VITIS VINIFERA CANES, A SOURCE OF STILBENOIDS AGAINST DOWNY MILDEW

Tristan RICHARD[1], Assia ABDELLI-BELHADJ[2], Xavier VITRAC[2],
Pierre WAFFO TEGUO[1], Jean-Michel MÉRILLON[1, 2*]

1: Université de Bordeaux, Unité de Recherche Œnologie EA 4577, USC 1366 INRA, INP
Equipe Molécules d'Intérêt Biologique (Gesvab) - Institut des Sciences de la Vigne et du Vin - CS 50008
210, chemin de Leysotte 33882 Villenave d'Ornon Cedex, France
2: Polyphénols Biotech, Institut des Sciences de la Vigne et du Vin - CS 50008
210, chemin de Leysotte 33882 Villenave d'Ornon Cedex, France

Abstract

Aim: To investigate the antifungal efficacy of grape cane extracts enriched in stilbenes against *Plasmopara viticola* by *in vivo* experiments on grape plants.

Methods and results: Experiments on grape plants were carried out from 2007 to 2008 at commercial vineyards in « Le Tourne » (southwest region of France) on *Vitis vinifera* L. cv Merlot plants and in a greenhouse, where 45-day-old plants (Merlot) were used. Grapevine plants were treated with stilbene extract (STE) or « Bordeaux mixture » (BM) as positive control. STE, prepared from *Vitis vinifera* canes, contained 33% of total stilbenes ranging from monomers to tetramers and was used at 5g L^{-1}. The degree of protection for downy mildew was determined as the percentage of infected leaf surface area and pathogen attack frequency. On young plants in greenhouse, *P. viticola* inoculation and three treatments with STE were carried out. After the treatments, the disease reduction was from 59 to 69% for pathogen attack frequency and from 83 to 88% for infected leaf surface for the two modes of infestation (artificial and secondary). STE triggered a similar intense decrease in downy mildew infection as recorded for BM. In the vineyard assay, downy mildew infection was allowed to occur naturally. Field grown plants were treated every 7th day for eight weeks or every 14th day for eight weeks during June and July. We observed that STE was also able to prevent the infection of *P. viticola* in vineyard but to a lesser extent. Treatment with STE every 7th day provided better results in terms of infection frequency and leaf area than that every 14th day. STE treatment of plants every 7th day in vineyard reduced the frequency of attack by 16 to 39% and leaf surface infection by 57 to 61%, which was lower than that recorded for BM. STE also displayed *in vitro* inhibitory effects on the release of zoospores from sporangia and the germination of zoospores of *P. viticola*. Zoospore germination was completely inhibited at a concentration of 100 mg L^{-1} and zoospore release was inhibited at 330 mg L^{-1}.

Conclusions: Treatment of young grapevine plants in greenhouse with stilbene extract from grapevine canes offered considerable protection against *P. viticola* that was identical to that provided by Bordeaux mixture. STE was also able to prevent the infection of *P. viticola* in vineyard but to a lesser extent. Treatment with STE every 7th day in vineyard provided better results than that every 14th day.

Significance and impact of the study: Stilbenes from grapevine canes are good candidates as natural fungicides against downy mildew. Given the very large quantity of grape canes available each year, this strategy could be scaled up to control this grapevine disease in a sustainable manner.

Key words: natural antifungal compounds, cane extract, stilbenes, downy mildew, grape plants, *Plasmopara viticola, Vitis vinifera* L.

manuscript received 24 February 2016 - revised manuscript received 31st May 2016

*Corresponding author : jean-michel.merillon@u-bordeaux.fr

OENO One, 2016, **50**, 3, 137-143
©Université de Bordeaux (Bordeaux, France)

INTRODUCTION

Downy mildew caused by an oomycete, *Plasmopara viticola*, is one of the most destructive diseases in vineyards. Protection against it involves several applications of fungicides between budburst/sprout and fruit-ripening depending on the disease severity and the climatic conditions of the year (Schnee *et al.*, 2013). These pesticides have a harmful impact on the environment as well as on human health. Even copper, a natural micronutrient used in organic farming against downy mildew, may have long-term harmful consequences owing to its accumulation in the soil (La Torre *et al.*, 2011). Thus, developing alternative strategies such as the use of natural products from plants that are biodegradable and usually less toxic has become a pressing need. Indeed, many secondary metabolites synthesized by plants to defend themselves against pathogens have antimicrobial properties. The alternative approach is to find elicitor compounds for grapevine defense stimulation, but only a few commercial products with low efficacy have been approved for this purpose (Aziz *et al.*, 2003; Saigne-Soulard *et al.*, 2015).

