

Proceedings of the

8th International Workshop on Grapevine Downy and Powdery Mildew

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Workshop History

1st Workshop

Geneva, New York, USA
1991

4th Workshop

Napa, California, USA
2002

7th Workshop

Vitoria-Gastiez, Spain
2014

2nd Workshop

Freiburg, Germany
1994

5th Workshop

Trentino, Italy
2006

8th Workshop

Corvallis, Oregon, USA
2017

3rd Workshop

Loxton, S. Australia, Australia
(1998)

6th Workshop

Bordeaux, France
2010

Sponsors



Science For A Better Life

Bayer Crop Sciences - Tuesday and Wednesday Lunch



Viticulture & Enology Program

WASHINGTON STATE UNIVERSITY

Washington State University Viticulture & Enology Program - Monday Lunch



WASHINGTON STATE WINE

Washington State Wine Commission - Monday Breakfast

Wine and Juice Donations

National Grape Cooperative
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Bethel Heights Vineyard
Oregon Wine Board
Argyle Winery
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In memory

Trevor Wicks, Plant Pathologist

1945 -2016

This article has been modified from its original publication in The Advertiser (advertiser.com.au) on 29 October 2016.

Trevor made a significant contribution as a plant pathologist to Australian primary industries over a remarkable 42 years. He started his career as a cadet for the SA Department of Agriculture in 1964 and studied at the same time for a Bachelor's degree in Agricultural Science at the University of Adelaide. He went on to complete a Masters in Crop Protection at Reading University (UK) and a PhD at the University of Adelaide with his doctoral research on 'Phytophthora crown rot of almonds and cherries'.

During Trevor's illustrious career at the South Australian Research and Development Institute, he contributed to research on disease identification and management in all areas of horticulture, making a significant contribution to the management of fungal disease in many crops. This was recognised when he received the prestigious Graham Gregory Award for horticulture in 1995, a national award recognising outstanding achievements in horticulture.

Trevor made an enormous contribution to the Australian wine industry over several decades through development and evaluation of many new fungicides and fungicide application programs that have revolutionised the viticulture industry. He was also interested in alternatives to conventional fungicides. Trevor was well-respected by researchers and wine industry personnel alike and was typically one of the first people to identify new disease problems and advise on their management. He made important contributions to the Cooperative Research Centre for Viticulture, particularly on the biology and control of grapevine powdery mildew and trunk diseases.

Internationally, Trevor was highly regarded for his wealth of knowledge with regular invitations to speak at industry and scientific meetings around the world. Through this he developed a great love of travel, established scientific collaborations between SARDI and many well-known international organisations and was a strong advocate for encouraging students and young scientists to develop their careers through gaining experience overseas.

Trevor was an affiliate Senior Lecturer at the University of Adelaide, where he contributed to mentoring students undertaking Honours, Masters and PhD research projects. He co-supervised at least 18 research students and was an advisor to many more. He particularly enjoyed introducing students to the joys and challenges of field work. He was also a partner investigator and collaborator on several research grants with the University. He is a co-author on numerous papers arising from this work and, over his career, published at least 60 scientific papers and many more articles in industry magazines.

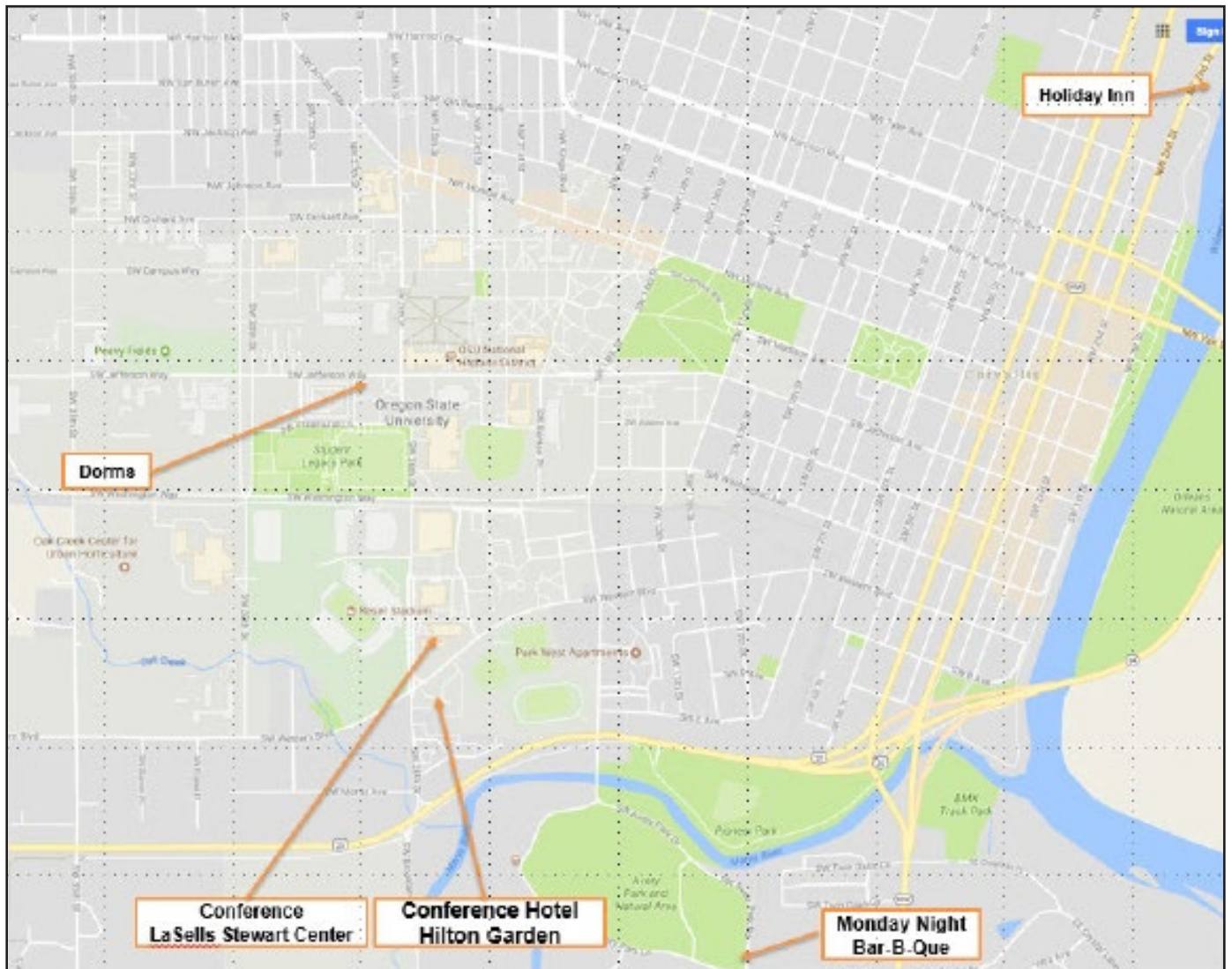
Trevor was diagnosed with a tumour in January 2016 and throughout his illness maintained his interest in



research and in people. Above all, Trevor was a wonderful colleague and friend to many in Australia and internationally, with a gift for making people feel valued. He was eternally curious, happiest in the field assisting growers who appreciated his practical and honest advice on disease management.

He was a wonderful mentor to staff in his close-knit research group and to students, contributing enthusiastically and interested to hear the news from colleagues until he died. He will be deeply missed by his wife Keren, and his many friends and colleagues in South Australia and around the world.

