits can only protect the exact areas they cover (Villedieu, 1932).

In some regions, legislative measures were taken to ensure obligatory and effective control of the parasites of the vine, in order to avoid significant losses. For example, in Switzerland, laws regulating the control of downy mildew were enacted in all 25 cantons (Faes, 1929).

Another approach to increase fungicide efficacy is to reduce the application rate focusing on the spray apparatus. With the earliest equipment, the type of apparatus was less important than how it was handled. In general, in the first studies researchers focused more on the concentration of copper than on the spray volumes since the vegetation was covered until runoff. In fact, very fine sprays, which deposit small, evenly spaced droplets on the leaves were less effective than heavier sprays that cover the leaves with a continuous film of the liquid. A minimum of spray material per unit area (e.g. 1,000 L ha^{-1}) was recommended as, below this level, the efficacy of the application rapidly decreases (Neyrac, 1930). Later, considerable progress was made in the improvement of spraying equipment, for example, the use of compressed air to improve spray application (Mahoux, 1933) (Figure 12).

Attention was also paid to agronomic factors. The level of downy mildew control in a sprayed crop can be increased by cutting off the apical portions of vine shoots during the crop's vegetative stages (Ravaz, 1930). Thinning the foliage throughout the vegetative period was considered necessary to ensure that the grape bunches would be easily reached by any fungicidal sprays and

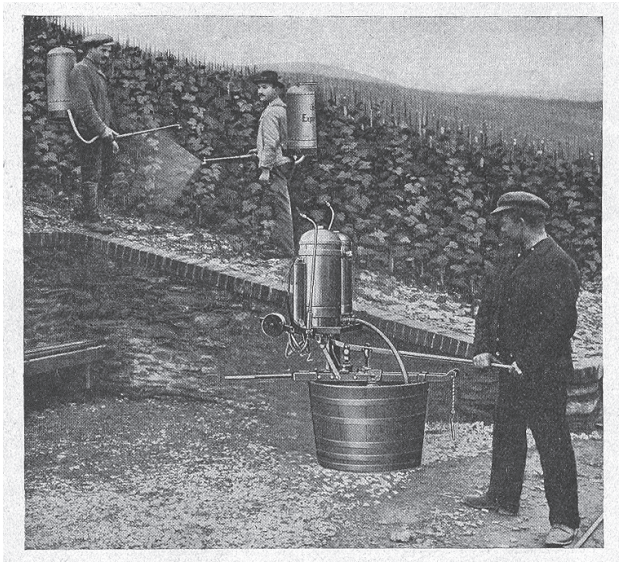

Figure 12. Application of Bordeaux mixture using pressure filling device and sprayer type Express, Ludwigshafen Germany (foto from Müller, 1918).

dusts that were applied. The disease was least severe in vineyards with weaker growth that were orientated from north to south, to ensure the good aeration of the plants (Cadoret, 1931). Suppression of the primary foci of infection in damp areas, which are quite favourable for the germination of oospores, were achieved by removing the buds and the leaves at the base of low stocks and grafts before any signs of the disease appear (Serviere, 1940).

Regarding the relations between phenological factors and the incidence of downy mildew, it was shown that the greatest amount of infection occured during the period of intensive growth (end of May to the middle of July) and that the development of the pathogen was favored by warm weather and frequent rain. At that time, based on an old viticultural rule and despite the fact that it was already clear that the first infection may occur when the largest leaves are only 10 cm in width, the practice was to start spraying when the shoots were 15 to 20 cm long. The combination of suitable temperatures and the continuous presence of moisture from persistent rain was known to provide the appropriate night-time conditions for infection. In 1930, Müller reported that incubation calendars and recommendations of experimental stations based on them were being used to guide the timing of preventive sprays (Müller, 1930).

A copper salt of oxyquinoline was shown to have very promising effects (Meyer, 1932a) against downy mildew. Researchers were also interested in copper-ammonia mixtures, because the ammonia helps to solubilize the copper, and the efficacy levels of these mixtures were mainly determined by their dissolved copper content (Chevalier, 1934; Bosc, 1935).

The copper sulfate-sodium bicarbonate spray mixture was prepared by adding powdered sodium bicarbonate to a copper sulfate solution. Carbon dioxide is freely generated in this mixture and this property was used to increase the pressure in the spraying apparatus. The apparatus consisted essentially of a hermetically sealed body, in which the two substances were mixed after it was closed. Field tests indicated that this mixture was as effective as Bordeaux mixture (Comte, 1935).

Laboratory studies of chemical treatments containing reduced quantities of copper that were prepared with bentonite and copper salts showed that, in these products, active copper was easily liberated to exert its toxic action against spores. Furthermore, activity of copper was naturally enhanced by the colloidal nature of the bentonitic clay (Malquori and Boezini, 1941).

Good results, in comparison with Bordeaux mixture, were also obtained with copper oxide (Osterwalder, 1939) and copper oxychloride (Gallay *et al.*, 1945). In some cases, copper oxide did not protect against downy mildew as well as Bordeaux mixture. However, copper oxide was considered preferable to Bordeaux mixture at the early stages of crop growth, since it does not scorch the plants and consequently permits the early development of luxuriant foliage (Blumer and Peyek, 1947).

In conclusion, Bordeaux mixture was always more effective than the less tenacious copper oxychlorides and copper oxides tested (Gallay, 1948). However, other forms of copper were found to be less phytotoxic and cause less injury to young shoots. Provided applications begin early and are made at frequent intervals, growers may choose to use Bordeaux mixture, copper oxide or oxychlorides during the early period of crop development. After flowering, Bordeaux mixture is preferable to other copper mixtures. If an oxide or an oxychloride is used, applications should be made very frequently. Bordeaux mixture should be used for the last two treatments to avoid premature leaf yellowing (Gallay and Staehelin, 1949). The frequency of application (usually every 10 days) should be increased when spots are present on the leaves and conditions favor infection (Gallay *et al.*, 1951).

Incubation calendars and methods of forecasting outbreaks of grapevine downy mildew were further developed to facilitate the prompt application of control measures and potentially reduce the number of treatments needed (Müller, 1937). Comparisons and criticisms of these tools have focused on the fact that they were reliable only in regions with relatively dry climates. In fact, the rapid succession of secondary infections in areas subject to heavy precipitation seemed to preclude accurate forecasts of the critical dates for spraying (Osterwalder, 1940). Over the years, several warning systems have been developed to optimize and reduce the use of copper and other fungicides for downy mildew control (Zillig and Niemeyer, 1942; Baldacci, 1949; Darpoux, 1949; Baldacci and Refatti, 1956; Viret *et al.*, 2005).

Limited availability of copper during World War II

During World War II (1939–1945), the availability of copper for agriculture was substantially reduced. In 1941, the annual European consumption of copper for agricultural purposes, including the control of downy mildew, was limited to 125,000 tons. In 1942 in Switzerland, the demand for copper to be used as fungicide was estimated to amount to 1,550 tons of metal but, because of the war, only 690 tons were available for agricultural use, of which only 320 tons were allocated for viticulture (Hadorn, 1942). Based on the demand for copper to be used as fungicide, experiments to find copper substitutes were carried out in connection with economy campaigns. Several products were tested without successful results (Mestbes, 1942).

The inability to find alternatives that provided control comparable to that of Bordeaux mixture fueled the use of lower rates of copper. Throughout German Switzerland, the effective control of grapevine downy mildew with dilute Bordeaux mixture proved perfectly feasible in the summer of 1941, during which the weather was not conducive to the disease. The correct timing of the treatments was found to be more important than the concentration of the Bordeaux mixture used (Peyer, 1942). Similar conclusions were drawn in Germany (Zillig and Niemeyer, 1942). In Switzerland in 1941, a series of experiments examined the possibility of combining effective protection of vines from downy mildew with the minimal use of copper dictated by the military situation. The best results were obtained with 1.5 or 2% Bordeaux mixture supplemented with an adhesive (Faes, 1943). Further tests concluded that copper cannot be replaced by zinc, aluminum, magnesium sulfates or other metallic salts (e.g., silver, cadmium, chrome, mercury and iron). In 1943, Hadorn stated that it was essential to use existing stocks of copper sulfate with the utmost economy consistent with adequate disease control (Hadorn, 1943). After several years of research, the conclusion was that there was very little likelihood of discovering any exceptionally active copper salts (Raucourt, 1943).

The age of acupric fungicides

After greenhouse tests confirmed the efficacy of zinc dimethyldithiocarbamate (ziram; introduced by du Pont de Nemours) against *P. viticola* (Morel, 1946), several dithiocarbamates were tested. In general, in the earliest trials, Bordeaux mixture often gave better results than dithiocarbamates or captan (introduced by Chevron Chemical Co.) (Pieri, 1952; Gaudineau and Messiaen, 1953; Boubals and Vergnes, 1954; Graniti, 1954; Boubals *et al.*, 1956; Lupetti Belviglieri, 1956). In later trials, captan often performed better than Bordeaux mixture at standard dosages and zineb (ENT 14 874, Dithane Z-78; introduced by Rohom & Haas Co.) provided almost the same level of control as Bordeaux mixture (Boubals and Vergnes, 1953; Gaudineau and Messiaen, 1953). The subsequent success of acupric fungicides was probably related to the development of new stable products, their reduced costs on the market (Zobrist, 1954) and the almost complete absence of any phytotoxicity, which was very often a problem when copper compounds were used (i.e., severe late-summer leaf scorch and punctures accompanied, in the case of oxychlorides, by chlorosis proceeding from the margins inward, and russeting of the grapes) (Kundert, 1956). Knowledge of the precise conditions under which these new products were likely to give the best control (Baldacci *et al.*, 1962) (i.e., treating according the incubation calendar or when copper tends to have phytotoxic effects and avoiding late applications that they may affect fermentation) (Cordonnier, 1955) and their consistent efficacy (Boubals *et al.*, 1955) further increased their popularity among growers. Other acupric products acting on contact, which have been found to successfully control downy mildew include: methiram (FMC 9102; Polyram, introduced by BASF); maneb (ENT 14 875, Dithane M-22, introduced by Rohm & Haas Co.); mancozeb (Dithane M-45, introduced by Rohm & Haas Co.); propineb (LH 30/Z, Antracol, introduced by Bayer); captafol (Ortho-5865, introduced by Chevron Chemical), folpet (introduced by Chevron Chemical) and dichlofluanid (KUE 13032c, Euparen, introduced by Bayer) (Cucchi *et al.*, 1974).

With time, the drawbacks of certain acupric fungicides became apparent: debated human toxicity and the promotion of mite populations. For example, it was shown that copper-maneb and copper-maneb-zineb mixtures enhanced damage caused by the mite *Eriophyes vitis*. Copper-mancozeb was initially considered to be less problematic (Chaboussou *et al.*, 1974), but opinions later changed, presumably because of the acaricidal ac-

tion of mancozeb mixture, which also affects beneficial predatory mites.

In the 1970's, copper compounds received little attention from researchers. From this period, the development of a trellis system with copper nets is the only major new achievement with copper-containing materials (Olivelli, 1976).

Cytotropic/systemic fungicides

In the 1980s, the discovery of cytotropic systemic fungicides with activity against *P. viticola* which can last for 9–18 days, which are not washed off by rain once adsorbed and which can cure established infections, was greeted with great enthusiasm (Boubals and Lafon, 1981). These systemic fungicides work best when treated plants are growing quickly and new, susceptible organs are being formed (i.e., formation of bunches, beginning of bloom and bloom). Sporulation of *P. viticola* is usually inhibited when lesions are sprayed with these products and the sporangia that do form following treatment have reduced capacity for germination (Wicks and Lee, 1982).

Cymoxanil (DPX-3217, Curzate, introduced by du Pont de Nemours) provided good control of *P. viticola* in field trials with no phytotoxicity, even at high application rates, with no adverse effects on must fermentation or wine flavor (Serres and Carraro, 1976). When applied in a mixture with mancozeb, it allowed a 50% reduction in the amount of mancozeb needed to control downy mildew, and treatment with this mixture at 10-day intervals provided satisfactory protection (Piglionica *et al.*, 1977). When applied during the incubation period, cymoxanil was found to have a curative effect (Klopping and Delp, 1980). When this product was applied at fixed times or in relation to the pathogen cycle, very good control was obtained with good economic savings compared to traditional chemical treatments (Rizzotto and Sartor, 1981). Thanks to its mechanism of action (Samoucha and Gisi, 1987b), growers began to apply cymoxanil as a curative treatment, with the idea of reducing the total number of treatments by applying it only when infection had already occurred (Perandin *et al.*, 1985).

Acylalanine metalaxyl (CGA 48988, Ridomil, introduced by Ciba-Geigy) was found to be very active against *P. viticola* thanks to its unique combination of residual and systemic properties. In field trials, it was more effective than products based on dithiocarbamates or folpet, and curative treatments were effective up to the 5th day of incubation, delaying the further development of symptoms by about 12 days and considerably reducing sporulation. Applications made when spots first appeared or later had partial eradicant effects, which were observed after a few days (Lafon *et al.*, 1978). The ascending systemic distribution and high level of activity of the chemical within plants allow new shoots to remain protected for 2–4 weeks. Since this molecule is rapidly absorbed by the green parts of the plants, its effect is independent of weather conditions (Urech and Schwinn, 1978). Acylalanine metalaxyl has been shown to provide good control of downy mildew in several environments (Vial *et al.*, 1978; Boubals *et al.*, 1979; Murolo and Stanich, 1980; Wicks, 1980; Cesari *et al.*, 1981; Marais and Tromp, 1981).

Similarly, aluminum ethylphosphite or fosetyl-Al (LS74 783, Aliette, introduced by Rhône-Poulenc) has also been shown to provide good protection against downy mildew (Boubals *et al.*, 1979; Raynal *et al.*, 1980). This chemical protects plant organs formed after treatment thanks to its systemic activity (Chazalet *et al.*, 1977), outstanding curative effects (Lafon *et al.*, 1977) and rapid penetration of plant tissues (Mur, 1978). Even if experiments have not provided any evidence that a phytoalexin response is the primary mode of action of fosetyl-Al, it has been suggested that the combination of direct and indirect modes of action exerts a double selection pressure on *P. viticola*. This may also explain why fosetyl-Al-tolerant strains are rarely found in the field (Dercks and Creasy, 1989b).

Phenylamide oxadixyl (SAN 371F, Sandofan, introduced by Sandoz) is another systemic fungicide effective against downy mildew. It exhibits preventative, curative and eradicative activity (Gisi *et al.*, 1983). It was also found to have a synergistic effect against *P. viticola* when applied together with other fungicides (Grabski and Gisi, 1987).

Dimethomorph (CME 151, Forum, introduced by Cyanamid) is a derivative of cinnamic acid. Dimethomorph is systemic and can penetrate grapevine leaves following foliar application. It has a long period of residual activity, is a good protectant and curative and is an excellent antisporulant.

Its novel mode of action has not been associated with any cross-resistance with phenylamide fungicides. Under field conditions, it is very effective, especially when applied in mixtures with contact fungicides, and does not cause any phytotoxicity (Albert *et al.*, 1988; Wicks and Hall, 1990; Reuveni, 1997). Other products that have been tested, but never became as popular as those listed above are chlorothalonil (Northover and Ripley, 1980) and the benzamide zrilamid (ICIA0001) (Heaney *et al.*, 1988).

The strobilurin fungicides (derived from a natural substance secreted by wood mushrooms *Oudemansiella mucida*) were initially successfully tested against *P. viticola* (Godet *et al.*, 1997), with the aim of controlling grapevine downy mildew and powdery mildew at the same time (Reuveni, 2001). With the rapid appearance of resistant strains of *P. viticola*, the developers of these fungicides shifted their attention toward powdery mildew and black rot. The preventive and curative effects of azoxystrobin (ICI A 5504, Strobi, introduced by Syngenta) (Bugaret *et al.*, 1998) and trifloxystrobin (CGA 279202, Flint, introduced by Bayer) (Margot *et al.*, 1998; Reuveni, 2001) against *P. viticola* should be mentioned.