Stilbenes are a class of phenolic compounds that are abundant in Vitaceae, particularly in grapevines. Over 60 stilbenes have been identified in *Vitis vinifera* to date, presenting a wide structural diversity from monomers to hexamers, with resveratrol as the predominant monomer subunit (Pawlus *et al.*, 2012; Rivière *et al.*, 2012). Several studies have shown the major involvement of these compounds in grapevine defense by acting as phytoalexins, mainly in leaves and berries (Douillet-Breuil *et al.*, 1999; Adrian *et al.*, 2000; Pezet *et al.*, 2003). They can also be constitutively accumulated in stems and roots (Lambert *et al.*, 2013). For example, a positive correlation has been shown between the level of stilbenes such as resveratrol, pterostilbene and viniferins in grapevine leaves and berries and resistance to diseases such as gray mold, powdery mildew and downy mildew (Langcake, 1981; Bavaresco *et al.*, 1997; Aziz *et al.*, 2003; Pezet *et al.*, 2004; Belhadj *et al.*, 2006; Dufour *et al.*, 2013). Alonso-Villaverde *et al.* (2011) showed that resistant grapevines react quickly to *P. viticola* infection by producing large concentrations of stilbenes at the exact sites of infection. Moreover, for the same cultivar, *V. vinifera* Cabernet Franc, some clones with a lower susceptibility for downy mildew produce a higher level of stilbenes in leaves (Van Leeuwen *et al.*, 2013). Additionally, the antimicrobial activity of various stilbenes toward zoospore mobility and *P. viticola* sporulation has been assayed *in vitro*. The stilbenes exhibiting the most toxic activity were δ- and ε-viniferins (dimers), vitisin B and hopeaphenol (tetramers), and methylated derivatives of resveratrol (pterostilbene and analogues) (Pezet *et al.*, 2004; Schnee *et al.*, 2013; Chalal *et al.*, 2014).

Stilbenes are constitutively accumulated in grape canes at the high level of about 10 g $kgDW^{-1}$ (Houillé *et al.*, 2015). Pruning results in the production each year of a very large quantity of grape canes. For example, in France, 1.4 million tons of dry weight are available per year (FranceAgrimer, 2012). Therefore, this strategy could be used to control grapevine diseases by using stilbenes extracted from these grape canes.

The aim of this study was to investigate the antifungal activities of stilbene-rich grape cane extracts against *P. viticola* by conducting two *in vitro* tests (zoospore germination and sporulation) and two *in vivo* experiments on plants (in greenhouse and in vineyard).

MATERIALS AND METHODS

1. Experimental sites and plant material

Experiments on grape plants were carried out in 2007 and 2008. Field trials were performed in commercial vineyards in « Le Tourne » (southwest region of France) on healthy *Vitis vinifera* L. cv Merlot plants. The grapevines (planted in 1970) were trained in a double Guyot system (two canes in opposite direction and two spurs) with a planting density of 2777 plants ha^{-1}. Greenhouse studies (SciencesAgro Atlantique, Loupes, France) were conducted on 45-day-old plants (Merlot) with 5 to 7 leaves.

2. Reagents

Stilbenoid extract (STE), named Vineatrol®, was purchased from Actichem (Montauban, France). It was prepared from *Vitis vinifera* canes collected in vineyards from the Bordeaux region and was used at a dose of 5 g L^{-1}. It contained 33% of total stilbenes, mainly ampelopsin A (2.3%), piceatannol (0.4%), pallidol (1.4%), resveratrol (5.8%), hopeaphenol (1.3%), isohopeaphenol (3.6%), ε-viniferin (12.3%), miyabenol C (2.0%), ω-viniferin (1.7%) and vitisin B (1.9%). These compounds were determined by HPLC-DAD-MS (Fig. 1) and calculated from a calibration curve of their own standards purified and identified in the lab (Lambert *et al.*, 2013; Pawlus *et al.*, 2013).

« Bordeaux mixture » RSR Disperss (BM) from UPL (Courbevoie, France) containing 20% of copper sulfate was used as cupric formulation at a dose of 7.5 g L^{-1}.