Oregon State University Campus



Program

DAY 1 - Monday

TIME	ACTIVITY
8:00 AM	Registration, Poster Set-up, Continental Breakfast - LaSells Stewart Center
8:30 AM	Introduction to Day
8:45 AM	Session 1: Disease Management 1
	Raynal, Marc - Study of the Variability of Vineyard Sensitivity to the Main Fungal Diseases: <i>A priori</i> Zoning of Physiological Behavior Units (PBU)
	Diez-Navajas, Ana - Reducing the Number of Fungicide Treatments to Control Downy and Powdery Mildew
9:45 AM	BREAK + Poster Viewing
10:00 AM	Session 1: Disease Management 1, con't
	Warneke, Brent - The Grape Powdery Mildew Conundrum: Fungicide Selection and Timing
	Gadoury, David - Why Light Matters: Opportunities to Suppress Powdery Mildews via Relationships that have Evolved Between the Pathogen and the Sun
(30 min)	Group Discussion on Disease Management 1
11:30 AM	Session 2 - Breeding and Host Resistance
	Wiedemann-Merdinoglu, Sabine - INRA-ResDur: A French Grapevine Breeding Program for Durable Resistance to Downy and Powdery Mildew
(15 min)	Group Discussion on Breeding and Host Resistance
12:15 PM	Lunch + Poster Viewing
1:15 PM	Session 3 - Fungicide Resistance
	Hall, Barbara - Fungicide Resistance in Australian Viticulture
	McKay, Suzanne - Incidence and Severity of QoI and DMI Fungicide Resistance of <i>Erysiphe necator</i> in Australia
(30 min)	Group Discussion on Fungicide Resistance
2:45 PM	BREAK + Poster viewing
3:00 PM	Session 4- Plant-Microbe Interactions and Population Genetics
	Scott, Eileen - Quantitative PCR, Fatty Acid Analysis and Mid-Infrared Spectroscopy for Assessment of Powdery Mildew on Grape Berries at Harvest
	Taylor, Andrew - Assessment of the Genetic Diversity of Western Australian <i>Plasmopara viticola</i> Populations
(30 min)	Group Discussion on Plant-Microbe Interactions and Population Genetics
4:30 PM	Discussion on Workshop Future Locations
5:00 PM	Introduction to Oregon and Washington Viticulture
	Overview of Oregon Viticulture (Patty Skinkis - OSU Viticulture Extension Specialist)
	Overview of Washington Viticulture (Michelle Moyer - WSU Viticulture Extension Specialist)
6:00 PM	Opening Reception and Light Refreshments
	Introduction to the PNW region by the Oregon Wine Board and Washington Wine Commission. Sample regional wines and enjoy local appetizers.

Program

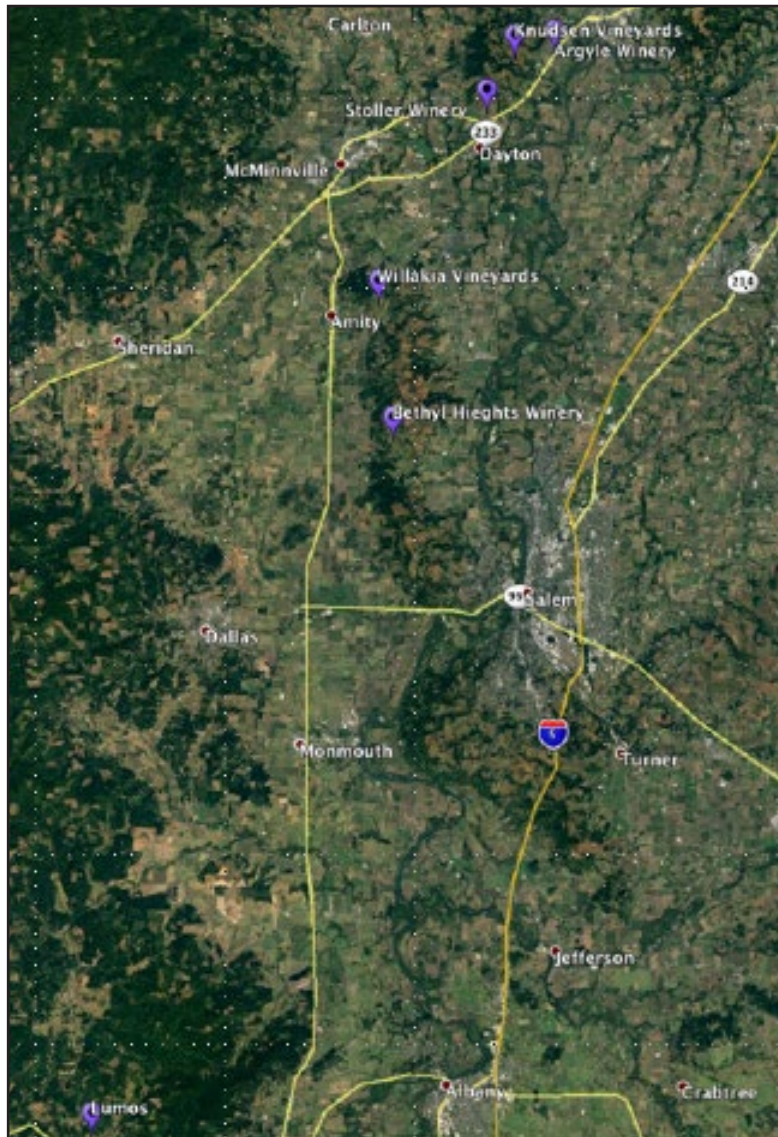
DAY 2 - TUESDAY

TIME	ACTIVITY
8:00 AM	Continental Breakfast - LaSells Stewart Center
8:30 AM	Session 5 - Disease Management 2 - Decision Support Systems
	Bleyer, Gottfried - Experiments to Optimize an Established Plant Protection Strategy Against Grapevine Downy Mildew Based on the Forecast Model "VitiMeteo Plasmopara"
	Magarey, Peter - GrowCare: An Easy-Access Web-Based Tool to Alert Australian Grape Growers to Better Manage Downy and Powdery Mildew
	Caffi, Tito - Sustainable Management of Vineyards: The Experience of a Large-Scale Application of a Web-Based Decision Support System
10:00 AM	BREAK + Poster Viewing
10:15 AM	Session 5 - Disease Management 2 - Decision Support Systems, con't
	Carisse, Odile - Network of Airborne Inoculum Monitoring for Informed Grape Disease Management Decisions
	Mahaffee, Walt - Assessing Transportability of Decision Support Systems - Ascospore Release Prediction
	Dubuis, Pierre-Henri - Contribution of the Swiss Agrometeo Platform to the Reduction of Pesticides Use and Risks
(45 min)	Group Discussion on Disease Management 2-DSS
12:30 PM	Lunch + Poster Viewing
1:30 PM	Session 6 - Epidemiology
	Scott, Eileen - PMapp and Supporting Website: New Tools to Facilitate Assessing Powdery Mildew on Grape Bunches
	Caffi, Tito - A Process-Based Model for Downy Mildew Epidemics on Partially-Resistant Grapevine
2:30 PM	BREAK + Poster Viewing
2:45 PM	Session 6 - Epidemiology, con't
	Dubuis, Pierre-Henri - Downy Mildew Development in Field-Artificially Infected Grape Bunches
	Mahaffee, Walt - Biophysical Modeling of Pathogen Dispersion
(30 min)	Group Discussion on Epidemiology
4:15 PM	Poster Reception
	Hong, Cheng-Fang - Who Can Swim? A New Approach for Determining Sporangia Viability of <i>Plasmopara viticola</i>
	Hong, Cheng-Fang - Epidemiology and Population Biology of Grape Downy Mildew (<i>Plasmopara viticola</i>) in Vineyards in Georgia, USA
	Neill, Tara - Detection and Monitoring of Fungicide Resistance in Oregon, Washington, and California <i>Erysiphe necator</i> Populations
	Pscheidt, Jay - Occurrence of Grape Powdery and Downy Mildew in Western Oregon
	Taylor, Andrew - Detection of <i>Plasmopara viticola</i> Oospores in Western Australian Vineyards
5:45 PM	Rest + Refresh
6:30 PM	Dinner at Avery Park - BBQ and Volleyball - Walking Distance from Hotel / LaSells Stewart Center

Program

DAY 3 - WEDNESDAY

TIME	ACTIVITY
7:15 AM	Continental Breakfast - LaSells Stewart Center
7:45 AM	Load Bus for Vineyard Tours (Leave from LaSells Stewart Center)
9:00 AM	Bethel Heights Vineyard Tour and Tasting with Ted Casteel
10:30 AM	Willakia Vineyard Tour with Geoff Hall
12:15 PM	Box Lunch at Stoller Family Estate, followed by Vineyard Tour and Tasting with Jason Tosch
2:00 PM	Knudsen Vineyard and Argyle Winery Tour with Allen Holstein
5:00 PM	Dinner at Lumos Winery - Vineyard Tour and Wine Tasting
Approx. 8:00 PM	Depart Lumos Winery for Hotel



ABSTRACTS

STUDY OF THE VARIABILITY OF A VINEYARD SENSITIVITY TO THE MAIN FUNGUS DISEASES: A PRIORI ZONING OF PHYSIOLOGICAL BEHAVIOR UNITS (PBU)

Raynal, M.^{1*}, Delfour, B.¹, Debord, C.¹, Vergnes, M.¹, Georges, M.², and Fulchic, R.²

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The systemic analysis of the performance of a vineyard at the scale of a wine-making exploitation is made possible by the use of sensors stemming from precision techniques, which allow precise and exhaustive geo-located measures.

The aim of our study is to exploit this kind of data and evaluate their information using geographical information systems (GIS) and crossing different layers representing characteristic and independent variables of the production system. The goal is then to elaborate an *a priori* zoning, likely to explain variations of the physiological development of vines and possible differences of the plants susceptibility to fungal diseases.