Special mention should be made of phosphorous acid and phosphonates (phosphorous acid neutralized mainly with potassium hydroxide), which cannot be considered true fungicides since they do not directly kill the pathogen and are commercialized as fertilizers. Nevertheless, these chemicals provide good protection against *P. viticola* (Magarey *et al.*, 1990; Magarey *et al.*, 1991a;). Phosphonate has both preventive effects as well as significant post-infection activity. When applied up to 13 days after infection, it has also been shown to reduce sporulation (Wicks *et al.*, 1991).

Waking up from a dream: resistant *P. viticola* strains and strategies for managing resistance

The discovery of systemic curative fungicides raised the possibility of treating crops only after infections had occurred, similar to the approach used to treat human infections. This possibility was greeted with great enthusiasm. Unfortunately, pesticide-resistant pathogen strains soon appeared around the world. One of the first reports of *P. viticola* resistance to metalaxyl, ofurace and milfuram dates to 1982 (Gay-Bellile *et al.*, 1983). This discovery led to efforts to develop new strategies for controlling *P. viticola* (Boubals and Mur, 1983).

Strains of *P. viticola* that are resistant to anilide fungicides have also been reported (Clerjeau *et al.*, 1984). Fungicide mixtures provide better control of phenylamide-resistant strains than any of the individual chemicals applied alone (Samoucha and Gisi, 1987a). In 1993, control failures were reported on grapevine in northern Italy under severe downy mildew pressure after post-infection applications of cymoxanil. The results of several trials suggested that *P. viticola* resistance to cymoxanil was the most probable explanation for this disease-control failure (Gullino *et al.*, 1997).

The strobilurin fungicides and the non-strobilurins, famoxadone and fenamidone, form a cross-resistance group referred to as QoI-STAR. *Plasmopara viticola* strains that are resistant to QoI fungicides (inhibitors of mitochondrial respiration at the Qo site of cytochrome b in the mitochondrial bc1 enzyme complex) have also been discovered (Wong and Wilcox, 2000). In one study, QoI resistance was not observed in vineyards that had never been treated with QoI fungicides and a decrease in resistance was generally observed in vineyards in which QoI treatments had been discontinued (Toffolatti *et al.*, 2007). Evolution of resistance to QoIs seems to depend mainly on early mitotic events. The selection process for resistant mutants in populations exposed to QoI treatments may involve mechanisms similar to those described for cases of resistance to other classes of fungicides that are controlled by single nuclear genes (Gisi *et al.*, 2002). The choice of the other fungicide in a spray mixture and the rate at which it is applied can significantly affect the success of strategies for managing QoI resistance (Genet *et al.*, 2006).

Several classes of fungicides with differing systemic properties, specificities, durations of activity and resistance risks are currently available. The major site-specific fungicides are the quinone outside inhibitors (QoIs; e.g., azoxystrobin), phenylamides (e.g., mefenoxam), carboxylic acid amides (CAAs; e.g., dimethomorph, mandipropamid) and cyano-acetamide oximes (cymoxanil). QoIs inhibit mitochondrial respiration and phenylamides inhibit the polymerization of rRNA; whereas the modes of action of the other two site-specific classes are unknown (Gisi and Sierotzki, 2008). Current

strategies to avoid the establishment of resistance involve the use of systemic products together with protective multi-site fungicides, alternation with products that have different mechanisms of action or the absence of cross-resistance, and restricting the number of applications (usually no more than two or three per year, generally applied during bloom and periods of frequent rain and rapid plant growth). Multi-site fungicides used in mixtures or in alternation with site-specific fungicides are, for example, mancozeb, folpet, chlorothalonil and copper formulations.

The newest molecules and current strategies

Current strategies for the control of grapevine downy mildew are mainly based on the use of preventive copper or mancozeb treatments from the beginning of the period during which plants are susceptible to infection. These are applied in the spring in environments in which primary infections are usually heavy, or after the end of the incubation period of the first probable primary infection in areas in which oospore infections are not usually problematic. These products are preferred when other pathogens as *Phomopsis viticola* are present. Systemic fungicides should be applied from before bloom to mid-summer, which is the period of the crop's fastest growth and highest level of susceptibility. Later in the season, preventive contact fungicides are generally preferred (depending on weather conditions). Copper is still widely used during the last part of the season because it tends to improve the lignification of grapevine shoots.

With the revision of plant protection products undertaken in the EU (Council Directive No 414/91), many older products have been removed from the market. However, new active ingredients that are highly effective against downy mildew and have favourable toxicological profiles have been developed and are now available to growers (Egger, 2008). Some of these chemicals are described below.

Iprovalicarb (SZX 722, Melody, introduced by Bayer) inhibits the growth of the germ tubes of zoospores and sporangia, mycelial growth and the sporulation of oomycetes, with consequent strong protective, curative and eradicative effects. It has systemic properties and is translocated in plants via the transpiration stream. No cross-resistance has been observed between this product and metalaxyl or cymoxanil. This suggests that this product has a different mode of action (Stenzel *et al.*, 1998), which would make it particularly useful in anti-resistance strategies (Dutzmann, 1999). Nowadays in viticulture, it is used in combination with various products. The proportion of active ingredient found in leaves increases over a period of days and the uptake is greatly dependent on temperature and the moisture level of the leaves (Stubler *et al.*, 1999).

Benthiavalicarb-isopropyl (KIF 230, introduced by Kumiai Chemical Industry) is also active against oomycetes. Although it has strong prophylactic and local activity and some translaminar activity, unlike iprovalicard, benthiavalicarb is not translocated from leaf to leaf. It can remain active on leaves for up to 28 days. Applied in combination with products that have different mechanisms of action, it may be useful in delaying the spread of resistance (Reuveni, 2003).

Famoxadone (DPX-JE874, Famoxate, introduced by du Pont de Nemours) is a highly active, broad-spectrum oxazolidinedione fungicide. Famoxadone inhibits sporangial differentiation and the release of zoospores and causes the lysis of zoospores within minutes. It has a strong preventative effect and good residual activity (at least 7–10 days), but no curative effect (Andrieu *et al.*, 2001). After application, famoxadone is found on the leaf surfaces or in association with epicuticular waxes, which probably explains its good rainfastness. Despite its low level of solubility in water, famoxadone spray residues on grape leaves are reactivated in surface water sufficiently quickly to prevent infections (Andrieu *et al.*, 2000). Famoxadone has also been combined with fosetyl-Al in the new commercial product Equation System (DPX-MP560) (Bassi *et al.*, 2000).

Fenamidone (RPA407213, Fenomen, introduced by Rhône-Poulenc) is an imidazolinone that inhibits mitochondrial respiration by blocking electron transport at the ubihydroquinone: cytochrome c oxidoreductase enzyme. It has both protective and curative activity against *P. viticola* on grapevine under a range of experimental conditions (Latorse *et al.*, 1998; Mercer *et al.*, 1998). The sprays of fenamidone should be limited in downy mildew control programmes that include products with a similar mode of action. It should be applied

in alternation (Lacombe *et al.*, 2001) or in mixture with other products with dissimilar modes of action (Gamberini *et al.*, 2000).

The benzamide Zoxamide (RH-7281, Electis, introduced by Rohm & Haas Co.) specifically targets oomycetes. It exhibits strong preventive activity and has excellent rainfastness and residual properties. Its mode of action involves binding tubulin and inhibiting nuclear division. It acts after spore germination to inhibit fungal penetration (Ruggiero and Regiroli, 2000). Because of its unique mode of action, it has good potential as a new alternative for use against fungal diseases of grapevine (Duriatti *et al.*, 2003).

Cyazofamid (IKF-916, Mildicut, introduced by ISK Biosciences Corporation) belongs to the cyanoimidazole family. Its activity against a broad spectrum of oomycetes was first reported in 1998. Under field conditions in several countries, this chemical exhibited excellent control of downy mildew (Mitani *et al.*, 1998). It has both preventive and curative activity. Its mechanism of action is based on the inhibition of the mitochondrial complex III. No cross-resistance with other QoI inhibitors was observed (Gentili *et al.*, 2002).

The benzamide Fluopicolide (AE C638206, introduced by Bayer), which is commercially combined with fosetyl-Al, is another systemic fungicide. To discourage the development of resistance, a maximum of three applications of fluopicolide are recommended per season (Gouot, 2006). Fluopicolide is active at several stages of the oomycete life cycle. It does not markedly affect zoospore release or cyst germination, but does inhibit zoospore mobility at very low concentrations. A significant reduction in sporulation has also been demonstrated following application, as well as the prevention of the liberation of viable motile zoospores from sporangia (Latorse *et al.*, 2006).

Mandipropamid (Pergado, introduced by Syngenta Crop Protection) is a mandelic acid amide (Lamberth *et al.*, 2006). Mandipropamid inhibits spore germination, mycelial growth and sporulation. It is recommended as a preventive treatment, but also provides curative activity during the incubation period. Because cross-resistance with the valinamide fungicides iprovalicarb and benthiavalicarb and the cinnamic acid amide fungicides dimethomorph and flumorph has been postulated, all five of these compounds are classified as carboxylic acid amide (CAA) fungicides (Gisi *et al.*, 2007). Novel analogues of mandipropamid have been designed and prepared and are expected on the market shortly (Lamberth *et al.*, 2007).

Alternative approaches to downy mildew control

Biological control and natural substances

The use of microorganisms for biocontrol of plant disease is, in theory, a powerful alternative to the use of chemical pesticides. Many microorganisms have been selected over the last decades for their antagonistic activities against *P. viticola* (Arnone *et al.*, 2008), but none of these have been successfully used as biofungicides. Some examples are outlined here.

Trichothecium plasmoparae (initially identified as *T. roseum*) is a hyperparasite on sporulating *P. viticola*, where it cause pink mildew patches. In spite of its aggressiveness, it was never further developed for use as a biocontrol agent (Arpai *et al.*, 1957). *Erwinia herbicola* was found to strongly inhibit the germination of *P. viticola* sporangia under laboratory conditions and its use led to fewer stomata being surrounded by zoospores in liquid cultures, but no further studies involving this microorganism have been reported (Tilcher *et al.*, 1994).

Post-infection applications of suspensions of microconidia of *Fusarium proliferatum* G6 reduced the production of *P. viticola* sporangia on grape leaf discs. Under field conditions, weekly applications of *F. proliferatum* G6 microconidia reduced disease development on leaves and fruit clusters. One of the major factors limiting the use of this microorganism as a biocontrol agent is the fact that it develops at higher temperatures than usually present at the time of application in the field. Therefore, a cold-tolerant strain of the same mycoparasite *F. proliferatum* (designated 1505) was obtained by UV mutagenesis of the G6 strain on grape leaves. This strain showed sufficient growth at 13°C, which is generally suboptimal for growth of *Fusarium* spp., but desirable for *P. viticola* (Bakshi *et al.*, 2001). The fumonisin mycotoxins that are also produced by the G6 strain probably hampered the commercial development of this microorganism as a biofungicide, even though they were not found in the berries or the

juice made from berries sprayed with microconidia in field trials (Falk *et al.*, 1996).

A strain of *Alternaria alternata* that inhibits *P. viticola* was isolated, along with 125 other microorganisms, from grapevine leaves that showed anomalous downy mildew symptoms (collected in Tuscany, Italy). Cytological observations highlighted the fact that, even without close contact with *A. alternata*, *P. viticola* mycelia exposed to this fungus showed severe ultrastructural alterations, such as the presence of enlarged vacuoles or vacuoles containing electron-dense precipitates. Their haustoria appeared necrotic and irregularly shaped or were coated with callose-like substances. In light of these findings, researchers hypothesized that *A. alternata* might be toxic to *P. viticola* (Musetti *et al.*, 2006). The main low-molecular-weight metabolites produced by the endophyte were three diketopiperazines that very effectively limited the sporulation of *P. viticola*. No necrotic lesions or other signs of phytotoxicity were observed on DKP-treated grapevine leaf tissues (Musetti *et al.*, 2007).

In a recent study (Thuerig *et al.*, 2006), an aqueous extract of the dry mycelium of *Penicillium chrysogenum* (Pen) provided grapevine downy mildew control comparable to that provided by copper. The extract had no major direct fungicidal effect and it was supposed that it protected plants by activating their defence mechanisms.

The endophytic fungus *Acremonium byssoides* was first isolated from the leaves of grapevine cv. Regina Bianca and later found in 34 grapevine varieties over a period of two years (Burruano *et al.*, 2008). This fungus can be considered to be a natural colonizer of grapevines and actively parasitizes *P. viticola*. Culture filtrates and a crude extract of *A. byssoides* completely inhibited the pathogen (Burruano *et al.*, 2008).

Trichoderma harzianum T39 was initially developed as commercial fungicide with the trade name of Trichodex (Makhteshim Agan, Chemical Works LTD, Be'er Sheva, Israel). This strain protects susceptible grapevine cultivars without directly inhibiting the germination of sporangia, through a plant-mediated resistance mechanism (see above, Induced resistance to *Plasmopara viticola* in grapevine). The systemic resistance is homogeneously activated, independent of leaf position (Perazzolli *et al.*, 2008).

In conclusion, despite good activity of microorganisms against *P. viticola* demonstrated under controlled conditions, microbial biocontrol agents have never shown good and consistent activity against *P. viticola* in the field. In the case of the polycyclic *P. viticola*, low-level control provided by a biocontrol agent can lead to significant losses due to the disease. Moreover, after it penetrates plants, the pathogen can no longer be controlled by antagonists after infection (Pertot and Gessler, 2007). The microorganisms tested so far have all been found to be active only for short periods after application, and those that induced resistance were not capable of completely controlling the disease. We conclude that a disease management programme based on the use of microorganisms or alternative products will need to target *P. viticola* at multiple sites and multiple stages of its life cycle to allow growers to successfully and reliably avoid crop damage (Vecchione *et al.*, 2007).

The effects of products of natural origin were recently the subject of extensive tests (Dagostin *et al.*, 2011). Some other examples of tests of natural products can be mentioned as well. In one study, 58 plant extracts were tested. Of these, only extracts of *Chloris virgata*, *Dalbergia hupeana*, *Pinus massoniana*, *Paeonia suffruticosa* and *Robinia pseudoacacia* inhibited the germination of sporangia and effectively controlled the disease on plants (Chen *et al.*, 2002). In another study, an extract of sage (*Salvia officinalis*) was shown to control grapevine downy mildew under field conditions (Dagostin *et al.*, 2010). An oily paste extract of *Inula viscosa* leaves has also been shown to effectively control downy mildew under field conditions (Cohen *et al.*, 2006). However, its high phytotoxicity on grapevine prevents its wide use as a fungicide. In general, even if plant extracts are highly effective and possess good toxicological profiles, their use is limited by their high costs, limited availability in large quantities, low persistence and low-level of rainfastness (Dagostin *et al.*, 2010).

The effects of an extract of the brown alga *Ascophyllum nodosum* on grapevine interactions with *P. viticola* were examined. Downy mildew outbreaks were greatly reduced when extracts of this alga were sprayed on grape leaves (Lizzi *et al.*, 1998). The β-1,3-glucan laminarin derived from the brown alga *Laminaria digitata* was shown to be an efficient elicitor of defence responses in

grapevine cells and plants and also effectively reduced *P. viticola* damages. Defence reactions elicited by laminarin in grapevine cells include calcium influx, alkalinization of the extracellular medium, an oxidative burst, activation of two mitogen-activated protein kinases, expression of 10 defence-related genes with different kinetics and intensities, increases in chitinase and β-1,3-glucanase activities and the production of two phytoalexins, resveratrol and epsilon-viniferin (Aziz *et al.*, 2003). Similarly, sulfated laminarin (PS3) has been shown to elicit plant defence activities and protect grapevine against downy mildew (Trouvelot *et al.*, 2008b).