©Université de Bordeaux (Bordeaux, France)

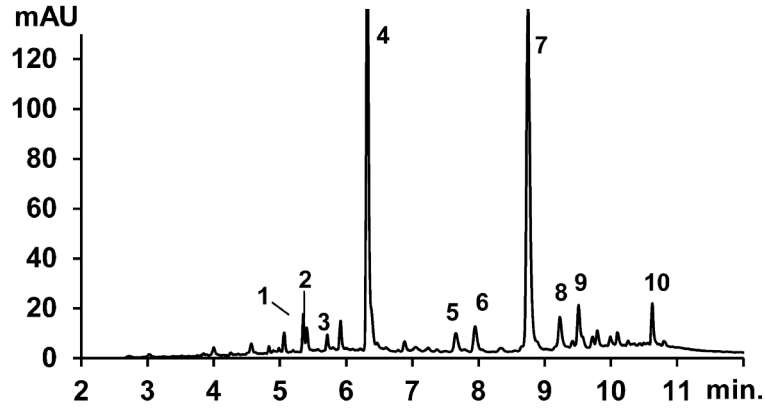

Figure 1 – HPLC chromatogram obtained at 280 nm of stilbene extract (STE) using HPLC-DAD-MS. (1) ampelopsin A, (2) piceatannol, (3) pallidol, (4) resveratrol, (5) hopeaphenol, (6) isohopeaphenol, (7) ε-viniferin, (8) miyabenol C, (9) ω-viniferin, (10) vitisin B.

3. Treatments in greenhouse

Six plants per treatment were sprayed entirely with the test material and each experiment was replicated in three sets (T1 = 17/10/2008). Lower surfaces of leaves of two plants were inoculated three days after the first treatment by spraying freshly prepared sporangia of *P. viticola* (40 000 mL^{-1}). The second treatment was given 14 days later (T2 = 31/10/2008) followed by the third 12 days later (T3 = 12/11/2008). For the four plants without artificial infection, natural secondary infestation was obtained by overhead irrigation three days after the second and third treatments (T2, T3).

Disease intensity was evaluated visually by estimating percentage of infected leaf surface and pathogen attack frequency on the following dates: 04/11 for the two plants artificially infected; 14/11, 21/11, and 26/11/2008 for the four plants with secondary infestation. The application volume was about 600 L ha^{-1}. The control plants remained untreated.

4. Treatments in vineyard

In this experiment, downy mildew infestation was natural. Three plants per treatment were sprayed entirely and each experiment was repeated in quadruplicate (T'1 = 07/06/2007). Afterwards, seven other treatments were performed every 7 days or three other treatments every 14 days. Disease intensity was evaluated visually by estimating the percentage of infected leaf surface and pathogen attack frequency on the following dates: 16/07, 23/07, and 01/08/2007. The application volume was about 600 L ha^{-1}. The control plants remained untreated with reagents.

Before the experiment, all plants were protected by using Dithane Neotec (3.5 kg ha^{-1}; 24/04/2007, 04/05/2007), Option Flash (4 kg ha^{-1}; 15/05/2007, 29/05/2007), Thiovit Jet (12.5 kg ha^{-1}; 15/05/2007), Olymp (0.3 L ha^{-1}; 29/05/2007) and Legend (0.2 L ha^{-1}; 22/06/2007, 04/07/2007).

5. Stilbenoid extract activity on release of zoospores from sporangia and germination of *P. viticola* zoospores

STE was tested on sporangia (50 000 mL^{-1}) using microplates at 21°C in the dark. After six hours, cotton blue was added to the wells and the numbers of germinated zoospores (viable) and empty sporangia (representing the release of zoospores) were determined using a microscope. STE was tested at different final concentrations, N (1 g L^{-1}), N/3, N/10, N/30 and N/100, using acetone to ensure dissolution at a concentration of 1% (v/v). The experiment was repeated three times.

6. Statistical analysis

Statistical analyses were performed using the Newman-Keuls test and the Student t-test at $P \leq 0.05$.