The study is based on the combination of two maps established on the property of Chateau Léoville Las Cases in the Medoc area of the Bordeaux vineyard (France). These maps represent the behavior of the two compartments, soil and plant, determined respectively by means of electric resistivity (R) and biomass index (B) evaluated by Normalized Differential Vegetation Index (NDVI) measures. Three levels - low, medium, high- are defined for each type of data. The combination of these indicators allows the elaboration of 9 classes of islets, named Physiological Behavior Units (PBU), whose distribution is bounded by the GIS on the whole vineyard.

Six of these nine PBU were selected by exclusion of the medium class of the biomass index. Each PBU is replicated twice, thus establishing an observation device of 12 PBU likely to identify differences in terms of physiological development and disease susceptibility. For this purpose, treated and non-treated zones were delimited for each PBU, and a weekly monitoring of these areas was performed during the 2014 - 2016 seasons.

Despite the difficulties related to the implementation of this kind of study at the scale of a 100 hectare vineyard, the first results are very encouraging. They reveal the value of the experimental design: the proposed zoning methodology shows differences regarding both physiological behavior of the vines and their susceptibility to diseases. The PBU concept thus appears relevant regarding analysis of both production and fungal attack variabilities. It should allow a better understanding of the functioning of our winemaking production systems.

The study confirms the interest of capitalizing on the exhaustive data stemming from remote sensors operated within the framework of emergent precision viticulture technics. These first results will need to be confirmed through experimentation over several vintages in order to better understand the temporal stability of the studied criteria and their effective role and impact according to climatic patterns. Moreover, we analyzed the explanatory power of a set of other variables stemming from the Chateau's database (e.g., age of plantation, rootstocks, clones, exposure), as it may relate the observed variability in diseases. This first global qualitative approach shows that all of these data explain part of the variability observed in the studied processes. Our next goal is to combine these factors to the existent PBU's in order to define a zoning of plot susceptibility and lower fungicide applications according to epidemic cycles and meteorological forecasts.

REDUCING THE NUMBER OF FUNGICIDE TREATMENTS TO CONTROL DOWNY AND POWDERY MILDEW

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European policies have been designed to achieve a sustainable use of pesticides by reducing their risk and impact on human health and the environment, and by promoting the use of integrated pest management and alternative techniques such as non-chemical approaches (DIRECTIVE 2009/128/EC). In this frame, LIFE FITOVID project evaluated different spray schedules to control grapevine downy and powdery mildew to reduce the usual number of applications to control both diseases. The action was performed in two areas with high disease pressure for each disease. The number of applied treatments during one growing season and under grapevine grower's criterion was compared to the different disease management schedules evaluated. Those schedules targeting powdery mildew were based on: (1) disease risk indicated by a weather station (Pessl Instruments), based in previous studies by Arens, Blaeser and Gehmann, (2) first treatment applied by degree-day accumulation and following ones by phenology, using conventional fungicides, and (3) a program similar to #2, but using organic-labeled fungicides. Downy mildew control was evaluated comparing to other two schedules based on (1) disease risk model by Gubler and Thomas provided by station and (2) phytochemicals for organic production applied according to phenology.

After treatment, disease symptoms were evaluated weekly in the growing season, disease evolution described, grape bunch and grapevine wood production measured, and reduction in the number of applied treatments analyzed.

The program based on degree-day accumulation and phenology and conventional chemicals application registered the lower number of treatments to control powdery mildew. Disease control results of this program became quite close to those obtained from plots treated under grower's criterion with conventional chemicals. Respect to downy mildew, the program based on risk disease showed encouraging results, decreasing the number of treatments by half with respect to the program applied by grower.

REFERENCE:

Pessl Instruments – Powdery Mildew Risk. <http://www.metos.at/tiki/tiki-index.php?page=The+Pessl+Instruments+Model+for+Grape+Vine+Powdery+Mildew+Risk>

THE GRAPE POWDERY MILDEW CONUNDRUM: FUNGICIDE SELECTION AND TIMING

Warneke, B.¹, Thiessen, L.², Neill, T.³, Mahaffee, W.^{3*}

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Grape powdery mildew (GPM, causal agent *Erysiphe necator*) is the most economically important disease of grapevine in the western U.S. The inflorescence and early infructescence growth stages are highly susceptible to GPM and its management during this window is critical to preventing economic losses. Targeting these stages with mobile fungicides could significantly reduce fruit infections. Leaf bioassays were conducted to characterize the mobility of common fungicides. All fungicides tested had protective vapor mobility in the lab and five of them were confirmed in the field. In addition, xylem and translaminar protective mobility were assessed and six had some degree of both attributes. A small-plot experiment was conducted in Corvallis, Oregon during the 2015 and 2016 growing seasons to examine the interaction between fungicide chemistry and application timing on GPM berry infection. Five commonly used fungicides with varying degrees of mobility were applied at three different growth stages around bloom. The controls consisted of a nontreated and sulfur, each applied every 14 days. The proportion of grape berries infected with GPM was significantly influenced by the interaction between fungicide selection and application timing (Drop-in-deviance test, $\chi^2 = 33.1$, $df = 8$, $P < 0.01$). All treatments significantly reduced fruit infection compared to the nontreated control (Z test, $P < 0.01$). The trifloxystrobin, quinoxyfen, and fluopyram applications made at berry set were most effective at reducing the proportion of berries infected, having 4.8, 8.0, and 11.5 times lower odds of berry infection, respectively, compared to the earliest timing (Z test, $P < 0.01$). This research indicates that using mobile fungicides during fruit development may improve disease control.

WHY LIGHT MATTERS: OPPORTUNITIES TO SUPPRESS POWDERY MILDEWS VIA RELATIONSHIPS THAT HAVE EVOLVED BETWEEN THE PATHOGEN AND THE SUN

Gadoury, D.M.¹, Suthaparan, A.², Giselsrød, H.-R.², Solhaug, K.A.², From, P.J.², Stensvand, A.^{2,3}, Rea, M.⁴, Bierman, A.⁴, Patel, J.⁴, Peres, N.⁵, Onofre, R.⁵, McCann, T.¹, and Cadle-Davidson, L.⁶

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Plants and their pathogens have evolved over millions of years amidst 24-hour cycles of light and dark. Many pathogens interpret and use light to direct their development. Among fungal pathogens, powdery mildews are nearly wholly external to their host, and occupy a niche that places them in a position to receive direct solar radiation, and (since the advent of electric lighting) supplemental radiation from various sources. In our most recent work, we have identified a number of light-sensitive processes in powdery mildews, and have used both visible and UV light to affect pathogen growth and suppress disease. Recent developments in solid-state lighting create additional opportunities to use light spectra, intensity, duration, and timing in ways that were never before possible. Both continuous broad spectrum illumination and brief treatments with specific wavelengths in the long-wavelength (red) region of the spectrum have been used to suppress powdery mildews without deleterious host effects. The discovery that nighttime applications of UVB or UVC can circumvent the active DNA repair mechanisms of powdery mildews, and that red light can be used synergistically with UV treatments permits greatly reduced UV doses in greenhouse and field studies. This presentation will review a number of recently published works and current research wherein the following has been demonstrated: (i) the effective suppression of a variety of powdery mildews by specific LED lighting, (ii) nighttime applications of UVB and UVC to suppress a variety of powdery mildews, (iii) light-mediated conidiation in powdery mildews, (iv) effects of specific spectra on various processes in powdery mildews, (v) considerations for design of lighting systems for field and greenhouse use, and (vi) static, mobile robotic, and mobile tractor-drawn systems for lighting treatments for disease suppression.

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INRA-RESDUR : A FRENCH GRAPEVINE BREEDING PROGRAM FOR DURABLE RESISTANCE TO DOWNY AND POWDERY MILDEW

Wiedemann-Merdinoglu, S.* , Prado, E., Dumas, V., Dorne, M-A., Lacombe, M-C., Onimus, C., Piron, M-C., Umar-Faruk, A., Duchene, E., Mestre, P., Schneider, C., and Merdinoglu, D.

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The current strategy to control grapevine downy and powdery mildew relies on chemical treatments. The most promising option to reduce the environmental impact of this practice is the use of resistant varieties. This is why a new INRA breeding program called INRA-ResDur was launched in 2000 to create varieties with a durable resistance to downy and powdery mildew and with a berry quality suitable for the production of high quality wines. Various American and Asian resistance sources have been described for a long time and during the last decade, intense genetic analyses of some of them have unveiled several resistance loci.

However, resistance breakdown has already been observed for the locus Rpv3 (resistance to *Plasmopara viticola* derived from the resistant variety Bianca) and for the locus Run1 (resistance to *Uncinula Erysiphe necator* derived from Muscadinia *Vitis rotundifolia*).

