The VISA network: a collaborative project between research institutes and vineyard owners to create the first epidemiological monitoring network of downy mildew epidemic based on aerial spore capture.

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Grapevine downy mildew, caused by the oomycete *Plasmopara viticola*, is one of the most devastating diseases of grapevine worldwide, especially in areas where climate conditions are more favourable to the disease development: intermediate to hot climate with sub-humid to humid conditions (Bois *et al.*, 2017). *P. viticola* is an obligate biotrophic pathogen with a known host-spectrum limited to *Vitis* spp. so far (Fontaine *et al.*, 2021), having direct effect on both bunches and leaves (oil spot symptoms, mosaic symptoms, berry dehydration) as well as indirect effect via defoliation. Grapevine downy mildew epidemics are frequent and can lead to serious yield losses, making this disease the first sanitary concern in grapevine owners in Europe (Bois *et al.*, 2017).

Since efficient preventive agronomic practices are still lacking, downy mildew epidemics are mainly controlled by fungicide applications. For example, in France, anti-downy mildew fungicide makes ~40 % of the total phytosanitary treatments of a vineyard on average (Chen, 2019). Regarding the negative effect of fungicide on the global biosphere health (*e.g.* Komárek *et al.*, 2010; Mercadante *et al.*, 2019; Raherison *et al.*, 2019), its use need to be reduced to its minimum tolerated dose. This reduction is accompanied by public policies (e.g. Ecophyto 2+) in order to find a multitude of solutions that would help reaching this goal while not jeopardising the actors of the sector. For example, it is necessary to be able to accurately characterise the epidemic development of mildew in order to apply phytosanitary products only when the risk is real.

P. viticola is heterothallic and alternate sexual and asexual cycles that both leads to the production of bi-flagellate zoospores from sporangia. Nonetheless, in the case of the sexual cycle, sporangia result from the germination starting in spring of oospores, *i.e.* an overwinter survival form resulting from the mating of the oogonium and antheridium of two different *P. viticola* individuals during the late summer, beginning of autumn of the previous season (Rossi, Caffi and Gobbin, 2013). Until recently, oospores germination - leading to primary infection - was thought to have a minor effect on the epidemic development compared to zoospores produced clonally after the first infection cycle and leading to secondary infection(s), as observed in some regions of the world (e.g. Rumbou and Gessler, 2006; Taylor *et al.*, 2019; Santos *et al.*, 2020). Nonetheless, the high level of genetic diversity found in *P. viticola* populations along the season of other regions, especially in western Europe, prove that primary infections can be the major source of the epidemic development (Gobbin *et al.*, 2005; Boso *et al.*, 2019; Hong *et al.*, 2020; Maddalena *et al.*, 2020; Santos *et al.*, 2020). Overall, wet conditions determine the production, transportation, and germination of zoospores into the host leaves (Rossi, Caffi and Gobbin, 2013). Aerial movement is also an important component of inoculum transports and have been correlated with epidemic development (Caffi *et al.*, 2013; Brischetto *et al.*, 2020).

Hence, epidemic risk prediction often relied on the modelling of infection caused by either or both inoculum source according to climate variables (*e.g.* Ronzon, 1987; Rossi, Giosuè and Caffi, 2009; Brischetto *et al.*, 2021). While these models are important to predict and understand the epidemic development in a region, their lack of accuracy at the plot scale limit their potential to adjust fungicide applications locally. Integrating airborne spore as a direct measure of the presence of the pathogen in the viticultural environment, before and during the epidemic development, is a promising approach to improve this risk prediction and adapt control strategies accordingly (Figure 1,Brischetto *et al.*, 2020). This approach was tested and validated using low-cost and field-transposable technique consisting in active spore capture followed by Loop-mediated isothermal Amplification and Quantification (qLAMP, Douillet et al., submitted). The same approach was then proposed to wine growers and advisors from the Bordeaux region vineyards during the vegetative season in 2021 to establish an epidemiological monitoring network based on spore capture. Indeed, we believe that synergizing the effort of data measurement at a regional scale appears to be a promising strategy to propose a global and accurate risk prediction.

In the next sections, we will answer the following questions: how does this participative research work and how does it improve research action? What information brings spore capture at the plot and on the regional level? How can this tool be adopted by vineyard owners in order to reduce fungicide consumption?

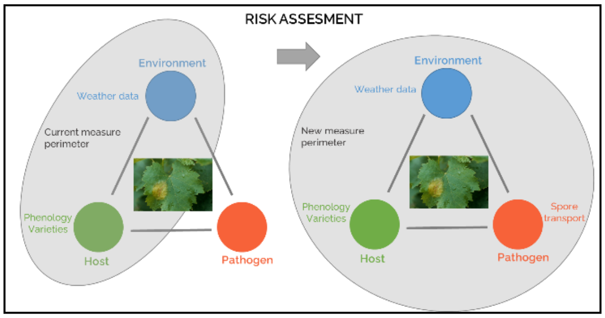

Figure 2: Toward a better epidemiological risk assessment by integrating spore capture as a proxy of pathogen presence in the field.

How does this participative research work and how does it improve research action?