RESULTS

1. Protection of grapevine plants by stilbenoid extract (STE) against downy mildew

On young plants in greenhouse. Grapevine plants pretreated with STE or BM as positive control three days before *P. viticola* inoculation, followed by two

OENO One, 2016, **50**, 3, 137-143
©Université de Bordeaux (Bordeaux, France)

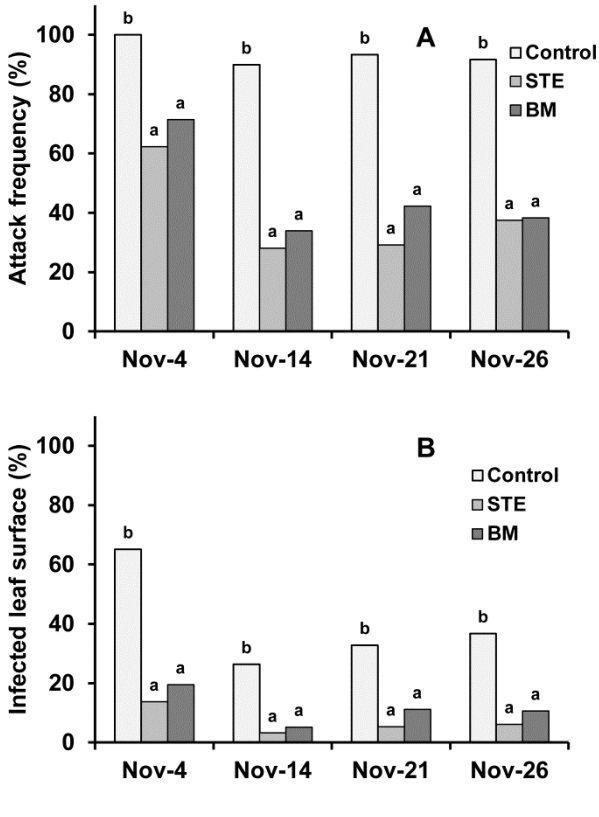

Figure 2 – Protection of grapevine plants (Merlot) by stilbenoid extract (STE) treatment against *P. viticola* in greenhouse: A. Pathogen attack frequency (%) and B. Infected leaf surface (%).
STE or Bordeaux mixture (BM) was sprayed on plants at a concentration of 5 g L^{-1} and 7.5 g L^{-1} respectively on the following dates: 17/10, 31/10 and 12/11/2008. Observation Nov 4 concerns two plants with artificial infestation three days after the first treatment; observations Nov 14, Nov 21 and Nov 26 concern four plants with secondary infestation three days after the second and third treatments. Letters above columns indicate significant differences at $P \leq 0.05$.

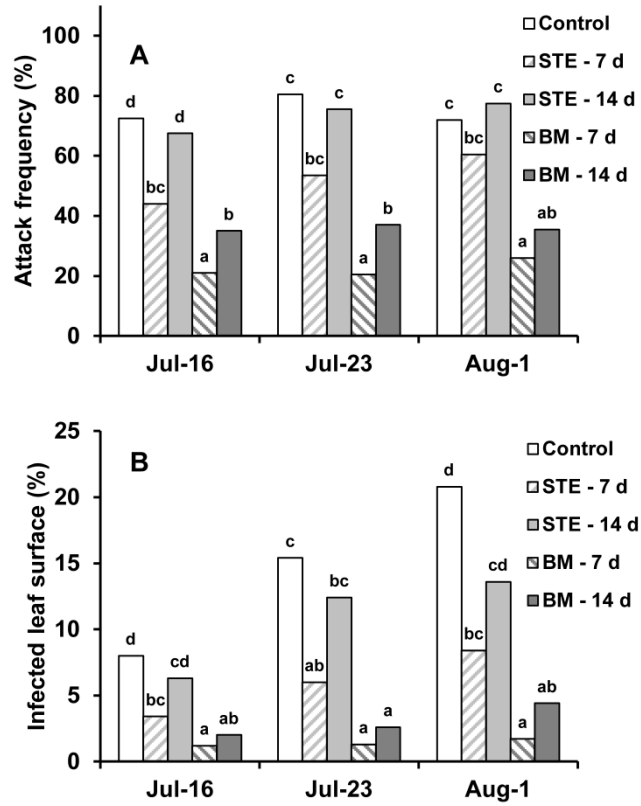

Figure 3 – Protection of grapevine plants (Merlot) by stilbenoid extract (STE) treatment against *P. viticola* (natural infestation) in vineyard: A. Pathogen attack frequency (%) and B. Infected leaf surface (%).
STE or Bordeaux mixture (BM) was sprayed on plants at a concentration of 5 g L^{-1} and 7.5 g L^{-1} respectively. From the first treatment (07/06/2007), seven other treatments were performed every 7 days or three treatments every 14 days. Letters above columns indicate significant differences at $P \leq 0.05$.