To ensure the durability of resistance, in the INRA-ResDur program, we used marker-assisted selection (MAS) to stack resistance factors derived from multiple sources. Thus, MAS allowed us to follow six resistance alleles: Rpv1, Rpv3 and Rpv10 for downy mildew and Run1, Ren3 and Ren3.2 for powdery mildew. This strategy led to the development of candidate varieties bearing not only one but two or three genes to control each disease. In addition, new cropping systems using a minimal plant protection regime are being assessed to limit the adaptation of pathogen populations to resistance factors and to avoid the emergence of secondary diseases (black rot, anthracnose) that may appear in the absence of phytosanitary treatments.

A set of approximately 30 new resistant varieties will be proposed for registration between 2017 and 2024. This project is the result of a national collaboration between INRA and the French Wine and Vine Institute (IFV) but also with other European institutes: the Julius Kühn-Institut (JKI) and the Staatliches Weinbauinstitut (WBI) in Germany and with the Agroscope in Switzerland.

FUNGICIDE RESISTANCE IN AUSTRALIAN VITICULTURE

Hall, B.H.^{1*}, McKay, S.F.¹, Lopez, F.², Harper, L.², Savocchia, S.³, Borneman, A.⁴, and Herderich, M.T.⁴

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Fungicide resistant populations add to the cost of disease management due to reduced fungicide efficacy and failure of spray programs to control disease. Following the discovery of populations of *Erysiphe necator* resistant to the strobilurin fungicides, with reports of field failure, research was initiated to determine the incidence and severity of resistance in vineyards in the main viticultural regions of Australia. Isolates of *E. necator* were tested for sensitivity to pyraclostrobin, penconazole, myclobutanil and tetraconazole, and *Plasmopara viticola* to metalaxyl M, mandipropamid and pyraclostrobin using leaf disc assays. Sequences of *E. necator* DNA were aligned to reference sequences to identify the Y136F (cyp51) or G143A (cytB) alleles and the presence and frequency of the G143A mutation in *P. viticola* DNA was determined using next generation sequencing (NGS) of a 180 bp amplicon that surrounded the G143A mutation.

Results showed that QoI resistance in *E. necator* was found in most viticultural regions, with 52% of the 94 sites tested having phenotypic resistance. Phenotypic resistance of *E. necator* to the DMIs was not widespread, with 14% of samples resistant to myclobutanil and none to penconazole or tetraconazole. The G143A allele was detected in 86% of the *E. necator* populations and the Y136F allele was present in over 68%. Limited samples of *P. viticola* were able to be tested, however 13 of the 18 sites tested from 5 regions were resistant to metalaxyl. The G143A allele in the cytB gene was detected in *P. viticola* populations from only one region in New South Wales. Fifteen additional samples were collected from this region and genotyped using NGS to determine the presence and frequency of the G143A allele. Eight were wild type, the others ranged from 10 to 98% G143A. Phenotypic testing from this area showed at least one sample was most likely resistant, however results were inconclusive and need to be repeated.

The results of this work have highlighted significant gaps in our knowledge, with the main area of uncertainty being the correlation between the laboratory testing results and field performance of a fungicide. Future research is concentrating on these knowledge gaps, including areas such as the relative fitness of resistant populations compared to wild type, the differences in DMI resistance and efficacy between the various products, and developing base line data for other fungicide groups. Sampling methods need to be refined to provide effective relationships between laboratory testing and field resistance status. Preliminary studies using NGS have shown that 10 random samples in a vineyard will give a reasonable indication of the proportion of resistant alleles present. However, the relationship between the phenotypic and genotypic results needs to be elucidated before the genotypic tests can be used to give an accurate indication of field resistance.

Until these knowledge gaps have been filled, the current advice to growers remains: adhere to the resistance management strategies and minimize exposure to any one of the “at risk” fungicides.

INCIDENCE AND SEVERITY OF QOI AND DMI FUNGICIDE RESISTANCE OF *ERYSIPHE NECATOR* IN AUSTRALIA

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One hundred and eight isolates of *Erysiphe necator* were established from 27 different areas within each of the major wine-grape growing regions of Australia. Isolates were tested, using *in vitro* assays, for sensitivity to the Quinone outside Inhibitor (QoI) fungicide pyraclostrobin, (Cabrio® 250 g/L ai) and the Demethylation Inhibitor (DMI) fungicides, penconazole (Topas®, 100g/L ai), myclobutanil (Mycloss™ Xtra, 250g/L ai) and tetraconazole (Domark®, 40g/L ai). Due to the widespread use of fungicides, only five isolates that had not been previously exposed to any synthetic fungicides were obtained. Isolates were also screened, using Sanger sequencing, to detect the presence of the mutations associated with resistance to both the QoI and DMI fungicide groups, G143A and Y136F respectively. Detection and quantification of both alleles using Next Generation Sequencing (NGS) was carried out for a subset of isolates. The EC₅₀ values for pyraclostrobin ranged between 0.008-14.4 µg/mL, with 27% of isolates tested in 2013/14 being > 1.0 µg/mL which were considered phenotypically resistant. Sanger sequencing revealed 70% of isolates were heterogenous G143/G143A genotype, with 12 % wild type (G143) and 18% G143A. NGS revealed G143A allele frequency (%) ranged between 3 and 100%. Sixty isolates, established from plant material collected during the 2015-16 growing season, were tested using a discriminatory-dose assay. Sixty-one percent were classified as resistant and NGS allele frequencies of G143 ranged between 8-100%. There was no clear relationship between EC₅₀ values, or discriminatory dose results and the % frequency of G143A. The EC₅₀ values were lower for those isolates unexposed to QoIs compared to those previously exposed. The EC₅₀ values for penconazole, myclobutanil and tetraconazole ranged between 0.0004-0.7, 0.002-6.6 and 0.0008-0.02 µg/mL respectively. The Y136F allele was present in over 60% of isolates, with 26% wild type, 29% heterogeneous Y136/Y136F, and 44% Y136F using Sanger methodology. Y136F allele frequency was between 0-100% using NGS. There was clear no relationship between DMI phenotype and genotype results. Although, the number of isolates previously unexposed to DMIs were low, there was no obvious relationship between EC₅₀ values and exposure for each of the DMIs tested. Our results show that QoI resistance, using either phenotype or genotype results, is widespread, and that decreased sensitivity to myclobutanil has been found for the first time in Australia, however the link between laboratory results and field performance needs to be further explored.

QUANTITATIVE PCR, FATTY ACID ANALYSIS AND MID-INFRARED SPECTROSCOPY FOR ASSESSMENT OF POWDERY MILDEW ON GRAPE BERRIES AT HARVEST

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Powdery mildew (*Erysiphe necator*) affects organoleptic properties of winegrapes and simple, objective and accurate measures are needed for assessment of fruit prior to grading. The aims of this study were to (i) develop a sensitive and powdery mildew-specific quantitative PCR assay suitable for discriminating healthy and infected individual berries, and (ii) investigate fatty acids and mid-infrared (MIR) spectroscopy as alternative approaches for distinguishing powdery mildew-affected berries.

Fully developed, intact berries (n=138) were detached from 35 Chardonnay bunches (17-18 °Brix) collected in a research vineyard (Waite Campus, University of Adelaide, Australia). All berries were visually assessed using a stereomicroscope (18x magnification), grouped as healthy, partly- or fully-infected, deseeded, homogenized and scanned in the MIR range (4000-400 cm⁻¹). A subsample of 54 berries was selected for subsequent qPCR analysis and 7 additional berries were included for fatty acid analysis (61 berries in total) using gas chromatography and thin layer chromatography. *E. necator* was quantified using a duplex TaqMan qPCR assay with MGB probes, with modified *E. necator* (from the original pEnA1 fragment described by Stummer et al. 2006) and *V. vinifera* primers (from the actin 1 gene). The limit of detection for pure *E. necator* DNA from conidia was 3 fg (Ct 31 ± 0.65). In the 54 berries, we detected 6 fg - 0.1 ng of pure *E. necator* DNA per 0.5 ng of total DNA (*E. necator* + *V. vinifera*). *E. necator* biomass among 20 visually healthy berries varied widely (6 fg to ca 0.003 ng of pure *E. necator* DNA), and was less variable on visually partly- and fully-infected berries (0.006 to ca 0.1 ng). Saturated fatty acids were prevalent in *E. necator* collected from the surface of berries, arachidic acid (C20:0) being most abundant. However, total saturated and total unsaturated fatty acids (C14-C24) were similar in healthy and infected berries. Arachidic acid content was strongly related to increasing *E. necator* biomass on berries (R²=0.76), and was a good discriminator for the visual infection groups. MIR spectra (1800-1185 cm⁻¹), with peaks corresponding mainly to fatty acid esters, water, carboxylic acid and proteins (amide I and II), contributed most to discrimination of berries with traces of *E. necator* from visually fully-infected berries but did not distinguish the intermediate group. The relationship between *E. necator* biomass and arachidic acid content in berries suggests that analysis of arachidic acid may be a simple and objective tool for measuring *E. necator* and powdery mildew severity.