Chitosan, a deacetylated derivative of chitin, was also shown to promote plant defence reactions and significantly reduce infection severity on grapevine leaves. Its oligomers trigger the accumulation of phytoalexins, trans- and cis-resveratrol and their derivatives, epsilon-viniferin and piceid in grapevine leaves (Aziz *et al.*, 2006).

The non-protein amino acid BABA (DL-3-amino-*n*-butanoic acid, beta-aminobutyric acid) has recently attracted the interest of many researchers. It has been reported to induce local and systemic resistance against downy mildew in grapevine leaves. BABA was able to stop fungal colonization even when applied to leaf discs after infection. The resistance of BABA persisted for more than 14 days. BABA has been shown to provide systemic protection (Cohen *et al.*, 1999). Treatment of a susceptible grapevine cultivar with BABA prior to inoculation with *P. viticola* primed the accumulation of specific phytoalexins that are undetectable in non-BABA-primed plants, increasing resistance (Slaughter *et al.*, 2008).

It is still unclear whether the inducers of grapevine resistance to *P. viticola* can represent an alternative to copper for downy mildew control, but inducers are likely to assist in reducing the susceptibility of plants to infection, improving the results obtained with other weak natural control agents.

Downy mildew control in organic viticulture

In 1991, the first European regulation (EEC No. 2092/91) concerning organic production of agricultural products was issued. This regulation introduced several limitations for the production of organic grapes. It stated that, in organic agricultural systems, diseases must be controlled using a combination of the following measures: choice of appropriate species and varieties, appropriate crop rotation programmes and mechanical cultivation techniques. It is obvious that such measures cannot sufficiently control grapevine downy mildew, especially when conditions are favourable for the disease. In cases of immediate threat to the crop, the law tolerates the use of some specific products, which are listed in its Annex II. In its list of fungicides, Annex II includes lecithin, plant oils, mineral oils, potassium permanganate, calcium hydroxide and microorganisms (bacteria, viruses and fungi). Sulfur and copper, the latter in the form of copper hydroxide, copper oxychloride, (tribasic) copper sulfate and cuprous oxide, were also included because they are of natural origin, but mainly because they are considered to be part of traditional organic farming practices.

It was soon realized that the use of copper may have long-term consequences, due to its accumulation in the soil, which would be incompatible with organic farming's objective of environmentally friendly farming. Therefore, a new Commission Regulation (EC) No 473/2002 amended Annex II, specified the conditions under which copper may be used and introduced limits on its use, expressed in terms of kilograms of copper that can be applied per hectare per year. The initial limit was a maximum of 8 kg copper per hectare per year. But, from 1 January 2006, that limit was reduced to 6 kg copper per ha per year. For perennial crops, growers are allowed to use a five-year average of 30 kg per hectare.

The new Council Regulation (EC) No. 834/2007 on organic production and labeling of organic products, which replaced Regulation 2092/91, basically confirmed the previous law's position on plant protection, stating that the prevention of damage caused by diseases shall rely primarily on protection provided by natural enemies, the choice of species and varieties, crop rotation, cultivation techniques and thermal processes. It stated that even plant protection products that have been authorized for use in organic production [Annex II of Commission Regulation (EC) No. 889/2008] may only be used in cases of an established threat to a crop. The previous limitations on the use of copper were maintained and the sole novelty was the inclusion of copper octanoate in the list of permitted

copper compounds. This strict limitation of the use of copper has encouraged research into optimized schedules and formulations, to help further reduce the quantities of copper applied while ensuring an acceptable level of disease control.

Among web-based decision support systems specifically designed to help organic growers optimize and track their use of copper, we can mention Coptimizer, a model-driven decision-support system. The ability of Coptimizer to produce recommendations that will provide good control without exceeding the copper limit was evaluated in simulations using historical data and tested in two experiments carried out in 2008 under field conditions. The results of these experiments showed that, using Coptimizer, growers could maintain the same level of protection they enjoyed when a traditional application schedule was followed while applying only half the amount of copper (Kuflik *et al.*, 2009). This system is based on a warning model and a decision-making procedure based on treating vines with variable rates of copper whenever there is an immediate risk of infection and the vegetation has not been sufficiently protected by previous treatments (Pellegrini *et al.*, 2010).

With the aim of evaluating most of the products available for use in organic viticulture, a wide study was carried out in Italy and Switzerland between 2004 and 2007, under controlled greenhouse conditions and in vineyards. A total of 112 different treatments, including biocontrol agents, materials of animal origin, homeopathic preparations, inorganic materials, microbial extracts, natural derivatives, plant extracts, physical methods and synthetic materials were tested, and none of them resulted to be a good substitute of copper in terms of disease control efficacy (Dagostin *et al.*, 2011). This information will help researchers avoid unnecessary tests of ineffective products that have already been examined.

The discussion concerning the inclusion of the phosponates in the Annex II list is ongoing. One of the major concerns is that applications of potassium phosphonate inevitably lead to phosphonate residues in the wine, which are not compatible with the reputation of organic wine among consumers. Phosponates, especially potassium phosphonate, are highly effective against downy mildew and allow growers to avoid most of the problems associated with the use of copper (phytotoxicity and accumulation in the soil). Phosphonate residues in wine are highly correlated with the total amount of potassium phosphonate applied during the crop's vegetative period, but not with the date of the last application. Potassium phosphonate is, therefore, an effective fungicide for the control of downy mildew (Speiser *et al.*, 1999).

Application technology

In the first 30 years of the *P. viticola* presence in Europe, many substances were tested, but only copper, in various forms, applied directly to the parts of the plant to be protected provided sufficient control of downy mildew. Experimental applications of copper to senescent leaves to reduce the oospore load were not successful, nor were soil applications (Müller-Thurgau, 1922). The customary application was aimed at producing a fine and regular film of drops on leaf surfaces, so that the zoospores liberated from the landing "conidia" would be killed. It was also assumed that the copper would penetrate the leaves, strengthening epidermis cells and retarding pathogen penetration.

The recognition that *P. viticola* can only penetrate leaves through stomata, which are almost all found on the lower surfaces of the leaves, and young berries, led to a need for an in-depth analysis of whether spraying the upper leaf surfaces contributed to disease control (Müller-Thurgau, 1912). The conclusion of that analysis, which was also confirmed in a scientific discussion included in this paper (see description and prediction of the life cycle: Sites of infection), was that lower surfaces must be treated. It was also suggested that sprayers be constructed in such a way that the leaves could be treated from below with high pressure, and include nozzles that create fine droplets (Figure 13).

Application technology evolved slowly. An important step forward came with the development of air-blast sprayers in the mid-1940's. Some of these air-blast sprayers produced droplets small enough to create mists. The liquid carrier of the fungicide was reduced from more than 2000 L ha^{-1}, and the custom of treatment until run-off was replaced with the use of spray volumes of less than 1600 L ha^{-1} and, later, ultra-low volumes of 75 to 150 L ha^{-1}. The use of these air blowers raised concerns about drift and the loss of a high percentage of the product to non-target surfaces, such as

soil, neighboring plots and sensitive areas, such as nearby bodies of water. Air speed and volume, droplet size, spray volume and driving speed all have to be considered in efforts to maximize the efficiency of these applications.

The techniques currently employed are: (a) traditional mechanical spraying at low or medium pressure (2 to 8 or 10 kg) at 600 to 1200 L ha^{-1} or high pressure (15 to 50 kg) at 400 to 800 L ha^{-1}; (b) atomization by means of pneumatic sprayers (air blowers) at 200 to 500 L ha^{-1}, which requires a large amount of motive force; (c) the Pintagram formula, which requires less than 100 L ha^{-1} of a highly concentrated spray solution and less motive force; (d) aerosols; and (e) foam mist sprayers. Manufacturers of application equipment have developed a range of models with their own specific advantages. But, a large proportion of the pesticide material will still inevitably be deposited outside of the target area (overview in Giles *et al.*, 2008). Developers of application machinery are currently taking full advantage of advances in computerization and sensorics, with target detection systems, as for example based on chlorophyll fluorescence (Cséfalvay *et al.*, 2009), that can be used for precise applications that take full account of the health of the crop being treated. However, most European growers still work with relatively simple air blowers that are mechanically adjusted. These growers can minimize off-target applications by correctly calibrating their sprayers based on the developmental stage of the crop being treated (Siegrfried *et al.*, 2007; Viret *et al.*, 2008).

An interesting alternative to the popular air-blower systems is the stationary spraying system, which was first tested at the Swiss Federal Station for Fruit-Growing, Viticulture, and Horticulture at Wädenswil (Jenny, 1937; Jenny and Huber, 1940), and later also south of the Alps (Jenny, 1941). Following these early trials, this type of

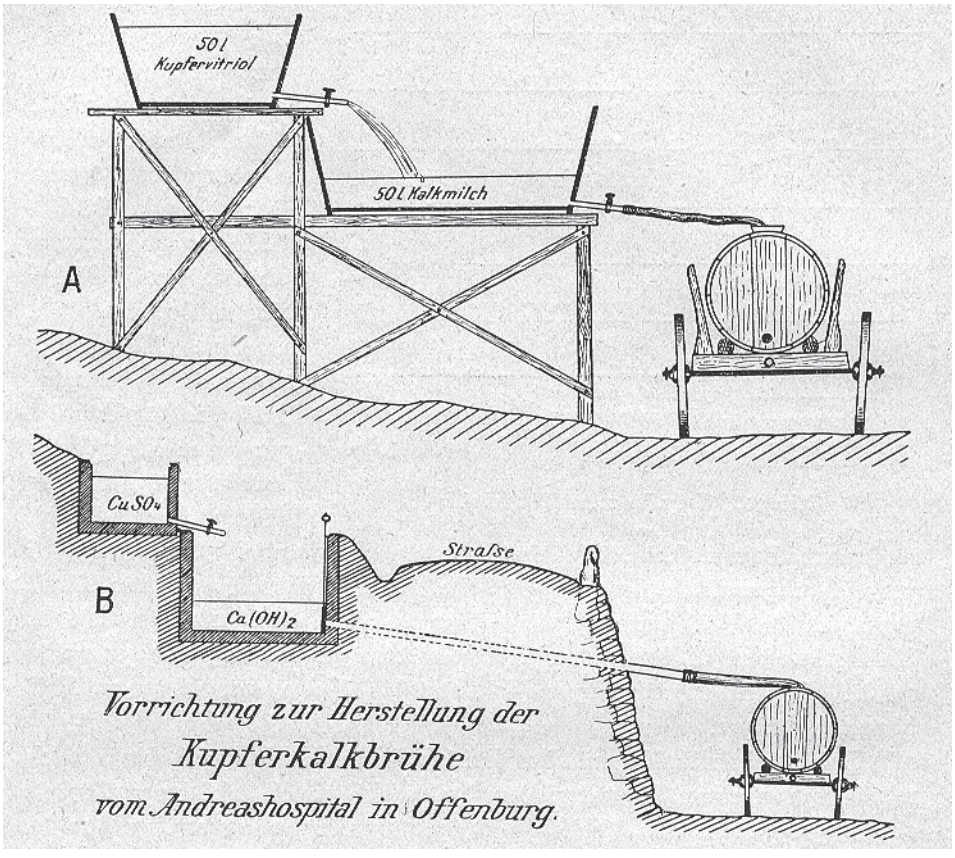

Figure 13. Gun form recommended for treatment of the lower leaf surface (drawing from Müller, 1918).

equipment was frequently used in sloped vineyards near farmhouses. The system consisted of a high-pressure pump, a tank and a fixed tube distribution system leading to and extending along the rows of the vineyard. There were valves at various points in the distribution system to which a flexibe hose with a spray gun could be attached. Treatments would be made by hand with this gun, until the leaves and bunches were completely wet. The advantage of this system was that it could be used in situations in which a driven blower could not be used, such as on steep slopes, and on terraces and when the ground was wet. An alternative to these rather costly installations was the knapsack sprayer (20 L capacity). The first knapsack sprayers were equipped with hand pumps and later models were equipped with motorized pumps. However, these sprayers could only be used in very small plots and have now been replaced by knapsack blowers that operate with much smaller spray volumes (around 250 L ha^{-1}).

In large, step-terraced vineyards, applications remained cumbersome and the use of modern low-volume spraying techniques capable of taking full advantage of modern fungicides remained difficult or impossible, but helicopters offered a solution. The first trials involving helicopters were conducted in the early 1950's. Zobrist and colleagues (Zobrist *et al.*, 1953a) reported on large Swiss experiments in which vines were sprayed against downy mildew from a helicopter. In that study, an area of 83 ha was covered in 20 working hours, which included 102 flights and a total flying time of 9 h, 42 min. This trial used 13,600 L (150 L ha^{-1}) of a 6% cuprosan mixture, at a cost of Fr. 63 per ha for the application alone (Fr. 120 including the cost of the fungicide). In a 30 ha vineyard, the work was accomplished in 5 h, including 2 h, 28 min of flying time, at a cost of Fr. 44 per application and Fr. 101 per ha (Zobrist *et al.*, 1953b). Low-volume spraying from helicopters is now routine in step-terraced vineyards.

There has been great interest in the idea of passive protection systems consisting principally of a source of fungicide that would deliver the chemical during rain. To this end, in the 1970's, hail nets interwoven with copper wires and nets impregnated with copper salts were tested (Olivelli, 1976) and sold. These products were apparently effective and profitable. However, to our knowledge, no similar products are currently on the market.

Acknowledgments

As well over 3000 papers (CAB 1910–2011) have dealt with *P. viticola*, not all could be cited in this review. We apologize to our colleagues whose work was not acknowledged appropriately. We tried, to the best of our knowledge, to cite the most complete papers or the first paper on a particular topic. As the overwhelming portion of the literature on grapevine downy mildew deals with chemical control and/or infection conditions in particular locations, we chose to omit those studies from this discussion. We offer special thanks to reviewers for their extensive and invaluable work. We thank Maria Cristina Palmieri for figures 6 and 9; Hanns-Heinz Kassemeyer for figures 7 and 8.

Literature cited

Albert G., J. Curtze and C.A. Drandarevski, 1988. Dimethomorph (CME 151), a novel curative fungicide In: *Proceedings of an International Conference, Brighton Crop Protection Conference: Pests & Diseases,* 16–19 November 1988. Brighton, UK, Vol. 1, 17–24.

Allègre M., M.C. Héloir, S. Trouvelot, X. Daire, A. Pugin, D. Wendehenne and M. Adrian, 2009. Are grapevine stomata involved in the elicitor-induced protection against downy mildew? *Molecular Plant-Microbe Interactions* 22, 977–986.

Alleweldt G. and J.V. Possingham, 1988. Progress in grapevine breeding. *Theoretical and Applied Genetics* 75, 669–673.

Alonso-Villaverde V., F. Voinesco, O. Viret, J.L. Spring and K. Gindro, 2011. The effectiveness of stilbenes in resistant *Vitaceae*: ultrastructural and biochemical events during *Plasmopara viticola* infection process. *Plant Physiology and Biochemistry* 49, 265–274.

Andrieu N., J.L. Genet, G. Jaworska and G. Bompeix, 2000. Behaviour of famoxadone deposits on grape leaves. *Pest Management Science* 56, 1036–1042.

Andrieu N., G. Jaworska, J.L. Genet and G. Bompeix, 2001. Biological mode of action of famoxadone on *Plasmopara viticola* and *Phytophthora infestans*. *Crop Protection* 20, 253–260.

Arens K., 1929a. Untersuchungen über Keimung und Zytologie der Oosporen von *Plasmopara viticola* (Berl, et de Toni). *Jahrbuch* für *Wissenschaftliche Botanik* 70, 57–92.

Arens K., 1929b. Physiologische Untersuchungen an *Plasmopara viticola*, unter besonderer Berücksichtigung der Infektionsbedingungen. *Jahrbuch für Wissenschaftliche Botanik* 70, 93–157.

Armet H., 1934. Les bouillies célestes au sulfate d'ammoniaque et solutions aux composes iartriques. *Progres Agricole et Viticole* 102, 86–89.