subsequent treatments spaced 12-14 days apart, showed no phytotoxic effect. The degree of protection against downy mildew was recorded as the percentage of infected leaf surface area and pathogen attack frequency. The first observation (Nov 4) of two plants 15 days after inoculation (artificial infestation), i.e. 18 days after the first treatment and four days after the second treatment (Fig. 2A and 2B), indicated a 38% reduction in pathogen attack frequency and a 79% reduction in infected leaf surface area. After the third treatment (Nov 12), three observations (Nov 14, 21 and 26) of four plants with secondary infestation showed a 59 to 69% reduction in pathogen attack frequency and a 83 to 88% reduction in infected leaf surface area. On young plants in greenhouse, STE triggered a similar intense decrease in downy mildew to that observed for BM (Fig. 2A and 2B).

In vineyard. In vineyard assays, downy mildew infection was allowed to occur naturally. Grapevine plants treated with STE or BM from June 7 to July 26 (treatments every 7th day) or to July 19 (treatments every 14th day) showed no phytotoxic effect. The pathogen attack frequency was similar in control plants (~75%) throughout the observation period (Fig. 3A). Concerning the treatment with STE at the 7th day, a low reduction of 39% was observed in attack frequency for the observation of Jul 16, 34% on Jul 23 and 16% on Aug 1. No significant decrease was observed for treatment with STE at the 14th day.

©Université de Bordeaux (Bordeaux, France)

Table 1 – Effects of stilbenoid extract (STE) on release of zoospores from sporangia and germination of *P. viticola* zoospores. Number of germinated zoospores and empty sporangia expressed as % (± SD). STE was tested at N (1 g L^{-1}), N/3, N/10, N/30 and N/100. (Means of three experiments ± SD).

10	Control		STE				
	water	acetone	N/100	N/30	N/10	N/3	N
Empty sporangia (%)	99.7±0.1	99.1±0.1	95.1±2.5	47.9±2.3	25.4±3.3	1.8±0.6	0
Germinated zoospores (%)	99.9±0.1	99.7±0.3	99.0±0.8	64.9±1.6	0	0	0

However, BM treatment induced a reduction in attack frequency of 64-74% (treatment every 7th day) and 51-54% (treatment every 14th day).

With regard to infected leaf surface area (Fig. 3B), downy mildew progressed constantly in control plants from 8 to 21% between July 16 and August 1. In contrast, in grapevines treated with STE every 7th day, the percentage of leaf surface infection by *P. viticola* progressed only from 3.4 to 8.4%, which represents a significant reduction of 57-61% for the three observations with this treatment. The reduction was 21-35% for treatment with STE at the 14th day. With BM, reductions of 85-92% (treatment every 7th day) and 75-83% (treatment every 14th day) were obtained. In vineyard, STE triggered a less intense decrease in downy mildew than that observed with BM (Fig. 3A and 3B).

2. Inhibitory effects of stilbenoid extract (STE) on release of zoospores from sporangia and germination of *P. viticola* zoospores

To investigate the mode of action of the stilbenoid extract, we studied various concentrations of STE from 10 mg L^{-1} to 1 g L^{-1} on the release of zoospores from sporangia and the germination of zoospores (Table 1). STE displayed a significant activity on both modalities from the concentration of N/30, which is equivalent to 33 mg L^{-1}. Zoospore germination was completely inhibited at the concentration of 100 mg L^{-1} and zoospore release was completely inhibited at 330 mg L^{-1}.

DISCUSSION

Treatment of young grapevine plants in greenhouse with STE led to considerable protection against *P. viticola* identical to that offered by Bordeaux mixture. After the treatments with STE, the disease reduction was from 59 to 69% for pathogen attack frequency and from 83 to 88% for infected leaf surface for the two modes of infestation (artificial and secondary). STE was also able to prevent the infection of *P. viticola* in vineyard but to a lesser extent. Treatment with STE every 7th day led to better results than treating every 14th day. STE treatment of plants every 7th day in vineyard reduced attack frequency by 16 to 39% and leaf surface infection by 57 to 61%, which was lower than the values obtained with BM.