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ASSESSMENT OF THE GENETIC DIVERSITY OF WESTERN AUSTRALIAN *PLASMOPARA VITICOLA* POPULATIONS

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The first detection of *Plasmopara viticola* in Western Australia (WA) occurred on grapevines in Kalumburu, East Kimberley in 1997. Despite these vines being destroyed further detections occurred in commercial production areas, approximately 3750km south of the initial detection, during the 1998/1999 vintage. Twenty years after the initial detection downy mildew is now established throughout all grape production areas of the state. During the 2014/15 and 2015/16 vintages, 65 isolates were collected from 35 separate vineyards across all major WA production regions; a geographic range of approximately 500km. Combined with four historic herbarium samples, including those from the original Kalumburu detection, the genetic diversity was assessed using thirteen microsatellite markers: ISA, CES, GOB, PV13, PV17, PV65, PV103, PV137, PV140, PV142, PV143, PV146, PV148. Preliminary results indicate that genetic diversity amongst the WA population is low. The common alleles in isolates collected 20 years apart suggest that the *P. viticola* in WA has occurred from a single incursion event. This is part of a larger population study that includes isolates from across Australia.

EXPERIMENTS TO OPTIMIZE AN ESTABLISHED PLANT PROTECTION STRATEGY AGAINST GRAPEVINE DOWNY MILDEW BASED ON THE FORECAST MODEL "VITIMETEO PLASMOPARA"

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The modeling platform "VitiMeteo" (VM) is a cooperative project between the State Institute for Viticulture and Enology, Freiburg (Germany), Agroscope (Switzerland) and the company GEOsens, Schallstadt (Germany). VM Models are widely used in other countries in Europe (Bleyer et al. 2014). The group is supported by other research institutes. "VM Plasmopara"; was the first VM - module created in 2002. It calculates the most important steps of the infection cycle of downy mildew (Bleyer et al. 2008).

The next component a growth model called "VM Growth", was programmed in cooperation with H.-R. Schultz from the Geisenheim University (Schultz 1992) to account for trellising systems, and was combined with the downy mildew model. During the past decades the State Institute in Freiburg has developed and established an effective growth- and model-based strategy to control grapevine downy mildew. According to the current strategy, the effective period of fungicides is limited by the growth. Under permanent and heavy infection pressure 300 to 400 cm² of unprotected leaf area (2 to 3 new leaves) / primary shoot can develop before the next treatment is necessary (Bleyer et al. 2003).

Potassium phosphonate (Veriphos®) has been a registered in Germany since 2014. With this systemic compound, four trials were carried out between 2013 and 2016 to optimize the established strategy. The objective of the experiments was to investigate whether the combination of potassium phosphonate with other fungicides can be used to extend the treatment intervals. Trials were carried out according to the different variants after the growth of 400 cm², of 600 cm², and of 800 cm² leaf area per primary shoot.

The findings show that potassium phosphonate increases the effectiveness of the preventive fungicide "Folpet" in the three tested spray intervals under high infection pressure. The results of the experiments also confirmed that it is possible to extend the treatment intervals from 400 cm² to 600 cm² leaf area, if potassium phosphonate was added to the preventive compound "Folpet". However the combination is not sufficient enough under high infection pressure in intervals of 800 cm² leaf area. The next step will be to test this further developed strategy in practice.

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GROWCARE®: AN EASY-ACCESS WEB-BASED TOOL TO ALERT AUSTRALIAN GRAPEGROWERS TO BETTER MANAGE DOWNY (AND POWDERY) MILDEW

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GrowCare®, a new web- and modified existing email-service for grape growers and industry personnel, provides: 1) alerts for infection risk and, when needed; 2) timely disease and vineyard management information. GrowCare®-web was established to improve the management of downy mildew (*Plasmopara viticola*) by identifying primary and secondary infection events and oilspot incubation periods. GrowCare®-web now also provides: 1) the risk of flower and fruit infection by bunch rot (*Botrytis cinerea*); 2) a degree-day calculator for pests; and 3) (in preparation) a daily guide to favorability of weather for powdery mildew (*Erysiphe necator*). Users may access an “easy-to-read, customized” graph of data from local weather station (AWS) networks established by South Australian grape industry associations in the Riverland, the Barossa and Clare Valleys, and in the Victorian Murray Valley. The AWS monitor vineyard canopies for temperature, relative humidity, rainfall and leaf wetness. A web-based version of DModel (Magarey et al. 1991) processes the data from a grower-nominated AWS. Users may select which data to graph and which sub-model(s) of DModel to show the progression of disease epidemiology. If all sub-models indicate positive, an infection alert appears and, if elected, an SMS-text and/or emailed message is automatically dispatched to the user. A progressive graph calculates the date oilspots will appear and thus by when a post-infection fungicide (e.g., metalaxyl) can be applied. An IFrame allows users to easily switch to/from Bureau of Meteorology weather forecasts and rainfall radars to assess risk of subsequent disease events and so, when to apply pre-infection fungicides. GrowCare® menu-bars provide other services including: a disease map showing the locality of reported occurrences of downy and powdery mildew; season-lists of predicted disease events; a preliminary version of a library of regional e-News bulletins and scientific and extension papers relating to the diseases of grapes; and Disease Diagnosis®, a web-based “wordless” identification of symptoms of diseases, insects and disorders in Australian and New Zealand vineyards. The GrowCare® e-News under name CropWatch®, has been educating Riverland growers for 22 seasons. Prior to its implementation in the early-1980's, an average of 4-6 sprays were applied for downy in dry seasons. Through the educational email-service, most growers now apply 1-2 sprays. In an era of the demise of “disease experts” (experienced personnel), the comprehensive but easy-to-use GrowCare® system is designed to disseminate epidemiological expertise and provide growers unprecedented precision in “self-managing” vineyard diseases. GrowCare®-web supersedes earlier systems such as the over-priced, more complicated and now defunct AusVit® DSS. Reasonably-priced and simple to operate GrowCare®, with annual-subscription of AUD\$99/season, might save a user one spray/season (@ AUD\$99/ha). Thus, an average vineyard of 30ha might save \$3,000/season returning an annual net gain of AUD\$2,901. Though available for 2-seasons (in beta-form), uptake to date has been unproductively insignificant with only approximately 10 subscribers out of 1,000 Riverland growers! A work-station will be set-up to allow delegate feedback.

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SUSTAINABLE MANAGEMENT OF VINEYARDS: THE EXPERIENCE OF A LARGE-SCALE APPLICATION OF A WEB-BASED DECISION SUPPORT SYSTEM

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Vite.net® is a Decision Support System (DSS) developed and provided by Horta (www.horta-srl.com), a spin-off company of the Università Cattolica del Sacro Cuore, which has been used on a large scale basis across Italy for the sustainable management of vineyards. The DSS is able to collect information in real-time about different vineyard components (air, soil, plants, pests, and diseases) and, by means of a web-based tool, is able to analyze these data by using advanced modelling techniques. The DSS provides up-to-date information for the management of the vineyard at plot level, in the form of alerts and decision supports. The DSS provides information about the grapevine phenology and canopy development, the risk about different diseases (i.e., downy and powdery mildews, black-rot) and pests (berry moth, American leafhopper, mealybug), the protection provided by the last application of plant protection product (PPP) performed and, eventually, the soil water status in the vineyard. The DSS was validated over a network of 21 organic farms in Italy, where disease control and control costs were compared in those parts of the vineyards managed using Vite.net® to those parts managed according to the usual farm practice. Over two seasons, the disease control obtained using the DSS was not statistically different from the one obtained with the usual farm practice, but the total amount of copper was reduced by almost 40% because of both reduced doses and fewer applications. The average saving obtained by organic growers using Vite.net® was 195 €/ha/year relative to their usual farm practice. The DSS platform is open to the integration of new components or add-on services provided by the last results from research. For instance, in 2015, an alert service was added to the DSS in order to provide the user with SMS and emails anytime a particular threshold or event was reached. In 2017, a new epidemiological model for *Botrytis cinerea* was added as a prototype to the DSS. The DSS Vite.net® has been made commercially available in January 2013. In 2016, it was consulted by more than 300 users (farmers and advisors, both public and private) and covered more than 10.000 ha of vineyards across Italy. Several locations were also reached across Europe (i.e., Greece, Spain, Portugal and England). Statistics about the use of the DSS by these growers based on their access to the web portal of vite.net® as well as the feedback collected during the regular contacts with them showed an average reduction in the number of treatments and, as a consequence, in the cost of grapevine's pest and disease control.