Arnone A., G. Nasini, W. Panzeri, O.V.d. Pava and L. Malpezzi, 2008. Acremine G, dimeric metabolite from

cultures of *Acremonium byssoides* A20. *Journal of Natural Products* 71, 146–149.

Arpai J., O. Eoltyn and O.G. Janotkova, 1957. A study of the appearance and fungistatic activity of *Trichothecium plasmoparae* Viola & Marsais. *Polnohospodarstvo* 4, 68–87.

Aziz A., B. Poinssot, X. Daire, M. Adrian, A. Bezier, B. Lambert, J.M. Joubert and A. Pugin, 2003. Laminarin elicits defense responses in grapevine and induces protection against *Botrytis cinerea* and *Plasmopara viticola*. *Molecular Plant-Microbe Interactions* 16, 1118–1128.

Aziz A., P. Trotel-Aziz, L. Dhuicq, P. Jeandet, M. Couderchet and G. Vernet, 2006. Chitosan oligomers and copper sulfate induce grapevine defense reactions and resistance to gray mold and downy mildew. *Phytopathology* 96, 1188–1194.

Bachala A., 1927. Les poudrages et la protection du vignoble. *Progres Agricole et Viticole* 88, 60–61.

Bakshi S., A. Sztejnberg and O. Yarden, 2001. Isolation and characterization of a cold-tolerant strain of *Fusarium proliferatum*, a biocontrol agent of grape downy mildew. *Phytopathology* 91, 1062–1068.

Baldacci E., 1947. Epifitie di *Plasmopara viticola* (1941–46) nell'Oltrepo pavese ed adozione del calendario di incubazione come istrumento di lotta. *Atti Istituto Botanico Laboratorio Crittogamico di Pavia* 8, 45–85.

Baldacci E., 1949. Il calendario d'incubazione della peronospora della vite e le sue possibili applicazioni. *Annali Academia Agricola Torino* 91, 8 pp.

Baldacci E., 1954. Le Mildiou en Italie. *Bulletin de l'Office International du Vin* 280, Tiré à part, 21 pp.

Baldacci E. and E. Refatti, 1956. La lotta contro le due maggiori ampelopatie: peronospora (*Plasmopara viticola*) e oidio (*Uncinula necator*) in relazione alla conoscenza dei periodi di incubazione e dei cicli biologici. *Annali della Facoltà di Agraria di Milano* V, 35, 151–178.

Baldacci E., E. Betto, G. Brazzola, M. Brazzola and G. Fogliani, 1962. Ricerche comparative sulle consequenze del fungicidi a base di zineb e di rame sulla vegetazione e sulla produzione dalla Vite e sulla modalita d'impiego nella lotta antiperonosporica. *Rivista di Patologia Vegetale* 2, 34–112.

Bassi A., R. Alber, J.L. Genet, W. Massasso and C. Sbriscia, 2000. DPX-MP560 (EquationReg.System) antiperonosporico della vite di nuova concezione. Profilo e risultati sperimentali (1996–99). In: *Atti Giornate Fitopatologiche,* Vol. 2, 16–20, Aprile 2000, Perugia, Italy, 129–136.

Beckers G.J.M. and U. Conrath, 2007. Priming for stress resistance: from the lab to the field. *Current Opinion in Plant Biology* 10, 425–431.

Bellin D., E. Peressotti, D. Merdinoglu, S. Wiedemann-Merdinoglu, A.F. Adam-Blondon, G. Cipriani, M. Morgante, R. Testolin and G. Di Gaspero, 2009. Resistance to *Plasmopara viticola* in grapevine 'Bianca' is controlled by a major dominant gene causing localised necrosis at the infection site. *Theoretical and Applied Genetics* 120, 163–176.

Berlese A.N. and J.B. de Toni, 1888. In Saccardo, *Sylloge Fungorum Omnium hucusque cognitorum*, VII. (Pars. I.) *Phycomyceteae* auctoribus, *Peronosporaceae*, 233–264.

Blaeser M., 1978. Untersuchungen zur Epidemiologie des falschen Mehltaus an Weinrebe, *Plasmopara viticola* (Berk. et de Toni). *PhD Thesis Universität Bonn Germany*, 127 pp.

Blaeser M. and H.C. Weltzien, 1978. Die Bedeutung von Sporangienbildung, -ausbreitung und -keimung für die Epidemiebildung von *Plasmopara viticola*. *Zeitschrift* für *Pflanzenkrankheiten und Pflanzenschutz* 85, 155–161.

Blaeser M. and H.C. Weltzien, 1979. Epidemiological studies of *Plasmopara viticola* for improving determination of spraying dates. *Zeitschrift* für *Pflanzenkrankheiten und Pflanzenschutz* 86, 489–498.

Blaise P. and C. Gessler, 1990. Development of a forecast model of grape downy mildew on a microcomputer. *Acta Horticulturae* 276, 63–70.

Blaise P., R. Dietrich and M. Jermini, 1996. Coupling a disease epidemic model with a crop growth model to simulate yield losses of grapevine due to *Plasmopara viticola*. *Acta Horticulturae* 416, 285–291.

Blaise P., M. Jermini and C. Gessler, 1997. Sampling system to estimate *P. viticola* severity in a vineyard based on tylor's power law: application to simulated data. *Viticultural and Enological Sciences* 52, 133–135.

Blaise P., R. Dietrich and M. Jermini, 1999. A new demand function for grapevine fruits in Vinemild. *Acta Horticulturae* 499, 253–260.

Blasi P., S. Blanc, S. Wiedemann-Merdinoglu, E. Prado, E. Rühl, P. Mestre and D. Merdinoglu, 2011. Construction of a reference linkage map of *Vitis amurensis* and genetic mapping of *Rpv8*, a locus conferring resistance to grapevine downy mildew. *Theoretical and Applied Genetics* DOI: 10.1007/s00122-011-1565-0.

Blattner V. and J. Schaller, 1990. Resistenzzüchtung zur Lösung weinbautechnischer Probleme. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 126, 201–203.

Bleyer G., B. Huber, V. Steinmetz and H.H. Kassemeyer, 2003. Growth models, a tool to define spray intervals against downy mildew (*Plasmopara viticola*). *Bulletin OILB/SROP* 26, 8, 7–11.

Bleyer G., H.H. Kassemeyer, R. Krause, O. Viret and W. Siegfried, 2008. "Vitimeteo Plasmopara" - Prognosemodell zur Bekampfung von *Plasmopara viticola* (Rebenperonospora) im Weinbau *Gesunde Pflanzen* 60, 91–100.

Blumer S. and E. Peyek, 1947. Versuche zur Bekämpfung der Peronospora im Weinbau. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 56, 62–67.

Bosc M., 1935. Bouillies cupriqiies au sulfate d'ammoniaque. *Progres Agricole et Viticole* 103, 562–566.

Boso S., V. Alonso-Villaverde, J.L. Santiago, P. Gago, M. Dürrenberger, M. Düggelin, H.H. Kassemeyer and M.C. Martinez, 2010. Macro- and microscopic leaf characteristics of six grapevine genotypes (*Vitis* spp.) with different susceptibilities to grapevine downy mildew. *Vitis* 49, 43–50.

Boubals D., 1959 Contribution a l'étude des causes de la ré-

sistance des vitacées au mildeou de la vigne (*Plasmopara viticola*) et de leur mode de transmission héréditaire. Thèse de doctorat Université de Montpellier. *Annales de l'amélioration des plantes I*, 5–233

Boubals D. and R. Lafon, 1981. Control of grapevine downy mildew by penetrating systemic fungicides. *Bulletin de l'organisation international de la vigne et du vin* 54, 319–335.

Boubals D. and G. Mur, 1983. Difficultés concernant des fongicides viticoles. *Progres Agricole et Viticole* 100, 109–113.

Boubals D., G. Mur and O.F. Rivaldo, 1979. Resultats de l'essai 1979 de nouveaux fongicides destines a lutter contre le mildiou de la vigne. *Progres Agricole et Viticole* 96, 448–454.

Boubals D. and A. Vergnes, 1953. Essais de fongicides organiques dans la lutte contre le mildiou de la Vigne. *Progres Agricole et Viticole* 139, 90–97.

Boubals D. and A. Vergnes, 1954. Essais de fongicides organiques dans la lutte contre le mildiou de la Vigne effectués en 1953. *Progres Agricole et Viticole* 142, 6–12.

Boubals D., A. Vergnes and H. Bobo, 1955. Essais de fongicides organiques dans la lutte contre le mildiou de la Vigne effectués en 1954. *Progres Agricole et Viticole* 143, 64–74.

Boubals D., A. Vergnes and H. Bobo, 1956. Essais de fongicides organiques et organo-cupriques dans la lutte centre le mildiou de la Vigne effectués en 1955. *Progres Agricole et Viticole* 145, 63–67.

Bouquet A., J. Pauquet, A.F. Adam-Blondon, L. Torregrosa, D. Merdinoglu and S. Wiedemann-Merdinoglu, 2000. Vers l'obtention de varietés de vigne résistantes a l'oidium et au mildiou par les methodes conventionnelles et biotechnologiques. *Bulletin de l'organisation international de la vigne et du vin* 73, 445–452.

Brook P.J., 1979. Effect of light on sporulation of *Plasmopara viticola*. *New Zealand Journal of Botany* 17, 135–138.

Brown M.V., J.N. Moore, P. Fenn and R.W. McNew, 1999. Evaluation of grape germplasm for downy mildew resistance. *Fruit Varieties Journal* 53, 22–29.

Bugaret Y. and L. Burosse, 2005. Contre le mildiou, quand commencer a traiter, comment continuer, et avec quels fongicides: prise de décision en protection raisonnée. *Phytoma* 583, 58–60, 62–64.

Bugaret Y., P. Sauris and M. Clerjeau, 1998. Mildiou et black-rot de la vigne: efficacité de l'azoxystrobine au vignoble. *Phytoma* 504, 69–72.

Burger P, Bouquet A and Striem MJ. 2009. Grape breeding. In: *Breeding Plantation Tree Crops: Temperate Species*. (Mohan J.S., Priyadarhan P.M., ed.). Springer Verlag Berlin, Germany, 161–189.

Burruano S., A. Alfonzo, S.l. Piccolo, G. Conigliaro, V. Mondello, L. Torta, M. Moretti and G. Assante, 2008. Interaction between *Acremonium byssoides* and *Plasmopara viticola* in *Vitis vinifera*. *Phytopathologia Mediterranea* 47, 122–131.

Burruano S., G. Conigliaro and M. Di-Graziano, 1990. Prime indicazioni sull'azione delle basse temperature sulla germinazione delle oospore di *Plasmopara viticola*. *Phytopathologia Mediterranea* 29, 73–75.

Busam G., H.H. Kassemeyer and U. Matern, 1997. Differential expression of chitinases in *Vitis vinifera* L. responding to systemic acquired resistance activators or fungal challenge. *Plant Physiology* 115, 1029–1038.

Cadle-Davidson L., 2008. Variation within and between *Vitis* spp. for foliar resistance to the downy mildew pathogen *Plasmopara viticola*. *Plant Disease* 92, 1577–1584.

Cadoret A., 1923. Bouillies bleues et vignes bleues. 7^{e} année d'experiences. *Progres Agricole et Viticole* 40, 380–382.

Cadoret A., 1927. Contribution a l'étude des traitements contre le mildiou. Détermination des époques d'attaques du champignon. *Progres Agricole et Viticole* 87, 362–365.

Cadoret A., 1931. Les léçons du mildiou en 1930. *Progres Agricole et Viticole* 95, 187–188.

Caffi T., V. Rossi and R. Bugiani, 2010. Evaluation of a warning system for controlling primary infections of grapevine downy mildew. *Plant Disease* 94, 709–716.

Caffi T., V. Rossi, R. Bugiani, F. Spanna, L. Flamini, A. Cossu and C. Nigro, 2009. A model predicting primary infections of *Plasmopara viticola* in different grapevine-growing areas of Italy. *Journal of Plant Pathology* 91, 535–548.

Caffi T., V. Rossi and M. Lusitani, 2011. Long-term survival of *Plasmopara viticola* oospores. *IOBC-bulletin* in press.

Cerasoli E., 1924. Il solfato di rame nei riguardi dell' agricultura. *Giornale Chimica Industriale ed Applicata* 6, 536–537.

Cesari A., R. Fiaccadori and E. Velicogna, 1981. Verifica tecnico-economica su fungicidi endoterapici applicati nella difesa antiperonosporica della vite. *Vignevini* 8, 43–49.

Chaboussou F., R. Lafon, J.P. Carles and G. Raise, 1974. Efficacité a l'égard du mildiou et répercussions sur l'érinose de la vigne de diverses formules fongicides cupro-organiques. *Phytiatrie-Phytopharmacie* 23, 89–106.

Chazalet M., P. Crisinel, D. Horriere and J. Thiolliere, 1977. Résultats de l'expérimentation réalisée avec les ethyl phosphites contre différentes maladies de la vigne. *Phytiatrie-Phytopharmacie* 26, 41–54.

Chen J., G. Dai, Z. Gu and Y. Miao, 2002. Inhibition effect of 58 plant extracts against grape downy mildew (*Plasmopara viticola*). *Natural Product Research and Development* 14, 9–13.

Chevalier G., 1934. Les bouillies cupro-ammoniacales. *Progres Agricole et Viticole* 102, 62.

Chong J., G. Le Henanff, C. Bertsch and B. Walter, 2008. Identification, expression analysis and characterization of defense and signaling genes in *Vitis vinifera*. *Plant Physiology and Biochemistry* 46, 469–481.

Clerjeau M., C. Moreau, B. Piganeau and G. Malato, 1984. Resistance of *Plasmopara viticola* to anilide-fungicides: evaluation of the problem in France. *Mededelingen van de Faculteit Landbouwwetenschappen Rijksuniversiteit Gent* 49, 179–184.

Cohen Y., M. Reuveni and A. Baider, 1999. Local and

systemic activity of BABA (DL-3-aminobutyric acid) against *Plasmopara viticola* in grapevines. *European Journal of Plant Pathology* 105, 351–361.

Cohen Y., W.Q. Wang, B.H. Ben-Daniel and Y. Ben-Daniel, 2006. Extracts of *Inula viscosa* control downy mildew of grapes caused by *Plasmopara viticola*. *Phytopathology* 96, 417–424.

Comte, F., 1935. Essais de bouilles cupriques an bicarbonate de soude dans la lutte centre le mildiou. *Progres Agricole et Viticole* 103, 229–234.

Conrath U., G.J.M. Beckers, V. Flors, P. Garcia-Agustin, G. Jakab, F. Mauch, M.A. Newman, C.M.J. Pieterse, B. Poinssot, M.J. Pozo, A. Pugin, U. Schaffrath, J. Ton, D. Wendehenne, L. Zimmerli and B. Mauch-Mani, 2006. Priming: getting ready for battle. *Molecular Plant-Microbe Interactions* 19, 1062–1071.

Cordonnier R., 1955. Etude, du point de vue œnologique, des fongicides organiques de synthèse a base d'ethylenebisdithiocarbamate de zinc et de N-trichloro-methylthio-tetrahydrophtalimide employés dans le traitement du mildiou de la Vigne. *Progres Agricole et Viticole* 143, 8–12.

Cséfalvay L., G. Di Gaspero, K. Matous, D. Bellin, B. Ruperti and J. Olejnickova, 2009. Pre-symptomatic detection of *Plasmopara viticola* infection in grapevine leaves using chlorophyll fluorescence imaging. *European Journal of Plant Pathology* 125, 291–302.

Cucchi N.J.S., C.D. Catania, A.E. Piratti and P.F. Rosell, 1974. Durée de l'efficacité des fongicides utilises en viticulture et l'influence de leurs résidus sur la fermentation vinique. *Bulletin de l'Organisation International de la Vigne et du Vin* 47, 209–217.