Pimbert (2011) classified participatory research in agricultural research depending on their level of participation from a scale to 1 (passive participation) to 7 (self-mobilisation). According to this classification, the VISA epidemiological network is best associated to the fifth level called "functional participation" and defined as "people participating by forming groups to meet predetermined objectives related to the project, which can involve the development or promotion of externally initiated social organisation. Such involvement does not tend to be at the early stages of project cycles or planning, but rather after major decisions have been made. These institutions tend to be dependent on external initiators and facilitators but may become self-dependent". Indeed, the action was proposed and initiated by the research group with the proposition to winegrowers of a robust protocol elaborated from previous work (thesis works of A. Douillet) with the shared goal to reduce fungicide uses. A total of 25 partners accepted to buy and install the spore trap on their property, to collect spore samples 3 times a week, and made a semi-expert health observation of the grapevines near the spore trap area (between 10 to 100 plants) once a week. Whereas some wine growers had no difficulties in buying material in the absence of guaranteed return on investment, the price of this material was for others - especially for family-run structures - the main barrier to membership. This observation highlights the importance of public financial support to encourage the development of participatory research. Once the spore captures were analysed in the research institute by qLAMP (UMT SEVEN, INRAE Bordeaux-Aquitaine), the results were shared online to the entire network (Figure 2), respecting the anonymity of each point individually. A time for exchange was then organised bringing together the different stakeholders during a dedicated event (vine growers, advisors, research unit and technical institutes) in order to show research results and collect feedbacks from the fields. Overall, this first year of experimentation was conclusive, with a global adhesion of the partners, and revealing an interesting potential to reduce the use of fungicide. Concerning the research institutes, the collected data are precious resources to understand spore production and transport on regional scale, and will be the focus of further and deeper investigations. The network is already extending in the Bordeaux region and elsewhere with a goal of 100 partners in 2022. Quantifying *Erysiphe necator* spores - the causing agent of powdery mildew - and Guignardia bidwellii spores - the causing agent of Black rot - during the same qLAMP reaction is also scheduled.

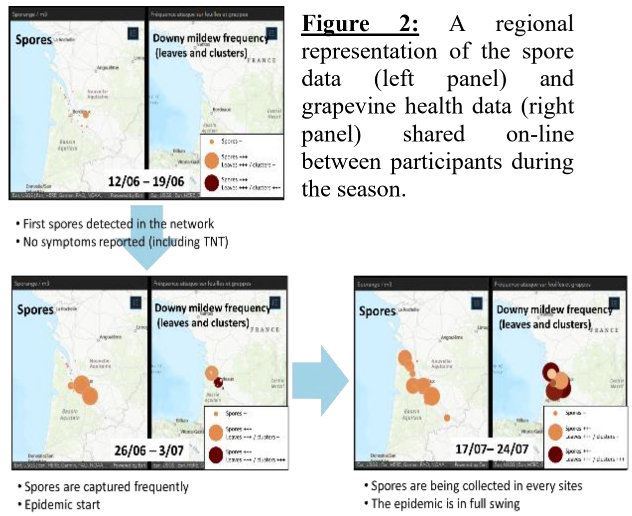

Figure 2: A regional representation of the spore data (left panel) and grapevine health data (right panel) shared on-line between participants during the season.

What information brings spore capture at the plot and on the regional level?

Spores were detected before symptoms apparition in 76% of the cases, with an average predictive length of 8 days for these anticipatory cases (*e.g.* Figure 3). The fact that spore capture is not systematically detected before symptom development highlights the importance of data sharing and monitoring network. Furthermore, informative data have been collected to study spore production and transport at the regional scale, in correlation to symptom development and climatic conditions, giving an overall picture of the epidemic dynamic in the region.

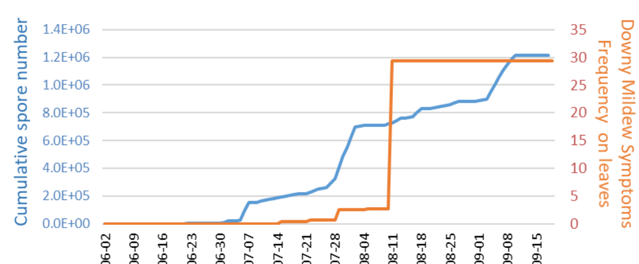

Figure 3: An example of field monitoring from a partner in Pessac-Léognan wine appellation along the 2021 growing season.

As seen on Figure 3, several periods of important spore production often occurred during the season, that was correlated with a delay with an increase of symptoms development. The implication of sexual vs clonal inoculum in the different phases of the spore capture dynamic is under investigation. Preliminary statistical analyse showed that numerous factors significantly influenced the quantity of spores captured at the regional level, including the type of vineyard management (organic, conditional, no treatment), wine appellations, climatic variables, seasonality, or the grape variety. However, all the factors tested explain a small part of the total variation, and other factors remain to be discovered. Of course, collecting data during several seasons and from different regions will strengthen the power of such analysis and will improve our knowledge on epidemic development.

How can this tool be adopted by wine grower in order to reduce their phytosanitary consumption?

The time from spore capture to qLAMP analysis and data sharing is critical to allow the wine growers a reaction time for their decision making. Publishing the results online in a rapid manner was not an objective for the 2021 campaign but is expected to be done three to four days after capture in 2022. Furthermore, the first partners will experiment – in collaboration with the UMT SEVEN – to add the spore capture data from the plot and from the network into an integrated decision support system for fungicide application that already included among other parameters a weather-driven disease risk prediction (Decitrait®) and field observations. A retrospective analysis of the 2021 growing season from an experimental plot of INRAE station (Bordeaux, Nouvelle-Aquitaine, France) showed that applying spore data into such decision rules would have saved ~30% of the total fungicide applied during this season.

In conclusion, combining several levers - including already established methods (*e.g.* weather-driven risk prediction) with new technological innovations, such as aerial spore monitoring, seems to be an effective strategy to achieve low-input production systems. Moreover, involving stakeholders in the development and the implementation of the research actions in the field is undeniably an important lever to accelerate the transition towards a sustainable viticulture.

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