NETWORK OF AIRBORNE INOCULUM MONITORING FOR INFORMED GRAPE DISEASE MANAGEMENT DECISIONS

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During the last decade, we collectively improved our capacity to monitor airborne plant pathogen inoculum, including *Plasmopara viticola* and *Erysiphe necator*. Sensitive and accurate DNA-based assays for detection and quantification of airborne inoculum are available. However, one of the main limitations for using airborne inoculum information to make disease management decision is the spatial resolution. Gregory in 1973 suggested that most spores of plant pathogens do not disperse beyond the field in which they were produced. Based on the limited information available on spatial distribution of airborne inoculum, it can be assumed that a large number of sampling sites would be required to reliably monitor airborne inoculum. However, empirical observations suggest that to estimate disease risk rather than inoculum size, a small number of sampling sites strategically organized in a network is sufficient. Consequently, the objective of this study was to assess the value of a network of *P. viticola*, *E. necator* and *Botrytis cinerea* airborne inoculum monitoring for downy and powdery mildew, and Botrytis bunch rot management. In 2016 and 2017, spore samplers were installed at 11 and 14 vineyards and airborne inoculum concentrations of *P. viticola*, *E. necator* and *B. cinerea* were monitored three times weekly using qPCR assays. Information on airborne inoculum was used to estimate disease risk, to time fungicide applications, and to monitor effectiveness of disease control. In 2016, the first airborne sporangia of *P. viticola* f.sp. *riparia* were detected on 29 May while those of *P. viticola* f.sp. *aestivalis* were detected 2 months later, on 26 July. Airborne inoculum remained below 5 sporangia/m³ until 18 August then it increased rapidly to reach 18 sporangia/m³. For powdery mildew, airborne inoculum remained low for most of the season and followed a previously developed degree-day model. Grape advisors and growers were involved in the development of the disease management decision scheme which was based on growth stage, disease history, cultivar susceptibility, weather conditions and airborne inoculum. Despite the time and cost required to collect and process air samples, it was concluded that the network of grape pathogen inoculum monitoring improved our predictive ability and our capacity to manage these diseases.

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ASSESSING TRANSPORTABILITY OF DECISION SUPPORT SYSTEMS - ASCOSPORE RELEASE PREDICTION

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There have been several empirical models recently developed to correlate environmental conditions to *Erysiphe necator* cleistothecia dehiscence and initiation of the grape powdery mildew epidemic in a number of different grape production regions. While these models adequately represent the validation datasets, there are questions as to how they will transport to regions with differing microclimate due to the empirical nature of the models. It is likely that models are not suitably predictive for environments where neither the host nor pathogen is native.

The Caffi, Carrise, Moyer, and Gubler/Thomas models were assessed for their suitability to predict ascospore availability in the Maritime climate of the Oregon Willamette Valley. To assess ascospore release, both natural and artificial grape trunk infestations were monitored for three overwintering seasons from 2012 to 2016. For the artificial infestations, cleistothecia were first collected by vacuuming from leaves collected prior to leaf drop and onset of fall rains. Then, the cleistothecia were overwintered outside by placing them onto natural and artificial grape trunk segments with custom impaction traps placed immediately under each trunk segment. In a research vineyard, airborne inoculum was also concurrently monitored by placing impaction traps adjacent to grape trunks trained in a bilateral cane-pruned guyot. Ascospore inoculum availability was assessed from leaf drop (BBCH 97) until the onset of the disease epidemic in the following growing season. Sample rods were collected from traps on a biweekly basis and qPCR used to estimate *E. necator* inoculum concentration. Weather and inoculum concentration data were used to assess models, and develop empirical models relating local environmental conditions to inoculum availability. Cleistothecia dehiscence and inoculum availability was predicted by all models prior to bud break (BBCH 08), and was observed from the first rain event following the start of inoculum monitoring until monitoring ceased. All models over-predicted cleistothecia dehiscence and inoculum availability in the Willamette Valley and predicted exhaustion of inoculum prior to bud break. The magnitude of inoculum availability from the vineyard or grape segments could not be correlated to environmental conditions, thus a binary model for inoculum availability was developed where inoculum presence is a function of the concurrent occurrence of the following factors within a 24-hour period: > 6 hours of cumulative leaf wetness during temperatures > 4 °C, precipitation > 2.5 mm, and relative humidity > 80%. These results indicate that a more mechanistic understanding of the factors influencing cleistothecia maturation is required to develop models that are transportable across grape production regions and that an improved understanding of factors affecting cleistothecia maturation is needed.

CONTRIBUTION OF THE SWISS AGROMETEO PLATFORM TO THE REDUCTION OF PESTICIDES USE AND RISKS

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The negative effects of plant protection products on the human health and the environment have become a major concern for consumers and politics in Switzerland as well as in most European countries. In the last years new policies for risk mitigation and reduction of use of synthetic pesticides have been enforced in Switzerland. A National Action Plan (NAP) to reduce the risks and the use of plant protection products will start in 2017. The NAP set goals, milestones and measures to reach these objectives. Viticulture is an important consumer of plant protection products especially for fungicides. In Switzerland typically 8 to 10 applications per year are needed to control powdery and downy mildew. In order to respond to the objectives of the NAP different strategies and tools can be enforced by the growers. A possible strategy is to spray according to the epidemic of diseases and pests by following decision support systems (DSS). The platform VitiMeteo offers a wide set of forecasting systems for viticulture which were developed in collaboration with the Staatliches Weinbauinstitut Freiburg (WBI, Germany) and the company Geosens (Germany). A validation of the VitiMeteo-Oidium model over six years in different Swiss vineyards showed an average reduction of one spray per year (~15%) with no reduction in efficacy compared to the reference program. The VitiMeteo-Plasmopara model enable to spray according to the epidemics of downy mildew and allow a better control of the disease. However, the reduction of the number of sprays strongly depends on the weather conditions of the year. A further significant tool is crop adapted spraying, a method which adapts the applied product quantity to the leaf surface to be protected. In Switzerland with this technique it is possible to reduce by 20% the quantity of fungicide use per season. The combination of these different DSS allow a significant reduction (up to 50%) of plant protection products used. All these forecasting systems and tools are available for the Swiss winegrowers on the platform <http://www.agrometeo.ch> free of charge. The use of this platform will help the winegrowers to meet the current society expectations.

PMAPP AND SUPPORTING WEBSITE: NEW TOOLS TO FACILITATE ASSESSING POWDERY MILDEW ON GRAPE BUNCHES

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Powdery mildew has the potential to affect wine quality. Many Australian wineries use a threshold of 3-5% powdery mildew severity on bunches close to harvest to inform decisions about quality and price. Assessment is based on visual estimation, which is acknowledged to be subjective. The quality of assessment strongly depends on the experience and training of field assessors in disease recognition and estimation of bunch area with disease. In collaboration with wine sector representatives, a free application, PMapp, for Apple and Android smart-phones and tablets was developed to facilitate assessment of powdery mildew in vineyards and adoption of uniform assessment practices. An assessment recording screen presents the user with categories from trace (0.5%) to 100%, with 1% increments from 1-10% and larger increments thereafter. The severity score for each bunch assessed is entered and the screen displays cumulative bunch count, incidence and severity on a row and patch basis. The data, including information about date, time and location (with latitude and longitude), can be exported on completion of the assessment for subsequent analysis. PMapp has a bank of computer-generated images of bunches to facilitate assessment of area, a self-calibration tool that allows the user to check his/her accuracy and a diagrammatic key with 2% increments in the range 2-12% for those who find a key helpful. PMapp was released in Australia in December 2015 and worldwide in November 2016 and was downloaded over 2700 times by May 2017 (Australasia 60%, North America 21%, Europe 17%). PMapp is available at <https://appsto.re/au/qe-e5.i> and <https://play.google.com/store/apps/details?id=com.lemuresoftware.pmapp>.

A website to support use of PMapp was developed at the request of wine sector collaborators. This website (www.pmassessment.com.au) offers a best-practice, stepwise guide to in-field assessment with links to training for disease recognition, area assessment and the diagrammatic key. The disease recognition component comprises 28 high-resolution photographs, in triplicate, of powdery mildew on red and white bunches at veraison and close to harvest; each photograph has areas outlined that might represent surface with visible powdery mildew. The user is asked to select the image (of three) that has powdery mildew outlined most correctly. The area assessment component features the computer-generated images in PMapp. The user takes a "test" comprising low-range (0.5-15%, 20 images) or full-range (0.5-90%, 30 images) severity in which certain images are shown three times to assess repeatability. Output comprises a chart showing agreement of the estimate with the actual image (Lin's concordance value), repeatability and time taken for each image. Output can be printed and used as proof of capability, should an employer require evidence. Results are retained and can be accessed by the user, via log-in and password, so that performance can be tracked and analysed over time.