Dagostin S., H. Schärer, I. Pertot and L. Tamm, 2011. Are there alternatives to copper for controlling grapevine downy mildew in organic viticulture? *Crop Protection*, in press.

Dagostin S., T. Formolo, O. Giovannini, I. Pertot and A. Schmitt, 2010. *Salvia officinalis* extract can protect grapevine against *Plasmopara viticola*. *Plant Disease* 94, 575–580.

Dai G.H., C. Andary, L. Mondolot-Cosson and D. Boubals, 1995. Histochemical studies on the interaction between three species of grapevine, *Vitis vinifera*, *V. rupestris* and *V. rotundifolia* and the downy mildew fungus, *Plasmopara viticola*. *Physiological and Molecular Plant Pathology* 46, 177–188.

Darpoux H., 1949. Les avertissements agricoles. *Bulletin. Technique Inform. Ingrs Service agriculture*, Fr, No. 41, 403–411.

De Bary A., 1863. Recherches sur le développement de quelques champignons parasites. *Annales Sciences Naturelles. Paris, France* Serie IV, 20, 5–148.

De Gibon J., 1929. Poudrages insecticides et anticryptogamiques en viticulture. *Vie Agricole et Rurale* 18, 251–253.

Delmotte F., W. J. Chen, s. Richard-Cervera, C. Greif, D. Papura, X. Giresse, G. Mondor-Genson and M.F. Corio-Costet, 2006. Microsatellite DNA markers for *Plasmopara viticola*, the causal agent of downy mildew of grapes. *Molecular Ecology Notes* 6(2), 379–381.

Denzer H., G. Staudt and E. Schlosser, 1995. Das Verhalten von *Plasmopara viticola* auf resistenten und anfälligen Rebsorten. *Vitis* 34, 113–117.

Dercks W. and L.L. Creasy, 1989a. The significance of stilbene phytoalexins in the *Plasmopara viticola*- grapevine interaction. *Physiological and Molecular Plant Pathology* 34, 189–202.

Dercks W. and L.L. Creasy, 1989b. Influence of fosetyl-Al on phytoalexin accumulation in the *Plasmopara viticola*-grapevine interaction. *Physiological and Molecular Plant Pathology* 34, 203–213.

Di Gaspero G. and G. Cipriani, 2003. Nucleotide binding site/leucine-rich repeats, Pto-like and receptor-like kinases related to disease resistance in grapevine. *Molecular genetics and genomics* 269, 612–623.

Di Gaspero G., G. Cipriani, A.F. Adam-Blondon and R. Testolin, 2007. Linkage maps of grapevine displaying the chromosomal locations of 420 microsatellite markers and 82 markers for R-gene candidates. *Theoretical and Applied Genetics* 114, 1249–1263.

Di Gaspero G., R. Marconi, G. Cipriani and R. Testolin, 2009. Localisation of NBS-LRR genes in the grape genome. *Acta Horticulturae* 827, 53–58.

Diez-Navajas A.M., S. Wiedemann-Merdinoglu, C. Greif and D. Merdinoglu, 2008. Nonhost versus host resistance to the grapevine downy mildew, *Plasmopara viticola*, studied at the tissue level. *Phytopathology* 98, 776–780.

Doddapaneni H., H. Lin, M.A. Walker, J.Q. Yao and E.L. Civerolo, 2008. VitisExpDB: a database resource for grape functional genomics. *Bmc Plant Biology* 8, 23.

Dubreuil-Maurizi C., S. Trouvelot, P. Frettinger, A. Pugin, D. Wendehenne and B. Poinssot, 2010. β-aminobutyric acid primes an NADPH oxidase-dependent reactive oxygen species production during grapevine-triggered immunity. *Molecular Plant-Microbe Interactions* 23, 1012–1021.

Duriatti A., R. Benzekri, J.P. Lefebvre and D. Driant, 2003. La zoxamide: fongicide antimildiou. *Phytoma* 557, 52–54.

Dutzmann S., 1999. Iprovalicarb (SZX 0722) - a novel fungicide with specific activity against oomycetes. *Pflanzenschutz-Nachrichten Bayer* 52, 15–32.

Egger E., 2008. La revisione non penalizza la lotta a peronospora e oidio. *Informatore Agrario* 13, 29–41.

Egger E., E. Marinelli and G. Greco, 1994. Un nuovo modello gestionale per il controllo della *Peronospora* della vite: prime verifiche nell'ambiente viticolo aretino. *Rivista di Viticoltura e di Enologia* 47, 15–32.

Eibach R., E. Zyprian, L. Welter and R. Topfer, 2007. The use of molecular markers for pyramiding resistance genes in grapevine breeding. *Vitis* 46, 120–124.

Faes H., 1929. La lutte contre les parasites de la Vigne et les traitements obligatoires en Suisse. *Revue de Viticulture* 70, 217–221.

Faes H., 1943. Station fédérale d'essais viticoles et arboricoles a Lausanne et Domaine de Pully. Rapports annuels 1941 et 1942. *Annuaire agricole de la Suisse* 44,

462–498.
Faes H. and M. Staehelin, 1927. La lutte contre les parasites, insectes et champignons de la Vigne en 1925 et 1936. *Annuaire Agricole de la Suisse* 28, 373–396.
Faes H. and M. Staehelin, 1930. La lutte contre les parasites de la Vigne, insectes et champignons, en 1929. *Annuaire agricole de la Suisse* 31, 123–133.
Falk S.P., R.C. Pearson, D.M. Gadoury, R.C. Seem and A. Sztejnberg, 1996. *Fusarium proliferatum* as a biocontrol agent against grape downy mildew. *Phytopathology* 86, 1010–1017.
Farlow W.G., 1876. The American grape mildew. *Bussy Institut Bulletin* 1, 423.
Ferri M., A. Tassoni, M. Franceschetti, L. Righetti, M.J. Naldrett and N. Bagni, 2009. Chitosan treatment induces changes of protein expression profile and stilbene distribution in *Vitis vinifera* cell suspensions. *Proteomics* 9, 610–624.
Figueiredo A., A.M. Fortes, S. Ferreira, M. Sebastiana, Y.H. Choi, L. Sousa, B. Acioli-Santos, F. Pessoa, R. Verpoorte and M.S. Pais, 2008. Transcriptional and metabolic profiling of grape (*Vitis vinifera* L.) leaves unravel possible innate resistance against pathogenic fungi. *Journal of Experimental Botany* 59, 3371–3381.
Fischer B.M., I. Salakhutdinov, M. Akkurt, R. Eibach, K.J. Edwards, R. Topfer and E.M. Zyprian, 2004. Quantitative trait locus analysis of fungal disease resistance factors on a molecular map of grapevine. *Theoretical and Applied Genetics* 108, 501–515.
Fung R.W.M., W.P. Qiu, Y.C. Su, D.P. Schachtman, K. Huppert, C. Fekete and L.G. Kovacs, 2007. Gene expression variation in grapevine species *Vitis vinifera* L. and *Vitis aestivalis* Michx. *Genetic Resources and Crop Evolution* 54, 1541–1553.
Galet P., 1977. *Les Maladies et les Parasites de la Vigne*. Tome 1. Imprimerie du Paysan du midi, Montpellier, France, 871 pp.
Gallay R., 1948. Etat actuel de la lutte antiparasitaire en Suisse. *Proceedings Ier. Congres International Phytopharmacologie*, 1946, 601–624.
Gallay R. and M. Staehelin, 1949. La lutte centre le mildiou de la Vigne vue a la lumière des expériences de l'année 1948. *Revue romande d'Agriculture, de Viticulture et d'Arboriculture* 5, 11–15.
Gallay R., M. Staehelin and G. Trivelli, 1945. La lutte contre le mildiou de la vigne. Les divers types de bouillies cupriques. *Revue Romande d'Agriculture, de Viticulture et d'Arboriculture* 1, 3–5.
Gallay R., C. Terrier and G. Trivelli, 1951. Le mildiou de la Vigne-les expériences de 1951. *Revue Romande d'Agriculture, de Viticulture et d'Arboriculture* 7, 81–90.
Gamberini O., V. Lazzari, G. Mitrangolo, P. Piazzi and A. Santini, 2000. Fenomen Reg. (fenamidone) nuovo fungicida antiperonosporico per la difesa della vite. In: *Atti Giornate Fitopatologiche,* Vol. 2, 16–20, Aprile 2000, Perugia, Italy, 9–14.
Gaudineau M. and C.M. Messiaen, 1953. Mildiou de la Vigne et nouveaux produits de lutte. *Ann Reck, agron* 4, 185–208.
Gäumann E., 1927. Die wirtschaftliche Bedeutung unserer wichtigsten Pflanzenkrankheiten. *Landwirtschaftliches Jahrbuch der Schweiz* 12, 319–324.
Gay-Bellile F., J. Lacouture, J.F. Sarrazin, Y. Courlit and E. Menard, 1983. Le point sur le mildiou de la vigne. Les fongicides systémiques. Les souches résistantes. *Progres Agricole et Viticole* 100, 83–89.
Gehmann K., G. Staudt and F. Grossmann, 1987. Der Einfluss der Temperatur auf die Oosporenbildung von *Plasmopara viticola*. *Zeitschrift für Pflanzenkrankheiten und Pflanzenschutz* 94, 230–234.
Genet J.L., G. Jaworska and F. Deparis, 2006. Effect of dose rate and mixtures of fungicides on selection for QoI resistance in populations of *Plasmopara viticola*. *Pest Management Science* 62, 188–194.
Gentili E., D. Tasselli, M. Coatti and M. Manaresi, 2002. Cyazofamid: fungicida a meccanismo d'azione innovativo nei confronti della peronospora della vite. In: *Atti Giornate Fitopatologiche*, Vol. 2, 7–11, Aprile 2000, Baselga di Pine, Trento, Italy, 125–130.
Gherardi I., A. Brunelli and R. Bugiani, 1999. Modello a prognosi negativa per le infezioni primarie di *Plasmopara viticola*. *Frustula Entomologica* 22, 87–91.
Giles N.B., N.B. Akesson and W.E. Yates, 2008. Pesticide application technology: research and development and the growth of the industry. *Transaction of the ASABE* 51, 397–403.
Gisi U., J. Harr, R. Sandmeijer and H. Wiedmer, 1983. A new systemic oxazolidinone fungicide (SAN 371 F) against diseases caused by *Peronosporales*. *Mededelingen van de Faculteit Landbouwwetenschappen Rijksuniversiteit Gent* 48, 541–549.
Gisi U., H. Sierotzki, A. Cook and A. McCaffery, 2002. Mechanisms influencing the evolution of resistance to Qo inhibitor fungicides. *Pest Management Science* 58, 859–867.
Gisi U. and H. Sierotzki, 2008. Fungicide modes of action and resistance in downy mildews. (Special Issue: The downy mildews - genetics, molecular biology and control.). *European Journal of Plant Pathology* 122, 157–167.
Gisi U., M. Waldner, N. Kraus, P.H. Dubuis and H. Sierotzki, 2007. Inheritance of resistance to carboxylic acid amide (CAA) fungicides in *Plasmopara viticola*. *Plant Pathology* 56, 199–208.
Giuntoli A. and S. Orlandini, 2000. Effects of downy mildew on photosynthesis of grapevine leaves. *Acta Horticulturae* 526, 461–466.
Gobbin D., I. Pertot and C. Gessler, 2003a. Identification of microsatellite markers for *Plasmopara viticola* and establishment of high throughput method for SSR analysis. *European Journal of Plant Pathology* 109, 153–164.
Gobbin D., I. Pertot and C. Gessler, 2003b. Genetic structure of a *Plasmopara viticola* population in an isolated Italian mountain vineyard. *Journal of Phytopathology* 151, 636–646.
Gobbin D., M. Jermini, B. Loskill, I. Pertot, M. Raynal and C. Gessler, 2005. Importance of secondary inoculum of *Plasmopara viticola* to epidemics of grapevine downy

mildew. *Plant Pathology* 54, 522–534.
Gobbin D., A. Rumbou, C.C. Linde and C. Gessler, 2006. Population genetic structure of *Plasmopara viticola* after 125 years of colonization in European vineyards. *Molecular Plant Pathology* 7, 519–531.
Gobbin D., G. Valsesia and C. Gessler, 2001. Genetic variability of *Plasmopara viticola* between and within selected populations. *Bulletin OILB/SROP* 24, 7, 19–23.
Godard S., I. Slacanin, O. Viret and K. Gindro, 2009. Induction of defence mechanisms in grapevine leaves by emodin- and anthraquinone-rich plant extracts and their conferred resistance to downy mildew. *Plant Physiology and Biochemistry* 47, 827–837.
Godet C., 1923. Rapport sur l'activité de la Station d'essais viticoles a Auvarnier. Exercices 1919–1920 et 1920–1921. *Annuaire Agricole de la Suisse* 24, 199–312.
Godet F., J.F. Roques and J.M. Compagnon, 1997. L'azoxystrobine: fongicide polyvalent cereales et vigne. *Phytoma* 50, 52–54.
Goidànich G., 1964. Le Peronosporales. In: *Manuale di Patologia Vegetale*, Vol. II. Edizione agricole, Bologna, Italy, 315–346, 1283 pp.
Goidanich G. and B. Casarini, 1961. II calendario dei trattamenti alla Vite. *Progresso Agricolo*, A. 7, (5), 620–631.
Gouot J.M., 2006. Field efficacy of Profiler, a fluopicolide and fosetyl-Al fungicide combination for the control of grape downy mildew (*Plasmopara viticola*). *Pflanzenschutz-Nachrichten Bayer* 59, 293–302.
Grabski C. and U. Gisi, 1987. Quantification of synergistic interactions of fungicides against *Plasmopara* and *Phytophthora*. *Crop Protection* 6, 64–71.
Grando M.S., D. Bellin, K.J. Edwards, C. Pozzi, M. Stefanini and R. Velasco, 2003. Molecular linkage maps of *Vitis vinifera* L. and *Vitis riparia* Mchx. *Theoretical and Applied Genetics* 106, 1213–1224.
Graniti A., 1954. Prove di lotta contro la Peronospora della Vite in Sardegna nell'anno 1952. *Notiziario sulle Malattie delle Piante* 24–25, 75–82.
Gregory C., 1915. Studies on *Plasmopara viticola*. *Reprint from official report of the Session of the internatioal congress of viticulture*, July 12–13, San Francisco, CA, USA, 126–150.
Grünzel H., 1960. Studien zur biologischen Differenzierung des Falschen Mehltaues der Weinreben (*Peronospora viticola* de Bary). *Phytopathologische Zeitschrift* 39, 149–194.
Grünzel H., 1961. Untersuchungen über die Oosporenbildung beim falschen Mehltau der Weinrebe (*Peronospora viticola* de Bary). *Zeitschrift für Pflanzenkrankheiten, Pflanzenpathologie und Pflanzenschutz* 68, 65–80.
Gullino M.L., E. Mescalchin and M. Mezzalama, 1997. Sensitivity to cymoxanil in populations of *Plasmopara viticola* in northern Italy. *Plant Pathology* 46, 729–736.
Guyot L., 1927. Le probleme actual des antioryptogamiques. IV. Les anticryptogamiques collcidaux. *Journal d'Agriculture pratique* 91, 78–80.
Hadorn C., 1942. Rationierung der Kupferspritzmittel in der Landwirtschaft. *Schweizerische Zeitschrift für Obst– und Weinbau* 51, 178–180.
Hadorn C., 1943. Vergleich der Wirksamkeit verschiedener Spritzbrühen gegen den falschen Rebenmehltau. *Schweizerische Zeitschrift für Obst- und Weinbau* 52, 54–69.
Hamiduzzaman M.M., G. Jakab, L. Barnavon, J.M. Neuhaus and B. Mauch-Mani, 2005. ß-Aminobutyric acid-induced resistance against downy mildew in grapevine acts through the potentiation of callose formation and jasmonic acid signaling. *Molecular Plant-Microbe Interactions* 18, 819–829.
Harm A., H.H. Kassemeyer, T. Seibicke and F. Regner 2011. Evaluation of chemical and natural resistance inducers against downy mildew (*Plasmopara viticola*) in grapevine. *American Journal of Enology and Viticulture*, doi: 10.5344/ajev.2011.09054.
Hayes M.A., A. Feechan and I.B. Dry, 2010. Involvement of abscisic acid in the coordinated regulation of a stress-inducible hexose transporter (VvHT5) and a cell wall invertase in grapevine in response to biotrophic fungal infection. *Plant Physiology* 153, 211–221.
Heaney S.P., J.C. Atger and J.F. Roques, 1988. ICIA0001: A novel fungicide for use against diseases on vines In: *Proceedings of an International Conference, Brighton Crop Protection Conference: Pests & Diseases,* 16–19 November 1988. Brighton, UK, Vol. 1, 245–250.
Hengl F., 1923. Vergleichende Versuche gegen verschiedene Rebenschädlinge. *Allgemeine Weinzeitschrift* 40, 1–5.
Hill G.K., 1998. Studies on the germination of *Plasmopara viticola* oospores with a floating disc test. *Bulletin OILB/SROP* 21, 2, 1–2.
Hill G.K., 2000. Simulation of *P. viticola* oospore-maturation with the model SIMPO. *Bulletin OILB/SROP* 23, 4, 7–8.
Jacquemain R. and Gravier, 1932. Note sur deux spécialités anticryptogamiques *Comptes rendus Congres Soc. Savantes Paris et Departements, Section des Sciences*, 293–294.
Jaillon O., J.M. Aury, B. Noel, A. Policriti, C. Clepet, A. Casagrande, N. Choisne, S. Aubourg, N. Vitulo, C. Jubin, A. Vezzi, F. Legeai, P. Hugueney, C. Dasilva, D. Horner, E. Mica, D. Jublot, J. Poulain, C. Bruyere, A. Billault, B. Segurens, M. Gouyvenoux, E. Ugarte, F. Cattonaro, V. Anthouard, V. Vico, C. Del Fabbro, M. Alaux, G. Di Gaspero, V. Dumas, N. Felice, S. Paillard, I. Juman, M. Moroldo, S. Scalabrin, A. Canaguier, I. Le Clainche, G. Malacrida, E. Durand, G. Pesole, V. Laucou, P. Chatelet, D. Merdinoglu, M. Delledonne, M. Pezzotti, A. Lecharny, C. Scarpelli, F. Artiguenave, M.E. Pe, G. Valle, M. Morgante, M. Caboche, A.F. Adam-Blondon, J. Weissenbach, F. Quetier and P. Wincker, 2007. The grapevine genome sequence suggests ancestral hexaploidization in major angiosperm phyla. *Nature* 449, 463–467.
Jean-Denis J.B., R. Pezet and R. Tabacchi, 2006. Rapid analysis of stilbenes and derivatives from downy mildew-infected grapevine leaves by liquid chromatography-atmospheric pressure photoionisation mass spectrometry. *Journal of Chromatography* 1112, 263–268.
Jeandet P., A.C. Douillet-Breuil, R. Bessis, S. Debord, M.