For these experiments, we used the concentration of 5 g L^{-1} on the basis of preliminary results obtained after a single treatment on young plants that showed a reduction in downy mildew correlated with STE concentration from 1 g L^{-1} (data not shown). No disease reduction was observed at a concentration lower than this concentration, although inhibition of zoospore germination was complete at a concentration of 100 mg L^{-1} and that of zoospore release at 330 mg L^{-1}. The chemical characterization of STE showed that it contains a variety of stilbenoids including monomers (resveratrol, piceatannol), dimers (ε- and ω-viniferins, ampelopsin A, pallidol), a trimer (miyabenol C), and tetramers (vitisin B, isohopeaphenol, hopeaphenol). Using *in vitro* antifungal assays, Schnee *et al.* (2013) also observed the inhibition of sporulation and the mobility of *P. viticola* zoospores with a methanolic extract of grapevine canes at a concentration of 1 g L^{-1}. To identify the stilbenes responsible for the antifungal activity, they fractionated the extract by solid phase extraction and obtained four fractions. All of them exhibited an activity against *P. viticola* at a concentration of 1 g L^{-1}, which indicated that many of these compounds were involved in the antifungal activity. A few compounds appeared particularly active in *in*

OENO One, 2016, **50**, 3, 137-143
©Université de Bordeaux (Bordeaux, France)

vitro tests against *P. viticola* with IC_{50} values between 10 and 100 µM, such as hopeaphenol, vitisin B, δ- and ε-viniferins; trans-resveratrol had a moderate activity with an IC value of 120-200 µM (Pezet *et al.*, 2004; Schnee *et al.*, 2013). Pterostilbene has been found to be the most toxic stilbene against various fungi, in particular *P. viticola* (Pezet *et al.*, 2004; Chalal *et al.*, 2014). However, to our knowledge, this has never been shown in grapevine canes (Pawlus *et al.*, 2012). Some of these stilbenes which are very active against *P. viticola*, such as pterostilbene, δ- and ε-viniferins, were also present in higher concentrations in clones of *V. vinifera* Cabernet Franc showing a lower susceptibility to downy mildew and in the resistant genotype *V. riparia* (Boso *et al.*, 2012; Van Leeuwen *et al.*, 2013).

CONCLUSION

Treatment of young grapevine plants in greenhouse with STE from grapevine canes offered considerable protection against *P. viticola* identical to that afforded by Bordeaux mixture. STE also prevented *P. viticola* infection in vineyard but to a lesser extent. Treatment with STE every 7 days gave better results than treatment every 14 days in vineyard.

Taken together, these findings indicate that stilbenes from grapevine canes are good candidates as natural fungicides against downy mildew. Nevertheless, the final product will require refinement to enhance its antifungal activity in the vineyard.

Acknowledgements: The support provided by the French National Research Agency (project ANR-06-EMPB-035-01) is gratefully acknowledged. Benoit Biais is acknowledged for his technical assistance. Analytical experiments were undertaken at the Metabolome Facility at the Functional Genomic Center in Bordeaux, France.

REFERENCES

Adrian M., Jeandet P., Douillet-Breuil A.C., Tesson L. and Bessis R., 2000. Stilbene content of mature *Vitis vinifera* berries in response to UV-C elicitation. *J. Agric. Food Chem.* **48**, 6103-6105.

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

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

Bavaresco L., Petegolli D., Cantu E., Fregoni M., Chiusa G. and Trevisan M., 1997. Elicitation and accumulation of stilbene phytoalexins in grapevine berries infected by *Botrytis cinerea. Vitis* **36**, 77-83.

Belhadj A., Saigne C., Telef N., Cluzet S., Bouscaut J., Corio-Costet M.F. and Mérillon J.M., 2006. Methyl jasmonate induces defense responses in grapevine and triggers protection against *Erysiphe necator*. *J. Agric. Food Chem*. **54**, 9119-9125.

Boso S., Alonso-Villaverde V., Martinez M.C. and Kassemeyer H.H., 2012. Quantification of stilbenes in *Vitis* genotypes with different levels of resistance to *Plasmopara viticola* infection. *Am. J. Enol. Vitic*. **63**, 419-423.

Chalal M., Klinguer A., Echairi A., Meunier P., Vervandier-Fasseur D. and Adrian M., 2014. Antimicrobial activity of resveratrol analogues. *Molecules* **19**, 7679-7688.

Douillet-Breuil A.C., Jeandet P., Adrian M. and Bessis R., 1999. Changes in the phytoalexin content of various *Vitis* spp. in response to ultraviolet C elicitation. *J. Agric. Food Chem.* **47**, 4456-4461.