Two of Australia's largest wine companies used these resources in 2017. One incorporated the recording component of PMapp into their standard operating procedures and made 375 assessments. The other used the website, image bank, self-calibration and key components to train new field staff (8 in 2017) to assess disease. Informal feedback from leaders in the sector indicates that these resources have improved the quality of disease assessment.

A PROCESS-BASED MODEL FOR DOWNY MILDEW EPIDEMICS ON PARTIALLY RESISTANT GRAPEVINE

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Process-based simulation modelling can be a powerful tool to guide phenotyping. This seems particularly interesting in the case of disease on perennials, or when plant material is hard to manipulate experimentally, as in the case of grapevine. A process-based approach is used to mobilize and synthesize the knowledge on downy mildew (DM) of grapevine, caused by the oomycete *Plasmopara viticola*. The system is described through a generic simulation model designed to analyze DM epidemics on both leaves and bunches, by considering phases of the disease cycle where components of partial host resistance are at play, such as infection efficiency, sporulation, infectious period and latency period. The model involves numerical integration. It involves five groups of processes: i) crop growth, development and senescence; ii) primary and secondary infection, iii) infection on leaves; iv) infection transmission to clusters; and v) effect of genetic and ontogenic resistance. The model is encoded using the STELLA® simulation software. Specifications of the model are described and simulated outputs are presented. Knowledge gaps and insights for further research are highlighted from this framework.

Federica Bove carried out this work within the Doctoral School Agrisystem of the Università Cattolica del Sacro Cuore (Italy).

DOWNY MILDEW DEVELOPMENT IN FIELD ARTIFICIALLY INFECTED GRAPE BUNCHES

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In previously published work (Gindro et al. 2012) *in vitro* artificial *Plasmopara viticola* infections on detached clusters of two susceptible grapevine cultivars were successful only at very early development stage (BBCH 53) when functional stomata were present, while no infections were observed at later stages (BBCH 69 and 75). This is in contradiction with field observations where later infections are frequently recorded. In attempt to explain this paradox, field experiments were conducted during two seasons on Chasselas plants (15-years old) covered with a plastic roof to protect them from rain and avoid natural infections. Artificial infections of clusters were made at four developmental stages of the grapes: BBCH 55 (inflorescences swelling, flowers closely pressed together), 65 (full flowering), 75 (pea-sized berries) and 81 (veraison). The development of downy mildew was recorded regularly during the overall growth period with microscopy and molecular methods. Results show that infections occur with visible symptoms observed for the first three infection stages. At BBCH 55, infections resulted in the complete desiccation of the inflorescence. At full flowering, infections led to desiccation of parts of the clusters with some berries developing normally. At BBCH 75, typical brown rot symptoms appeared, while no infections were possible at veraison. These results are in agreement with field observations but differ from previous *in vitro* experiments. Such differences may result from the incubation conditions in the laboratory where detached bunches were observed during two weeks only. Microscopic observations showed that downy mildew invades almost all bunch tissues (rachis, pedicel, cap and berry including anthers, ovary and seed). Nevertheless mycelium and haustoria were never observed in vascular tissues (xylem and phloem). Based on these observations the hypothesis of downy mildew systemic development in clusters tissues is rejected. The same experimental design was applied to the downy mildew resistant grape variety Divico (=IRAC 2091, Gamaret x Bronner). Infections at early developmental stages (BBCH 55 and 65) resulted in sparse infections with very limited colonization. The rapid synthesis of high concentration of toxic stilbenes, such as d-viniferin and pterostilbene, may explain these results. Nevertheless, Divico is not totally immune and need a minimal protection during flowering and onset of the berries.

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BIOPHYSICAL MODELING OF PATHOGEN DISPERSION

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Researchers continue to develop empirical models to describe disease epidemics and aid growers in decision making. However, these models are often developed from datasets with limited environmental heterogeneity, and then used across a production region(s) with greater heterogeneity. This approach has been useful in the past, but is not likely optimal since regional differences in model performance have been observed. Recent advances in computation and computer sciences combined with an increased understanding of the biophysical interaction of the vineyard system could allow the development of highly spatially resolved disease risk estimates at the sub-block scale. The first step in such an approach is to develop an improved understanding of how terrain and canopy geometry affect the in- and above-canopy air turbulence and pathogen dispersion and deposition. To accomplish this, we have taken a two-prong approach where we use large-eddy simulations to examine the theoretical physical properties underpinning turbulent transport in trellised canopies and field experimentation to characterize air turbulence in vineyards and examine how particle plumes move throughout vineyards. The results of these studies are then used to build modeling environments that examine the probability of pathogen dispersion and deposition. This new information is then combined with three-dimensional plant growth models to examine the spatial and temporal distribution of disease development. The culmination of this research will be a vineyard simulation environment where growers can experiment with various vineyard practices before implementing in the field; similar to an engineering design process where parts are stress tested before being built.

EPIDEMIOLOGY AND POPULATION BIOLOGY OF GRAPE DOWNY MILDEW (*PLASMOPARA VITICOLA*) IN VINEYARDS IN GEORGIA, U.S.A.

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The state of Georgia (U.S.A.), with its hot and humid climate, comprises an extreme and diverse environment for growing wine grapes. *Vitis vinifera*, *V. aestivalis* and French-American hybrids are produced in the foothills of the Appalachian Mountains in the north, whereas *V. aestivalis*, hybrids, and muscadine grapes (*V. rotundifolia*) are grown in the Piedmont and Coastal Plains regions in the central and southern areas of the state. Most vineyards have been planted relatively recently and are geographically isolated from each other. Downy mildew, caused by *Plasmopara viticola*, is one of the most damaging diseases on *V. vinifera* and French-American hybrids throughout the state, but little is known about the epidemiology of the disease and population biology of its causal agent in this environment. Preliminary mating tests with single-sporangium isolates from an experimental vineyard in north Georgia indicate the presence of both mating types and the formation of oospore, although the oospore density in naturally infected leaf litter was low in that vineyard. Epidemic onset on foliage typically occurs in the late spring through early summer, and fruit infection is not commonly observed, presumably because fruit have reached the stage of ontogenic resistance by the time of disease onset. A statewide survey of 195 single-lesion isolates from 12 vineyards over 3 years revealed that 77.9% of isolates belonged to *P. viticola* clade *aestivalis* (Pva), whereas 22.1% (all from two vineyards in the Coastal Plains region) had a *P. viticola* clade *vinifera*-like (Pvv-like) genetic signature. Analysis of 130 of Pva isolates with seven microsatellite markers revealed high genetic diversity ($\hat{G}=0.98$), supporting a role of sexual overwintering. To further investigate the temporal dynamics of *P. viticola* populations and the contribution of sexual and asexual inoculum to the epidemic, 344 single-lesion isolates were collected in the experimental vineyard in northern Georgia from six temporal subpopulations between fall 2014 and fall 2016. Isolates were genotyped using the aforementioned seven microsatellite markers. There were 173 multilocus genotypes (MLG), of which 44 were identified more than once; the clonal population made up 62.5% ($n = 215$) of the total population. One dominant MLG was identified 57 times, representing 26.5% of the clonal population. On the other hand, 5 MLG were collected across different years. This study thus reveals high statewide genetic diversity of downy mildew populations. Both sexually- and asexually-derived inoculum contributes to epidemics at the vineyard scale. In addition, the same MLG observed across years indicates the possibility of asexual overwintering. We found only one cryptic species (Pva) in the northern part of the state, whereas two cryptic species (Pva and Pvv-like) are involved in epidemics in the Coastal Plains region. Additional research is needed to clarify asexual overwintering of the pathogen and to determine the factors affecting genetic diversity and distribution of cryptic species within and among populations in Georgia.