Sbaghi and M. Adrian, 2002. Phytoalexins from the *Vitaceae*: biosynthesis, phytoalexin gene expression in transgenic plants, antifungal activity and metabolism. *Journal of Agricultural and Food Chemistry* 50, 2731–2741.

Jenny J., 1937. Eine moderne stationäre Spritzanlage mit elektr. Antrieb. *Schweizerische Zeitschrift für Obst- und Weinbau* 46, 229–232.

Jenny J., 1941. Stationäre Spritzanlage im Tessin. *Schweizerische Zeitschrift für Obst- und Weinbau* 1, 477–480.

Jenny J. and H. Huber, 1940. Stationäre Spritzanlagen, namentlich für den Weinbau. *Annuaire Agriculture Suisse* 41, 171–185.

Jermini M., P. Blaise and C. Gessler, 2010a. Response of 'Merlot' (*Vitis vinifera*) grapevine to defoliation caused by downy mildew (*Plasmopara viticola*) during the following growing season. *Vitis* 49, 161–166.

Jermini M., P. Blaise and C. Gessler, 2010b. Influence of *Plasmopara viticola* on gas exchange parameters on field-grown *Vitis vinifera* 'Merlot'. *Vitis* 49, 87–93.

Jermini M., C. Gessler and C. Linder, 2006. The use of know-how on the interaction between grapevine and pests or diseases to improve integrated protection strategies. *Bulletin OILB / SROP* 29, 11, 9–14.

Jermini M., D. Gobbin, P. Blaise and C. Gessler, 2003. Influence of the overwintering methods on the germination dynamic of downy mildew (*Plasmopara viticola*) oospores. *Bulletin OILB / SROP* 26, 8, 37–41.

Jürges G., H.H. Kassemeyer, M. Durrenberger, M. Duggelin and P. Nick, 2009. The mode of interaction between *Vitis* and *Plasmopara viticola* Berk. & Curt. Ex de Bary depends on the host species. *Plant Biology* 11, 886–898.

Kast, W. K., 1998. First indications for a race specific resistance against *Plasmopara viticola*. *Bulletin OILB / SROP,* 21(2), 19–21.

Kast W.K., 2001. Inter-isolate variation of virulence of *Plasmopara viticola* on resistant vine varieties. *Bulletin OILB / SROP* 24(7), 45–49.

Kennelly M.M., D.M. Gadoury, W.F. Wilcox, P.A. Magarey and R.C. Seem, 2005. Seasonal development of ontogenic resistance to downy mildew in grape berries and rachises. *Phytopathology* 95, 1445–1452.

Kennelly M.M., D.M. Gadoury, W.F. Wilcox, P.A. Magarey and R.C. Seem, 2007. Primary infection, lesion productivity and survival of sporangia in the grapevine downy mildew pathogen *Plasmopara viticola*. *Phytopathology* 97, 512–522.

Killigrew B.X., K. Sivasithamparam and E.S. Scott, 2005. Absence of oospores of downy mildew of grape caused by *Plasmopara viticola* as the source of primary inoculum in most Western Australian vineyards. *Plant Disease* 89, Abstract 777.

Klopping H.L. and C.J. Delp, 1980. 2-Cyano-N-[(ethylamino) carbonyl]-2-(methoxyimino)acetamide, a new fungicide. *Journal of Agricultural and Food Chemistry* 28, 467–468.

Koopman T., C.C. Linde, P.H. Fourie and A. McLeod, 2007. Population genetic structure of *Plasmopara viticola* in the Western Cape Province of South Africa. *Molecular Plant Pathology* 8, 723–736.

Kortekamp A., 2006. Expression analysis of defence-related genes in grapevine leaves after inoculation with a host and a non-host pathogen. *Plant Physiology and Biochemistry* 44, 58–67.

Kortekamp A. and E. Zyprian, 2003. Characterization of *Plasmopara*-resistance in grapevine using *in vitro* plants. *Journal of Plant Physiology* 160, 1393–1400.

Kotte W., 1924. Laboratoriumsversuche zur Chemotherapie der Peronosporakrankheit. I. Die Wirkung von Metallen und Salzen. *Zentralblatt für Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene* 61, 367–378.

Kozma P., Jr., 1998. Evaluation of fungus-resistant winegrape varieties. *Acta Horticulturae* 473, 93–103.

Kramer O., 1925. Die Krankheiten und Schädlinge der Reben in Württemberg im Jahre 1924. *Mitteilungen der Deutschen Landwirtschafts-Gesellschaft* 40, 23–26.

Kramer O., 1927. Ueber die Bekämpfung der Rebschädlinge in Württenberg im Jahre 1926. *Nachrichten über Schädlingsbekämpfung* 3, 26–29.

Kuflik T., D. Prodorutti, A. Frizzi, Y. Gafni, S. Simon and I. Pertot, 2009. Optimization of copper treatments in organic viticulture by using a web-based decision support system. *Computers and Electronics in Agriculture* 68, 36–43.

Kump I., C. Gessler and P. Blaise, 1998. Primary infections of *Plasmopara viticola*: should we revise our ideas about the quantitative relevance of oospores? *Bulletin OILB / SROP* 21(2), 9–11.

Kundert J., 1956. Die Peronospora der Rebe und ihre Bekämpfung im Jahre 1955. *Schweizerische Zeitschrift für Obst- und Weinbau* 65, 135–139.

Lacombe J.P., L. Patty and D. Steiger, 2001. La fenamidone: antimildiou de la vigne et de la pomme de terre. *Phytoma* 535, 42–44.

Lafon R., Y. Bugaret and J. Bulit, 1977. Nouvelles perspectives de lutte contre le mildiou de la vigne (*Plasmopara viticola*) (B.C.) (Berlese et de Toni) avec un fongicide systémique, l'éthylphosphite d'aluminium. *Phytiatrie-Phytopharmacie* 26, 19–40.

Lafon R., Y. Bugaret and J. Bulit, 1978. Essais d'un nouveau fongicide de la famille des acylalanines (CGA 48 988) contre le mildiou de la vigne (*Plasmopara viticola* (B.C.) Berl. et de Toni. *Phytiatrie-Phytopharmacie* 27, 263–273.

Lalancette N., L.V. Madden and M.A. Ellis, 1988. A quantitative model for describing the sporulation of *Plasmopara viticola* on grape leaves. *Phytopathology* 78, 1316–1321.

Lamberth C., F. Cederbaum, A. Jeanguenat, H.J. Kempf, M. Zeller and R. Zeun, 2006. Synthesis and fungicidal activity of *N*-2-(3-methoxy-4-propargyloxy)phenethyl amides. Part II: Anti-oomycetic mandelamides. *Pest Management Science* 62, 446–451.

Lamberth C., H.J. Kempf and M. Kriz, 2007. Synthesis and fungicidal activity of *N*-2-(3-methoxy-4-propargyloxy) phenethyl amides. Part 3: Stretched and heterocyclic mandelamide oomyceticides. *Pest Management Science*

63, 57–62.
Latorse M.P., J.M. Gouot and R. Pepin, 1998. Curative activity of RPA407213 against downy mildew on grapevine: biological and microscopical study. In: *Proceedings of an International Conference, Brighton Crop Protection Conference: Pests & Diseases,* 16–19 November 1998. Brighton, UK, Vol. 3, 863–868.
Latorse M.P., D. Holah and R. Bardsley, 2006. Fungicidal properties of fluopicolide-based products. *Pflanzenschutz-Nachrichten Bayer* 59, 185–200.
Laviola C., S. Burruano and S. Strazzeri, 1986. Influenza della temperatura sulla germinazione delle oospore di *Plasmopara viticola* (Berk. et Curt.) Berl. et De Toni. *Phytopathologia Mediterranea* 25, 80–84.
Lippincott J.S., 1866. Observations on the atmospheric humidity. *U.S. Dept. Agricultur Report 1865*, 542–550.
Lizzi Y., C. Coulomb, C. Polian, P.J. Coulomb and P.O. Coulomb, 1998. L'algue face au mildiou: quel avenir? Des résultats de laboratoire très encourageants. *Phytoma* 508, 29–30.
Loskill B., D. Gobbin, B. Berkelmann-Lochnertz and C. Gessler, 2006. Disease dynamics and genetic variability of *Plasmopara viticola* in three untreated vineyards. *Bulletin OILB/SROP* 29, 11, 33–36.
Lupetti Belviglieri R., 1956. Osservazioni sull'impiego degli acuprici in provincia di Verona. Anno 1955. *Rivista di Viticoltura e di Enologia* 9, 77–90.
Lustner G., 1922. Results of the testing of new methods for the control of *Peronospora Oidium and* Vine moth in the years 1920 and 1921. *Berichte hoh. staatliche Lehranstalt Wein-, Obst-, und Gartenbau zu Geisenheim-am-Rhein, 1920-21.*, 79–85.
Lyon A.V., 1924. Problems of the viticultural industry. *Bulletin Australia* 28, 56–72.
Magarey P.A., M.F. Wachtel and M.R. Newton, 1991a. Evaluation of phosphonate, fosetyl-Al and several phenylamide fungicides for post-infection control of grapevine downy mildew caused by *Plasmopara viticola*. *Australasian Plant Pathology* 20, 34–40.
Magarey P.A., M.F. Wachtel, P.C. Weir and R.C. Seem, 1991b. A computer-based simulator for rational management of grapevine downy mildew (*Plasmopara viticola*). *Plant Protection Quarterly* 6, 29–33.
Magarey P.A., T.J. Wicks and M.F. Wachtel, 1990. Phosphonic (phosphorous) acid controls *Plasmopara viticola* the cause of downy mildew of grapevines. *Australasian Plant Pathology* 19, 126–127.
Magnien C., D. Jacquin, N. Muckensturm and P. Guillemard, 1991. MILVIT: a descriptive quantitative model for the asexual phase of grapevine downy mildew. *Bulletin OEPP* 21, 3, 451–459.
Mahoux J., 1933. Le sulfatage avec bouteilles d'air comprime. *Progres Agricole et Viticole* 99, 519–522.
Malquori A. and G. Boezini, 1941. Rame-Bentonite come anticrittogamico. *Bollettino Patologia vegetale* 21, 185–220.
Marais P.G. and A. Tromp, 1981. Metaxanine, a systemic fungicide against *Plasmopara viticola* on wine grapes: disease control, residues and effect on fermentation and wine quality. *South African Journal for Enology and Viticulture* 2, 67–70.
Margot P., F. Huggenberger, J. Amrein and B. Weiss, 1998. CGA 279202: a new broad-spectrum strobilurin fungicide. In: *Proceedings of an International Conference, Brighton Crop Protection Conference: Pests & Diseases,* 16–19 November 1998. Brighton, UK, Vol. 3, 375–382.
Marguerit E., C. Boury, A. Manicki, M. Donnart, G. Butterlin, A. Nemorin, S. Wiedemann-Merdinoglu, D. Merdinoglu, N. Ollat and S. Decroocq, 2009. Genetic dissection of sex determinism, inflorescence morphology and downy mildew resistance in grapevine. *Theoretical and Applied Genetics* 118, 1261–1278.
Martìnez-Zapater J.M., M.J. Carmona, J. Dìaz-Riquelme, L. Fernandez and D. Lijavetzky, 2009. Grapevine genetics after the genome sequence: Challenges and limitations. *Australian Journal of Grape and Wine Research* 16, 33–46.
Masson E., 1887. La Bouille bourguignonne. *Progess Agricole et Viticole*, 511–513.
Matasci C.L., M. Jermini, D. Gobbin and C. Gessler, 2010. Microsatellite based population structure of *Plasmopara viticola* at single vine scale. *European Journal of Plant Pathology* 127, 501–508.
Maurin G., 1983. Application of a model of infection potential to *Plasmopara viticola*. *EPPO Bulletin* 13, 263–269.
McKirdy S.J., I.T. Riley, I.J. Cameron and P.A. Magarey, 1999. First report of grapevine downy mildew (*Plasmopara viticola*) in commercial viticulture in Western Australia. *Plant Disease* 83, 301.
Mercer R.T., G. Lacroix, J.M. Gouot and M.P. Latorse, 1998. RPA 407213: a novel fungicide for the control of downy mildews, late blight and other diseases on a range of crops. In: *Proceedings of an International Conference, Brighton Crop Protection Conference: Pests & Diseases,* 16–19 November 1998. Brighton, UK, Vol. 2, 319–326.
Merdinoglu D., S. Wiedemann-Merdinoglu, P. Coste, V. Dumas, A. Haetty, G. Butterlin and C. Greif, 2003. Genetic analysis of downy mildew resistance derived from *Muscadinia rotundifolia*. *Acta Horticulturae* 603, 451–456.
Merjanian A.S. and M.A.D. Lipetzkaya, 1936. Effect of constant and fluctuating temperatures on the length of the incubation period of downy mildew of the Vine. *Sovetskoya Botanica* 3, 68–77.
Mestbes J.A., 1942. La lucha contra el mildiu. *Agricultura* 11, 139–140.
Meyer A., 1932a. Recherches sur l'utilisation des matières colorantes organiques et de lab-oxyquinoleine dans la lutte contre les maladies cryptogamiques de la Vigne. *Revue de Viticulture* 77, 117–120.
Meyer A., 1932b. Sur l'emploi des colorants et de diverses substances organiques dans la lutte contre les maladies cryptogamiqnes, en particulier le mildiou de la Vigne. *Revue de Viticulture* 76, 197–202.
Millardet A., 1883. Adress to the Society on oospore infection work. *Societée Sciences Bordeaux Bul* 2, 24–27.
Millardet A., 1885. Traitement du mildiou par le mélange de sulphate de cuivre et chaux. *Journal Agriculture Pratique* 49(2), 707–710.