Dufour M.C., Lambert C., Bouscaut J., Mérillon J.M. and Corio-Costet M.F., 2013. Benzothiadiazole-primed defence responses and enhanced differential expression of defence genes in *Vitis vinifera* infected with biotrophic pathogens *Erysiphe necator* and *Plasmopara viticola*. *Plant Pathol*. **62**, 370-382.

FranceAgrimer, 2012. L'observatoire national des ressources en biomasse – Évaluation des ressources disponibles en France, 96 pp – *Les Études de FranceAgrimer*, Edition octobre 2012.

Houillé B., Besseau S., Courdavault V., Oudin A., Glévarec G., Delanoue G., Guérin L., Simkin A.J., Papon N., Clastre M., Giglioli-Guivarch N. and Lanoue A., 2015. Biosynthetic origin of *E*-resveratrol accumulation in grape canes during postharvest storage. *J. Agric. Food Chem.* **63**, 1631-1638.

Lambert C., Richard T., Renouf E., Bisson J., Waffo Téguo P., Bordenave L., Ollat N., Mérillon J.M. and Cluzet S., 2013. Comparative analyses of stilbenoids in canes of major *Vitis vinifera* L. cultivars. *J. Agric. Food Chem.* **61**, 11392-11399.

Langcake P., 1981. Disease resistance of *Vitis* spp. and the production of the stress metabolites resveratrol, ε-viniferin, α-viniferin and pterostilbene. *Physiol. Plant Pathol.* **18**, 213-226.

La Torre A., Pompi V., Mandalà C. and Cioffi C., 2011. Grapevine downy mildew control using reduced copper amounts in organic viticulture. *Commun. Agric. Appl. Biol. Sci.* **76**, 727-735.

Pawlus A.D., Waffo Téguo P., Shaver J. and Mérillon J.M., 2012. Stilbenoid chemistry from wine and the genus *Vitis*, a review. *J. Int. Sci. Vigne Vin* **45**, 57-111.

Pawlus A.D., Sahli R., Bisson J., Rivière C., Delaunay J.C., Richard T., Gomes E., Bordenave L.,

©Université de Bordeaux (Bordeaux, France)

Waffo Teguo P. and Mérillon J.M., 2013. Stilbenoid profiles of canes from *Vitis* and *Muscadinia* species. *J. Agric. Food Chem.* **61**, 501-511.

Pezet R., Perret C., Jean-Denis J.B., Tabacchi R., Gindro K. and Viret O., 2003. δ-viniferin, a resveratrol dehydrodimer: one of the major stilbenes synthesized by stressed grapevine leaves. *J. Agric. Food Chem.* **51**, 5488-5492.

Pezet R., Gindro K., Viret O. and Spring J.L., 2004. Glycosylation and oxidative dimerization of resveratrol are respectively associated to sensitivity and resistance of grapevine cultivars to downy mildew. *Physiol. Mol. Plant Pathol*. **65**, 297-303.

Rivière C., Pawlus A.D. and Mérillon J.M., 2012. Natural stilbenoids: distribution in the plant kingdom and chemotaxonomic interest in Vitaceae. *Nat. Prod. Rep.* **29**, 1317-1333.

Saigne-Soulard C., Abdelli-Belhadj A., Télef-Micouleau M., Bouscaut J., Cluzet S., Corio-Costet M.F. and Mérillon J.M., 2015. Oligosaccharides from *Botrytis cinerea* and elicitation of grapevine defense, pp 939-958. *In: Polysaccharides, Bioactivity and Biotechnology*. Ramawat K.G. and Mérillon J.M. (Eds.), Springer International Publishing, Switzerland.

Schnee S., Queiroz E.F., Voinesco F., Marcourt L., Dubuis P.H., Wolfender J.L. and Gindro K., 2013. *Vitis vinifera* canes, a new source of antifungal compounds against *Plasmopara viticola*, *Erysiphe necator*, and *Botrytis cinerea*. *J. Agric. Food Chem.* **61**, 5459-5467.

Van Leeuwen C., Roby J.P., Alonso-Villaverde V. and Gindro K., 2013. Impact of clonal variability in *Vitis vinifera* Cabernet Franc on grape composition, wine quality, leaf blade stilbene content and downy mildew resistance. *J. Agric. Food Chem.* **61**, 19-24.

©Université de Bordeaux (Bordeaux, France)