WHO CAN SWIM? A NEW APPROACH FOR DETERMINING SPORANGIA VIABILITY OF *PLASMOPARA VITICOLA*

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Plasmopara viticola is an obligate phytopathogenic oomycete causing downy mildew of grapevine. Sporangia, produced by the pathogen, release zoospores which swim in a film of water toward stomata where they enter and subsequently infect leaves and berries. Evaluating the viability of the pathogen's propagules enables us to estimate the effect of environmental factors or various treatments on infection risk by this important pathogen. Currently, the most common methods to test the viability of sporangia include inoculation tests and viability stains. Inoculation tests are laborious and time- and space-consuming. With viability staining, the percentage of viable sporangia can be quantified, but this method cannot quantify the time that zoospore remain viable, only limited sample numbers can be processed at one time, and stained sporangia cannot be re-used for inoculation. Therefore, a high-throughput method for determining viability of sporangia is needed to evaluate the ability of sporangia to release zoospores following various treatments. To overcome these limitations, a spectrophotometer-based method for determining sporangia viability is proposed and tested. Sporangia suspensions were inoculated on leaf disks of *Vitis vinifera* cv. Merlot, and sporangia produced at 4, 6, 10 and 17 days after inoculation (DAI) were collected to yield differences in sporangia viability. Sporangia suspensions were placed in a cuvette and their germination was monitored in a spectrophotometer at 2-min intervals for 5 h. The principle is based on the nature of zoospore release in the aqueous suspension of sporangia in the cuvette: absorbance will be high initially, but will decrease quickly as sporangia are settling within the suspension. As soon as zoospores are released and begin to swim, absorbance will increase and will remain high as long as the zoospores are swimming. When zoospores encyst, they will stop swimming and a decrease of absorbance will be detected. Thus, viability of sporangia (as measured by their ability to release viable zoospores) is related to the increase in absorbance following settling of the sporangia, as well as the duration of high absorbance reflecting swimming of zoospores. Absorbance started to increase after sporangia were suspended in water for ~30 to 60 min followed by major peak(s) for younger (4, 6, 10 DAI) sporangia, but no such increase was observed for 17 DAI (senescent) sporangia. Consistent patterns of sporangia germination demonstrated the results obtained with this method are reproducible. This method could provide new insights into basic biology and physiology of sporangia germination and will be useful to test the effect of various treatments on this important biological variable. In addition, this approach should be applicable to other oomycetes, fungi, or algae that germinate by releasing zoospores. Although further microscopic validation and statistical modeling are still needed, the concept presented here shows potential as a high-throughput technique for determining sporangia and zoospore viability and the timing of zoospore release.

DETECTION AND MONITORING OF FUNGICIDE RESISTANCE IN OREGON, WASHINGTON, AND CALIFORNIA *ERYSIPHE NECATOR* POPULATIONS

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Grape Powdery Mildew (GPM), caused by *Erysiphe necator*, can cause complete crop loss in the western U.S.A. if mismanaged. GPM control failures in 2015 lead to the exploration of the possibility of quinone outside inhibitor (QoI, FRAC Group 11) and demethylase inhibitor (DMI, FRAC Group 3) fungicide resistant *E. necator* presence in Oregon and California grape growing regions. GPM infected field samples were collected from vineyards in 2015 and 2016, yielding 87 *E. necator* isolates and 120 infected field samples for analysis. Of these isolates, 72% were resistant to QoI fungicides based on quantitative PCR (qPCR) analysis, which targets the G143A mutation in cytochrome *b* gene (*cytb*). These qPCR results were confirmed with a conidia germination bioassay on fungicide amended water agar. Sensitive isolates exhibited EC₅₀ values of 0.003-0.01 µg/ml to trifloxystrobin (Flint 50 WG) and 0.018-0.05 µg/ml to kresoxim-methyl (Sovran WG); resistant isolates exhibited EC₅₀s of >100 µg/ml to both fungicides. Of the infected field samples, 80% had detectable G143A mutation. Analysis of air samples collected using impaction spore samplers from commercial vineyards from 2013-2016 showed that QoI resistance was detected 2 years prior to control failures. Of 30 field samples collected from Southwestern Washington vineyards during May, 2017, all exhibited only the QoI resistant genotype based on qPCR analysis. In fields that experienced very high populations of QoI resistant GPM at the beginning of the 2016 growing season, and where QoI fungicides were not used in the 2016 growing season, the relative abundance of QoI resistant GPM dropped to near detection limits by the end of the growing season. Current 2017 sampling of *E. necator* from one field has not detected QoI resistance. This trend indicates that we may be able to restore the efficacy of these fungicides through fungicide rotation and monitoring. A leaf disk bioassay was used to determine the EC₅₀ of 30 single chain isolates for myclobutanil (Rally 40 WSP) based on disease severity. The EC₅₀s ranged from < 0.1 to > 16 µg/ml and were used to set discriminatory doses of 1 and 3 µg/ml for characterization of isolates as susceptible, moderate or resistant, respectively, to both myclobutanil and tebuconazole (Elite 45 WP/Toledo 45 WP). Based on this discriminatory dose bioassay, of the 46 isolates collected in 2015, 24% were characterized as moderately resistant and 56% as resistant to myclobutanil and 52% and 4%, respectively, to tebuconazole. Similar results have been seen for the 2016 isolates tested thus far. Sequencing of the *cyp51* gene only identified the Y136F mutant allele in moderate and resistance isolates. We are currently developing qPCR and Digital Droplet assays to detect and quantify the Y136F mutation.

OCCURRENCE OF GRAPE POWDERY AND DOWNY MILDEW IN WESTERN OREGON

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The Fruit and Ornamental Disease Management Testing Program has annually provided Oregon growers with relative efficacy of fungicides, fungicide schedules or cultural practices for control of plant diseases since 1989. Testing at the Botany and Plant Pathology Research Farm, Corvallis, OR occurs on many crops including 5 acres of vineyards established for grape powdery mildew (*Erysiphe necator*) or bunch rot (primarily *Botrytis cinerea*) management. Vineyards were managed for weeds, insects, cane or spur pruned and trained with vertical shoot positioning. Weather conditions were favorable for powdery mildew development such that non-fungicide treated plots frequently developed 100% incidence on leaves and/or clusters each year. Vineyards were surveyed intensively most years for the first occurrence of powdery mildew to determine fungicide program initiation. The first symptoms were recorded either as individual colonies, flag shoots (infected buds from the previous year) or both. The first symptoms did not always correspond to non-fungicide treated plots and occurred in multiple cultivars at the same time but were widely scattered through various vineyards. The date of first occurrence was as early as 26 April 2016 or as late as 30 June 1993. Powdery mildew occurred an average of 21 days prior to Pinot Noir bloom ranging from 1 to 45 days before bloom. Flag shoots occurred in 9 out of 22 years surveyed (from 1993 to 2016) and in 5 out of the last 6 years (2011 to 2016). Overall trends from 1993 to 2016 indicate that powdery mildew has occurred earlier in the calendar year, prior to Pinot Noir bloom and more often as flag shoots.

Downy mildew (*Plasmopara viticola*) has not been reported from grape grown in the Pacific Northwest. One unconfirmed OSU Plant Clinic specimen from a homeowner was diagnosed with downy mildew in 1966. Downy mildew was detected on Boston ivy (*Parthenocissus tricuspidata*) plants in Oregon in 2001. Subsequent surveys in 2002 found downy mildew on both Boston ivy and Virginia creeper (*Parthenocissus quinquefolia*) plants in wholesale and retail nurseries throughout grape-producing areas of Oregon. One landscape planting of Boston ivy, without overhead irrigation, had symptoms of downy mildew. This may indicate that western Oregon can sustain natural spread and expression of this disease.

Growers in western Oregon are advised to begin powdery mildew fungicide management programs well before bloom (no later than BBCH 57) especially when not monitoring for spores of the fungus. Chemical applications for grape downy mildew are unwarranted since the disease has not occurred in the PNW.

DETECTION OF *PLASMOPARA VITICOLA* OOSPORES IN WESTERN AUSTRALIAN VINEYARDS

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Sexual reproduction of *Plasmopara viticola* in European and American populations is known to be heterothallic. The historical absence of oospores in Western Australia (WA) was thought to indicate the presence of a single mating type. However, the detection of oospores in leaf samples collected in 2015 suggests that sexual reproduction is indeed occurring in WA, and that past absence may be the result of a population bottleneck, due to the recent introduction of the pathogen.

P. viticola was detected for the first time in commercial WA vineyards in 1998. Examination of cleared leaf discs infected with *P. viticola*, collected between 2001 and 2003, failed to detect oospores. In autumn 2015 vineyards in the Swan Valley and Margaret River regions of WA that had experienced downy mildew infections during spring 2014 were revisited. Whole leaves exhibiting mosaic symptoms were removed from randomly selected vines, cut into strips, hydrated overnight in warm water and examined using a compound microscope for the presence of oospores. Oospores were detected in leaf samples from all vineyards.

For confirmation that both mating types exist in WA, 11 isolates from geographically distant vineyards were paired. A 7 µL droplet containing 2.5×10^4 mL⁻¹ sporangia of each isolate was co-inoculated on each of six replicate leaf discs, within a petri dish containing moistened filter paper, and incubated at 20°C on the laboratory bench. After 5 weeks they were assessed for the presence of oospores. Three of the crosses produced oospores. As no reference isolates exist in Australia, specific mating type ratios could not be determined. This study has shown both mating types of *P. viticola* are present in WA and that the lack of oospores in the surveys conducted in earlier studies is likely the result of a population bottleneck in that both mating types had yet to become established in all growing regions.