Mitani S., S. Araki, N. Matsuo and P. Camblin, 1998. IKF-916 - a novel systemic fungicide for the control of oomycete plant diseases In: *Proceedings of an International Conference, Brighton Crop Protection Conference: Pests & Diseases,* 16–19 November 1998, Brighton, UK, Vol. 2, 351–358.

Moreira F.M., A. Madini, R. Marino, L. Zulini, M. Stefanini, R. Velasco, P. Kozma and M.S. Grando, 2011. Genetic linkage maps of two interspecific grape crosses (*Vitis* spp.) used to localize quantitative trait loci for downy mildew resistance. *Tree Genetics & Genomes* 7, 153–167.

Morel G., 1946. Essais de laboratoire sur le mildiou de la Vigne. *Revue de Viticulture* 92, 210–213.

Moroldo M., S. Paillard, R. Marconi, L. Fabrice, A. Canaguier, C. Cruaud, V. De Berardinis, C. Guichard, V. Brunaud, I. Le Clainche, S. Scalabrin, R. Testolin, G. Di Gaspero, M. Morgante and A.F. Adam-Blondon, 2008. A physical map of the heterozygous grapevine 'Cabernet Sauvignon' allows mapping candidate genes for disease resistance. *BMC Plant Biology* 8, 66.

Muckensturm N., C. Magnien and D. Jacquin, 1990. MILVIT, un nouveau modèle de prévision des risques mildiou. *Phytoma* 419, 34–36.

Müller K., 1918. *Rebschädlinge und Ihre Neuzeitliche Bekämpfung*. G. Braunsche Hofbuchdruckerei und Verlag, Karlsruhe, Germany, 205 pp.

Müller K., 1930. Phaenologie und Pflanzenschutz im Weinbau. *Weinbau und Kellerwirtschaft* 9, 3 pp.

Müller K., 1937. Ein Vierteljahrhundert Bekämpfung der Reben-Peronospora (*Plasmopara viticola*). *Angewandte Botanik* 19, 110–118.

Müller K., 1938. Entwicklung der Reben-Peronosporabekämpfung in Baden. *Nachrichtenblatt des Deutschen Pflanzenschutzdienstes* 12, 195–205.

Müller K. and H. Sleumer, 1934. Biologische Untersuchungen über die Peronosporakrankheit des Weinstockes. *Landwirtschaftliche Jahrbucher* 79, 509–576.

Müller-Thurgau H., 1911. Die Ansteckung der Weinrebe durch *Plasmopara* (*Peronospora viticola*). *Schweizerische Zeitschrift* für *Obst und Weinbau 20*, 337–344.

Müller-Thurgau H., 1912. Die Bekämpfung der Peronospora auf Grund neuer Forschung. *Mitteilungen des Deutschen Weinbau-Vereins* Sonder-Abdruck, 16 pp.

Müller-Thurgau H., 1922. Zur Bekämpfung der Peronosporakrankheit und des roten Brenners im unbelaubten Zustand der Rebe. *Schweizerische. Zeitschrift für Obst und Weinbau* 5, Separata 6 pp.

Mur G., 1978. Etude du mode d'action des nouveaux fongicides anti-mildiou pénétrants et systémiques. *Progres Agricole et Viticole* 95, 570–585.

Murolo G. and I. Stanich, 1980. Prove di lotta antiperonosporica in Campania con Ridomil. *Vignevini* 7, 31–32.

Musetti R., R. Polizzotto, A. Vecchione, S. Borselli, L. Zulini, M. D'Ambrosio, L.S.D. Toppi and I. Pertot, 2007. Antifungal activity of diketopiperazines extracted from *Alternaria alternata* against *Plasmopara viticola*: an ultrastructural study. *Micron* 38, 643–650.

Musetti R., A. Vecchione, L. Stringher, S. Borselli, L. Zulini, C. Marzani, M. D'Ambrosio, L.S.d. Toppi and I. Pertot, 2006. Inhibition of sporulation and ultrastructural alterations of grapevine downy mildew by the endophytic fungus *Alternaria alternata*. *Phytopathology* 96, 689–698.

Musiani A., 1930. Quando applicare i trattamenti contro la peronospora della Vite. *Istria Agricola* 10, 175–179.

Mzid R., C. Marchive, D. Blancard, L. Deluc, F. Barrieu, M.F. Corio-Costet, N. Drira, S. Hamdi and V. Lauvergeat, 2007. Overexpression of VvWRKY2 in tobacco enhances broad resistance to necrotrophic fungal pathogens. *Physiologia Plantarum* 131, 434–447.

Neyrac, 1930. Expose des traitements contre le mildiou en Charente. *Progres Agricole et Viticole* 94, 613–619.

Northover J. and B.D. Ripley, 1980. Persistence of chlorothalonil on grapes and its effect on disease control and fruit quality. *Journal of Agricultural and Food Chemistry* 28, 971–974.

Olivelli V., 1976. Protezione della vite dalla peronospora con reti antigrandine in plastica impregnate di sali rameici. *Vignevini* 3, 23–27.

Orlandini S., A. Giuntoli and M. Moriondo, 2001. Agrometeorological and ecophysiological approach to the analysis of grapevine response to downy and powdery mildew infections. (Special issue: Grapevine research in Italy.). *Advances in Horticultural Science* 15, 92–96.

Orlandini S., B. Gozzini, M. Rosa, E. Egger, P. Storchi, G. Maracchi and F. Miglietta, 1993. PLASMO: a simulation model for control of *Plasmopara viticola* on grapevine. *Bulletin OEPP* 23, 4, 619–626.

Orlandini S., L. Massetti and A. Dalla Marta, 2008. An agrometeorological approach for the simulation of *Plasmopara viticola*. *Computers and Electronics in Agriculture* 64, 149–161.

Orlandini S., A.d. Marta, I. D'Angelo and R. Genesio, 2003. Application of fuzzy logic for the simulation of *Plasmopara viticola* using agrometeorological variables. *Bulletin OEPP* 33, 415–420.

Osterwalder A., 1939. Prüfung von Peronospora-Bekämpfungsmitteln im Sommer 1938. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 48, 244–249.

Osterwalder A., 1940. Vom Inkubationskalender der Peronospora. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 49, 3–9.

Ovsepyan R.A., 1983. New recommended grape varieties in Armenia. *Vinodelie i Vinogradarstvo SSSR* 2, 40–43.

Pee-Laby E., 1926. L'invasion du mildiou en 1925. Résistance de quelques hybrides producteurs. *Revue de Viticulture* 64, 31–33.

Pellegrini A., D. Prodorutti, A. Frizzi, C. Gessler and I. Pertot, 2010. Development and evaluation of a warning model for the optimal use of copper in organic viticulture. *Journal of Plant Pathology* 92, 43–55.

Perandin G., M. Ruffoni and G. Pasqualin, 1985. Lotta guidata contro la peronospora della vite con l'impiego di fungicidi preventivi e curativi in pieno campo. *Informatore Agrario* 41, 47–53.

Perazzolli M., S. Dagostin, A. Ferrari, Y. Elad and I. Pertot, 2008. Induction of systemic resistance against *Plas-*

mopara viticola in grapevine by *Trichoderma harzianum* T39 and benzothiadiazole. *Biological Control* 47, 228–234.

Perazzolli M., B. Roatti, E. Bozza and I. Pertot, 2011. *Trichoderma harzianum* T39 induces resistance against downy mildew by priming for defence without costs for grapevine. *Biological Control,* DOI:10.1016/j.biocontrol.2011.04.006

Peressotti E., S. Wiedemann-Merdinoglu, F. Delmotte, D. Bellin, G. Di Gaspero, R. Testolin, D. Merdinoglu and P. Mestre, 2010. Breakdown of resistance to grapevine downy mildew upon limited deployment of a resistant variety. *Bmc Plant Biology* 10, 147.

Pertot I. and C. Gessler, 2007. Is it possible to improve biocontrol efficacy in some plant/pathogen systems? *Bulletin OILB/SROP* 30, 6, 3–9.

Pertot I., D. Gobbin and C. Gessler, 2003. Occurrence of *Plasmopara viticola* primary and secondary infections in the early stage of the season in Northern Italy (Trentino). *Bulletin OILB/SROP* 26(8), 47–50.

Pertot I. and L. Zulini, 2003. Studies on *Plasmopara viticola* oospore germination in Trentino, Italy. *Bulletin OILB/SROP* 26(8), 43–46.

Peyer E., 1942. Die Erfahrungen mit schwach konzentrierter Bordeauxbrühe bei der Mehltaubekampfung in den Reben der deutschen Schweiz im Sommer 1941. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 51, 173–178.

Pezet R., K. Gindro, O. Viret and H. Richter, 2004. Effects of resveratrol, viniferins and pterostilbene on *Plasmopara viticola* zoospore mobility and disease development. *Vitis* 43, 145–148.

Pichi P., 1890. Una nuova forma di Peronospora nel peduncolo dei giovani grappoli *Nuova Rassegna di Viticoltura ed Enologia, Conegliano* 15–19.

Pieri G., 1952. Sull' uso del calendario d'incubazione della Peronospora della Vite nella provincia di Treviso. *Annali agricoltura* 6, 1577–1586.

Pieterse C.M.J., A. Leon-Reyes, S. Van der Ent and S.C. Van Wees, 2009. Networking by small-molecule hormones in plant immunity. *Nature Chemical Biology* 5, 308–316.

Piglionica V., P. Tarantini and G. Ferrara, 1977. Lotta contro "muffa grigia", "oidio" e "peronospora" della vite condotta in Puglia. *Informatore Fitopatologico* 27, 17–22.

Polesani M., L. Bortesi, A. Ferrarini, A. Zamboni, M. Fasoli, C. Zadra, A. Lovato, M. Pezzotti, M. Delledonne and A. Polverari, 2010. General and species-specific transcriptional responses to downy mildew infection in a susceptible (*Vitis vinifera*) and a resistant (*V. riparia*) grapevine species. *BMC Genomics* 11, 117.

Polesani M., F. Desario, A. Ferrarini, A. Zamboni, M. Pezzotti, A. Kortekamp and A. Polverari, 2008. cDNA-AFLP analysis of plant and pathogen genes expressed in grapevine infected with *Plasmopara viticola*. *BMC Genomics* 9, 142.

Prillieux E., 1895. *Maladie des Plantes Agricoles et Arbres Fruitiers et Forestiers Causées par des Parasites Végétaux*. 104 Firmin-Didot, Paris, France, 421 pp.

Rafaila C. and Z. David, 1963. Morphological modifications in the sporangia and sporangiophores of *P. viticola* in relation to Vine vars. and environmental conditions. *Lucrarile Gradinii botanice din Bucuresti* 2, 983–992.

Rafaila C., V. Sevcenco and Z. David, 1968. Contributions to the biology of *Plasmopara viticola*. *Phytopathologische Zeitschrift* 63(4), 328–336.

Raucourt M., 1943. Vue d'ensemble sur les essais anticryptogamiques de 1942. *Annales*. *Epiphyt*. 9, 163–167.

Ravaz L., 1927. Chronique: Notes sur le mildiou. *Progres Agricole et Viticole* 87, 429–436.

Ravaz L., 1930. Chronique. Toujours le mildiou. *Progres Agricole et Viticole* 94, 101–105.

Ravaz L. and G. Verge, 1913. La germination des spores d'hiver de *Plasmopara viticola*. *Compte rendues de Academie des Science* 156, 800–802.

Ravaz L. and G. Verge, 1921. Sur la germination des spores du mildiou de la Vigne. *Compte Rendu Hebdomadaire des Seances de l'Academie des Sciences* 173, 1421–1423.

Raynal G., A. Ravise and G. Bompeix, 1980. Action du tris-O-ethylphosphonate d'aluminium (phosethyl d'aluminium) sur la pathogenie de *Plasmopara viticola* et sur la stimulation des reactions de defense de la vigne. *Annales de Phytopathologie* 12, 163–175.

Reuveni M., 1997. Post-infection applications of K_3PO_3, phosphorous acid and dimethomorph inhibit development of downy mildew caused by *Plasmopara viticola* on grapevines. *Journal of Small Fruit & Viticulture* 5, 27–38.

Reuveni M., 2001. Activity of trifloxystrobin against powdery and downy mildew diseases of grapevines. *Canadian Journal of Plant Pathology* 23, 52–59.

Reuveni M., 2003. Activity of the new fungicide benthiavalicarb against *Plasmopara viticola* and its efficacy in controlling downy mildew in grapevines. *European Journal of Plant Pathology* 109, 243–251.

Reuveni M., T. Zahavi and Y. Cohen, 2001. Controlling downy mildew (*Plasmopara viticola*) in field-grown grapevine with beta -aminobutyric acid (BABA). *Phytoparasitica* 29, 125–133.

Rizzotto N. and G. Sartor, 1981. Nuove prospettive di lotta antiperonosporica della vite. *Informatore Agrario* 37, 47–55.

Rossi V., T. Caffi, R. Bugiani, F. Spanna and D. della-Valle, 2008. Estimating the germination dynamics of *Plasmopara viticola* oospores using hydro-thermal time. *Plant Pathology* 57, 216–226.

Rossi V., S. Giosue and T. Caffi, 2009. Modelling the dynamics of infections caused by sexual and asexual spores during *Plasmopara viticola* epidemics. *Journal of Plant Pathology* 91, 615–627.

Ruggiero P. and G. Regiroli 2000. Zoxamide, a novel fungicide for vines and vegetables. In: *Atti Giornate Fitopatologiche*, Vol. 2, 16–20, Aprile 2000, Perugia, Italy, 215–220.

Rumbolz J., S. Wirtz, H.H. Kassemeyer, R. Guggenheim, E. Schafer and C. Buche, 2002. Sporulation of *Plasmopara viticola*: differentiation and light regulation. *Plant Biology* 4, 413–422.

Rumbou A. and C. Gessler, 2004. Genetic dissection of *Plasmopara viticola* population from a Greek vineyard in two consecutive years. *European Journal of Plant Pathology* 110, 379–392.

Rumbou A. and C. Gessler, 2006. Particular structure of *Plasmopara viticola* populations evolved under Greek island conditions. *Phytopathology* 96, 501–509.

Rumbou A. and C. Gessler, 2007. Greek epidemics of grapevine downy mildew are driven by local oosporic inoculum: a population biology approach. *Journal of Biological Research* 7, 3–18.

Salomon R., 1927. La situation viticole. Vignoble de Seine-et Marne. *Revue de Viticulture* 67, 188–190.

Samoucha Y. and U. Gisi, 1987a. Use of two- and three-way mixtures to prevent buildup of resistance to phenylamide fungicides in *Phytophthora* and *Plasmopara*. *Phytopathology* 77, 1405–1409.

Samoucha Y. and U. Gisi, 1987b. Systemicity and persistence of cymoxanil in mixture with oxadixyl and mancozeb against *Phytophthora infestans* and *Plasmopara viticola*. *Crop Protection* 6, 393–398.

Schellenberg H., 1927. Zur Bekämpfung des falschen Mehltaues. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 36, 178–179.

Scherer E. and U. Gisi, 2006. Characterization of genotype and mating type in European isolates of *Plasmopara viticola*. *Journal of Phytopathology* 154, 7/8, 489–495.

Schmidlin L., A. Poutaraud, P. Claudel, P. Mestre, E. Prado, M. Santos-Rosa, S. Wiedemann-Merdinoglu, F. Karst, D. Merdinoglu and P. Hugueney, 2008. A stress-inducible resveratrol O-methyltransferase involved in the biosynthesis of pterostilbene in grapevine. *Plant Physiology* 148, 1630–1639.

Schröter J. 1886. Fam. *Peronosporaceae*. In: *Kryptogamenflora von Schlesien*, Band 3, Heft 3, Pilze J.U. Kern's Verlag, Breslau, Germany, (F. Cohn, ed.), 1885–1889, 814 pp.

Serra S., A. Zanzotto and M. Borgo, 1997. Esperienze di lotta contro la *Peronospora* della vite: verifica sperimentale del modello EPI-*Plasmopara* nel Veneto. *Informatore Fitopatologico* 47, 48–55.

Serres J.M. and G.A. Carraro, 1976. DPX-3217, a new fungicide for the control of grape downy mildew, potato late blight and other *Peronosporales*. *Mededelingen van de Faculteit Landbouwwetenschappen, Rijksuniversiteit Gent* 41, 645–650.

Serviere H., 1940. Chronique. Organisation de la défense contre les attaques du mildiou. *Progres Agricole et Viticole* 113, 197–199.

Siegfried W. and T. Temperli, 2008. PIWI-Reben im Vergleich - ein Zwischenbericht. *Obst- und Weinbau* 144, 6–9.

Siegfried, W., O. Viret, B. Huber, R. Wohlhauser, 2007. Dosage of plant protection products adapted to leaf area index in viticulture. *Crop Protection* 26, 2, 73–82.

Slaughter A.R., M.M. Hamiduzzaman, K. Gindro, J.M. Neuhaus and B. Mauch-Mani, 2008. Beta-aminobutyric acid-induced resistance in grapevine against downy mildew: involvement of pterostilbene. (Special Issue: The downy mildews - genetics, molecular biology and control). *European Journal of Plant Pathology* 122, 185-195.

Sohad C., 1943. Etude des lacteurs de 1'inflection primaire et de la duree de 1'incubation en vue de la prevision des epoques de traitements contre to mildiou de la Vigne. *Annales Epiphyt.* 9, 19–25.

Speiser B., A. Berner, A. Haseli and L. Tamm, 1999. Control of downy mildew of grapevine with potassium phosphonate: effectivity and phosphonate residues in wine. *Biological Agriculture & Horticulture* 17, 305–312.

Spera G., R. Bugliosi, A.l. Torre and M. Gianferro, 2009. Results of a two years trials of a new forecasting model for *Plasmopara viticola* infection realized with an artificial neural network. *Progres Agricole et Viticole* 126, 40–44.

Spring J.I., M. Jermini, D. Maigre and F. Murisier, 1998. Regent, un nouveau cépage résistant aux maladies. Expériences en Suisse romande et au Tessin. *Revue Suisse de Viticulture, d'Arboriculture et d'Horticulture* 30, 347–351.

Spring J.L., 2001. Premières expériences avec les cépages interspécifiques Merzling, Johanniter, Bronner et Solaris en Suisse romande. *Revue Suisse de Viticulture, Arboriculture et Horticulture* 33, 57–64.

Spring J.L., 2003. Expérimentation des cépages interspécifiques d'origine hongroise Bianca, Lilla et Nero en Suisse romande. *Revue Suisse de Viticulture, Arboriculture et Horticulture* 35, 159–164.

Staudt G. and H.H. Kassemeyer, 1995. Evaluation of downy mildew resistance in various accessions of wild *Vitis* species. *Vitis* 34, 225–228.

Stellwaag F., 1943. Stand und Krisis der Schädlingsbekämpfung im Weinbau. *Zeitschrift* für *Pflanzenkrankheiten, Pflanzenpathologie und Pflanzenschutz* 53, 113–124.

Stenzel K., R. Pontzen, T. Seitz, R. Tiemann and A. Witzenberger, 1998. SZX 722: a novel systemic oomycete fungicide. *Brighton Crop Protection Conference: Pests & Diseases*, Vol. 2, 367–374.

Stiegler A., 1923. Der echte Meltau (*Oidium tuckeri*) und der falsche Mehltau (*Peronospora viticola*) sowie deren Bekämpfung. *Allgemeine Weinzeitschrift* 40, 51–52

Stubler D., U. Reckmann and G. Noga, 1999. Systemic action of iprovalicarb (SZX 0722) in grapevines. *Pflanzenschutz-Nachrichten Bayer* 52, 33–48.

Sung C.R.M. and M. Clerjeau, 1988. Techniques for formation, maturation and germination of *Plasmopara viticola* oospores under controlled conditions. *Plant Disease* 72, 938–941.

Sung C.R.T.M. and M. Clerjeau, 1989. Influence of climatic conditions in the autumn on the maturation of oospores of *Plasmopara viticola* and the degree of attack of mildew in the spring: models of the phenomenon Influence of environmental factors on the control of grape pests,

diseases and weeds. *Proceedings of a Meeting of the EC Expert's Group, Thessaloniki*, 6–8 Oct. 1987, 237–243.

Thuerig B., A. Binder, T. Boller, U. Guyer, S. Jimenez, C. Rentsch and L. Tamm, 2006. An aqueous extract of the dry mycelium of *Penicillium chrysogenum* induces resistance in several crops under controlled and field conditions. *European Journal of Plant Pathology* 114, 185–197.

Thuerig B., A. Slaughter, E. Marouf, M. Held, B. Mauch-Mani and L. Tamm, 2010. Site-specific field resistance of grapevine to *Plasmopara viticola* correlates to altered gene expression and was not modulated by the application of organic amendments. *European Journal of Plant Pathology* 129, 255–265.

Tilcher R., G.A. Wolf and G. Brendel, 1994. Effects of microbial antagonists on leaf infestation, sporangia germination and zoospore behaviour of *Plasmopara viticola* (Berk. & Curtis) Berl. & de Toni. *Mededelingen - Faculteit Landbouwkundige en Toegepaste Biologische Wetenschappen, Universiteit Gent* 59, 919–929.

Toffolatti S.L., G. Rho, P. Fremiot, M. Prandato, A. Vavassori, F. Zerbetto and A. Vercesi, 2006. Dinamica di germinazione delle oospore di *Plasmopara viticola* e comparsa delle infezioni primarie. In: *Atti Giornate Fitopatologiche,* Vol. 2, 145–152.

Toffolatti S.L., L. Serrati, H. Sierotzki, U. Gisi and A. Vercesi, 2007. Assessment of QoI resistance in *Plasmopara viticola* oospores. *Pest Management Science* 63, 194–201.

Topfer R. and R. Eibach, 2002. Rebenzüchtung für eine okologischere Weinbergsbewirtschaftung. *Beitrage zur* Züchtungsforschung - *Bundesanstalt* für Züchtungsforschung *an Kulturpflanzen* 8, 55–62.

Tran Manh Sung C., S. Strizyk and M. Clerjeau, 1990. Simulation of the date of maturity of *Plasmopara viticola* oospores to predict the severity of primary infections in grapevine. *Plant Disease* 74, 120–124.

Trouvelot S., A.L. Varnier, M. Allègre, L. Mercier, F. Baillieul, C. Arnould, V. Gianinazzi-Pearson, O. Klarzynski, J.M. Joubert, A. Pugin and X. Daire, 2008a. A beta-1,3 glucan sulfate induces resistance in grapevine against *Plasmopara viticola* through priming of defense responses, including HR-like cell death. *Molecular Plant-Microbe Interactions* 21, 232–243.

Trouvelot S., A.L. Varnier, M. Allègre, L. Mercier, F. Baillieul, C. Arnould, V. Gianinazzi-Pearson, O. Klarzynski, J.M. Joubert, A. Pugin and X. Daire, 2008b. A beta -1,3 glucan sulfate induces resistance in grapevine against *Plasmopara viticola* through priming of defense responses, including HR-like cell death. *Molecular Plant-Microbe Interactions* 21, 232–243.

Tucci M., M. Ruocco, L. De Masi, M. De Palma and M. Lorito, 2010. The beneficial effect of *Trichoderma* spp. on tomato is modulated by the plant genotype. *Molecular Plant Pathology* DOI: 10.1111/j.1364-3703.2010.00674.x.

Unger S., C. Buche, S. Boso and H.H. Kassemeyer, 2007. The course of colonization of two different *Vitis* genotypes by *Plasmopara viticola* indicates compatible and incompatible host-pathogen interactions. *Phytopathology* 97, 780–786.

Urech P.A. and F.I. Schwinn, 1978. Properties of a new chemical family active against *Peronosporales* and of CGA 48 988 in particular. *Phytiatrie-Phytopharmacie* 27, 239–247.

Van der Auwera G.D.B.R., Van de Peer Y, P. De Rijk, I. Van den Broeck and R. De Wachter, 1995. The phylogeny of the *Hyphochytriomycota* as deduced from ribosomal RNA sequences of *Hyphochytrium catenoides*. *Molecular Biology and Evolution* 12, 671–678.

van Hulten M., M. Pelser, L.C. van Loon, C.M.J. Pieterse and J. Ton, 2006. Costs and benefits of priming for defense in *Arabidopsis*. *Proceedings of the National Academy of Sciences of the United States of America* 103, 5602–5607.

Vecchione A., L. Zulini, I. Pertot and R. Musetti, 2007. Biological control of *Plasmopara viticola*: a multisite approach. *Acta Horticulturae* 754, 361–366.

Velasco R., A. Zharkikh, M. Troggio, D.A. Cartwright, A. Cestaro, D. Pruss, M. Pindo, L.M. Fitzgerald, S. Vezzulli, J. Reid, G. Malacarne, D. Iliev, G. Coppola, B. Wardell, D. Micheletti, T. Macalma, M. Facci, J.T. Mitchell, M. Perazzolli, G. Eldredge, P. Gatto, R. Oyzerski, M. Moretto, N. Gutin, M. Stefanini, Y. Chen, C. Segala, C. Davenport, L. Dematte, A. Mraz, J. Battilana, K. Stormo, F. Costa, Q. Tao, A. Si-Ammour, T. Harkins, A. Lackey, C. Perbost, B. Taillon, A. Stella, V. Solovyev, J.A. Fawcett, L. Sterck, K. Vandepoele, S.M. Grando, S. Toppo, C. Moser, J. Lanchbury, R. Bogden, M. Skolnick, V. Sgaramella, S.K. Bhatnagar, P. Fontana, A. Gutin, Y. van de Peer, F. Salamini and R. Viola, 2007. A high quality draft consensus sequence of the genome of a heterozygous grapevine variety. *Plos One* 2, 1326.

Vercesi A., F. Zerbetto and G. Rho, 1999. Impiego dei modelli EPI e PRO nella difesa antiperonosporica del vigneto. *Frustula Entomologica* 22, 92–97.

Verhagen B.W.M., J. Glazebrook, T. Zhu, H.S. Chang, L.C. van Loon and C.M.J. Pieterse, 2004. The transcriptome of rhizobacteria-induced systemic resistance in *Arabidopsis*. *Molecular Plant-Microbe Interactions* 17, 895–908.

Vial J., J. Trespeuch and M. Sylvestre, 1978. Field trials of CGA 48 988 (Acylon) for the control of grapevine mildew. *Phytiatrie-Phytopharmacie* 27, 249–261.

Villedieu G., 1932. Cuivre et mildiou. *Progres Agricole et Viticole* 98, 536–539.

Viret O., B. Bloesch, A.L. Fabre, W. Siegfried, G. Bleyer, B. Huber, H.H. Kassemeyer and V. Steinmetz, 2005. Vitimeteo: un nouveau modèle de prévision pour le mildiou de la vigne (www.agrometeo.ch). *Revue Suisse de Viticulture, Arboriculture et Horticulture* 37, 65–68.

Viret, O., W. Siegfried, B. Bloesch, 2008. *Dosage of fungicides adapted to leaf area index in viticulture in Switzerland*. Progres Agricole et Viticole 125, 20, 549–551.

Voglino R., 1922. Servizio di segnalazione degli attacchi di *Plasmopora viticola* nel 1921 nelle Province di Torino, Cuneo, Novara. *Nuovi Annali del Ministero per l'Agricoltura* 2, 72–80.

Voitovich K. A. and A. N. Nikolaev, 1970. On the presence

of physiologic races of *P. viticola* in the USSR. *Mikologiya i Fitopatologiya* 4, 4, 281–285.
Walters D.R. and M. Heil, 2007. Costs and trade-offs associated with induced resistance. *Physiological and Molecular Plant Pathology* 71, 3–17.
Welter L.J., N. Gokturk-Baydar, M. Akkurt, E. Maul, R. Eibach, R. Topfer and E.M. Zyprian, 2007. Genetic mapping and localization of quantitative trait loci affecting fungal disease resistance and leaf morphology in grapevine (*Vitis vinifera* L). *Molecular Breeding* 20, 359–374.
Wicks T., 1980. The control of *Plasmopara viticola* by fungicides applied after infection. *Australasian Plant Pathology* 9, 2–3.
Wicks T. and B. Hall, 1990. Efficacy of dimethomorph (CME 151) against downy mildew of grapevines. *Plant Disease* 74, 114–116.
Wicks T. and T.C. Lee, 1982. Evaluation of fungicides applied after infection for control of *Plasmopara viticola* on grapevine. *Plant Disease* 66, 839–841.
Wicks T.J., P.A. Magarey, M.F. Wachtel and A.B. Frensham, 1991. Effect of postinfection application of phosphorous (phosphonic) acid on the incidence and sporulation of *Plasmopara viticola* on grapevine. *Plant Disease* 75, 40–43.
Wille F., 1927. Untersuchungen über die Beziehungen zwischen Immunität und Reaktion des Zellsaftes. *Zeitschrift für Pflanzenkrankheiten und Pflanzenschutz*. 37, 129–158.
Wissotzky S., 2007. Mildiou de la vigne: éléments de prévision et de gestion. *Progres Agricole et Viticole* 124, 173–179.
Wong F.P. and W.F. Wilcox, 2000. Distribution of baseline sensitivities to azoxystrobin among isolates of *Plasmopara viticola*. *Plant Disease* 84, 275–281.
Wu J., Y. Zhang, H. Zhang, H. Huang, K.M. Folta and J. Lu, 2010. Whole genome wide expression profiles of *Vitis amurensis* grape responding to downy mildew by using Solexa sequencing technology. *Bmc Plant Biology* 10, 234.
Zachos D.G., 1959. Recherches sur la biologie et l'épidémiologie du mildiou de la Vigne en Grèce. Bases de prévisions et d 'avertissements. *Annales de l'Institut Phytopathologique Benaki* 2, 193–355.
Zillig H. and L. Niemeyer, 1942. Wie spart man Kupfer bei der Plasmoparabekämpfung ? *Wein und Rebe* 24, 25–47.
Zobrist L., 1954. L'efficacité des fongicides organiques en viticulture. *Proceedings III int. Congress on Phyto-pharmacologie*, Vol. 2. 682–688.
Zobrist L., R. Clausen and P. Mühlethaler, 1953a. Rebspritzungen mit dem Helikopter - vom Versuch zur praktischen Anwendung. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 62, 283–287.
Zobrist L., R. Clausen and P. Mühlethaler, 1953b. Rebspritzung mit Helikopter- Der erste Versuch in der Schweiz. *Schweizerische Zeitschrift* für *Obst- und Weinbau* 62, 246–248.
Zyprian E., L.J. Welter, M. Akkurt, S. Ebert, I. Salakhutdinov, N. Gokturk-Baydar, R. Eibach and R. Topfer, 2009. Genetic analysis of fungal disease resistance in grapevine. *Acta Horticulturae* 827, 535–538.

Accepted for publication: March 21, 2011