

Portrait of the distribution of grapevine trunk diseases in Quebec, Canada, and consideration of the microbiome in their epidemiology.



CAROLINE PROVOST¹, PHILIPPE CONSTANT², and AUDREY-ANNE DURAND².

¹CENTRE DE RECHERCHE AGROALIMENTAIRE DE MIRABEL, 9850 Belle-Rivière, Mirabel, Québec, Canada, J7N2X8.

²INRS-ARMAND-FRAPPIER SANTÉ BIOTECHNOLOGIE, 531 Boul des Prairies, Laval, Québec, Canada, H7V 1B7.

<u>cprovost@cram-mirabel.com</u>

Institut national de la recherche scientifique

OBJECTIVES and CONTEXT

Grapevine trunk diseases (GTD) are very damaging for the sustainability of the vineyard heritage in all major wine regions of the world. A multitude of fungi are responsible for these diseases and attack the perennial organs of the vine, which causes the death of the plant in the short or medium term. GTD can affect young plantations as well as aging vineyards. In both cases, the risk of contamination by these diseases is increasing in Quebec vineyards, as many of them are more than 20 years old, and many producers plan to expand. However, the presence and distribution of GTD is unknown in Quebec. Moreover, climate change can promote the spread of these diseases, including higher temperatures and heavy rainfall. Knowledge of the etiology and epidemiology of MBV is necessary to develop control methods against these diseases. Knowledge of infection times in combination with the development of management strategies using appropriate cultural practices, such as 'replacement surgery', and the application of fungicides or biological control agents to pruning wounds (when registered) are needed to control MBV in vineyards (Bertsch et al. 2013; Gramaje et al. 2018).

The objective was to acquire knowledge on the etiology and epidemiology of grapevine trunk diseases in the vineyards of Quebec, Canada, according to different criteria, such as grape varieties, cultural practices and age of the vineyards.

In total, 203 samples from 46 vineyards were received and analyzed. The age distribution of the plots concerned is 56 plots (71%) over 9 years old (older plots) and 23 plots (29%) under 8 years old (young plots). We find 68% of the plots sampled under conventional management and 32% under organic. A total of 30 grape varieties were sampled, including the 12 main grape varieties found in Quebec (mainly hybrids).

- The results demonstrate that both young vine plots and older plots are affected by GTD (p > 0.05) (Tab. 1). The prevalent GTD in young plots is Esca with the fungus *P. aleophilum*, followed by *D. seriata* causing Botryosphaeria dieback (Tab. 1). The three GTD found in older vineyard plots are Botryosphaeria dieback (*D. seriata* and *B. dothidae*), Esca (*P. aleophilum*) and eutypiosis (*E. lapa*).
- The different GTD are found in all the main wine-growing regions of Quebec (Fig. 1, 2). Esca is Quebec's most widespread GTD, covering all producing regions from east to west (Fig. 1). The Botryosphaeria dieback is found throughout the Quebec wine-growing region except for the Ottawa Valley (Outaouais) (Fig. 2). It is common to detect more than one fungus causing Botryosphaeria dieback on the same site.
- Of the 79 plots sampled, 64 were infected with at least one GTD, representing 81% of the samples. It should, however, be mentioned that plots showing signs of decline were targeted.
- The cultural practices did not demonstrate a significant effect on the presence of GTD (p > 0.05) (Tab. 1). The four main GTD were detected both in organic and conventional vineyards.
- Detection of GTD was similar in different parts of the vine.
- The most sampled grape varieties were Frontenac, Frontenac blanc, Ste-Croix, St-Pépin and Marquette. Frontenac, Frontenac blanc and Marquette were greatly affected by GTD with several detections of fungi. It has also been observed that *Vitis vinifera* seems more affected than hybrid grape varieties. Indeed, one to two samples were collected for Chardonnay, Merlot, Pinot gris and Pinot noir, and there were at least 3 GTD, and even 4 in each sample.
- The prevalence of two genera of bacteria (*Oxalicibacterium* spp. and *Salinibacterium* spp.) was found to be correlated with tissues where no *E. lata* infection was observed (Fig. 3). For *D. seriata*, twelve genera of bacteria were prevalent in the absence of infection (Fig. 3). Genotypes affiliated with the genus *Salinibacterium* spp. were prevalent in the absence of the two phytopathogenic fungi.

METHODS

The project was divided into two parts: 1) an assessment of the presence and distribution of GTD across Quebec was carried out according to various criteria (region, cultural practices, grape variety, age of the vineyard, and part of the plant); and 2) the characterization of microbiome related to the epidemiology of GTD.

We collected 79 samples from 46 vineyards across Québec, and detection of these GTD were executed: Esca (*Phaeomoniella chlamydospora*, *Phaeoacremonium* sp.), Botryosphaeria dieback (*Botryosphaeriaceae* sp., *Diplodia* sp., *Neofusicoccum* sp.), excoriosis (*Diaporthe ampelina*), Eutypa dieback (*Eutypa lapa*). For each sample, three plant parts were collected: 1) part of a lignified arm (cord), 2) a shoot of the year (cours/shoot), and 3) part of the bark of the trunk with canker. Then, a total of 203 samples were processed. Results were analyzed considering the following parameters: 1) the grape varieties; 2) age classes of vineyards (young vs mature); 3) cultural practices (organic or conventional); and 4) the part of the plant.

Genomic DNA samples extracted from vine samples subjected to vine wood disease detection tests were used to portray the vine microbiome. The composition of microbial communities was determined by PCR amplicon sequencing targeting the 16S rRNA of bacteria. The profile of microbial communities was linked to the distribution of *D. seriata* and *E. lata*.

RESULTS

Table 1: Distribution of samples infected by different vine fungi.

		Organic		Conventionnal	
Disease	Fungi	< 8 years old	> 9 years old	< 8 years old	>9 years old
Botryosphaeria dieback	Botryosphaeria dothidae	0	1	4	10
	Diplodia seriata	7	5	7	18
	Neofusicoccum parvum	1	0	1	9
Eutypa dieback	Eutypa lata	4	9	5	19
Esca	Phaeoacremonium aleophilum	8	6	11	26
	Phaeomoniella chlamydospora	1	0	0	1
Excoriosis	Diaporthe ampelina	1	4	5	7
Total number of plots tested positive		10	13	15	41
Total number of positive vineyards		6	10	10	28

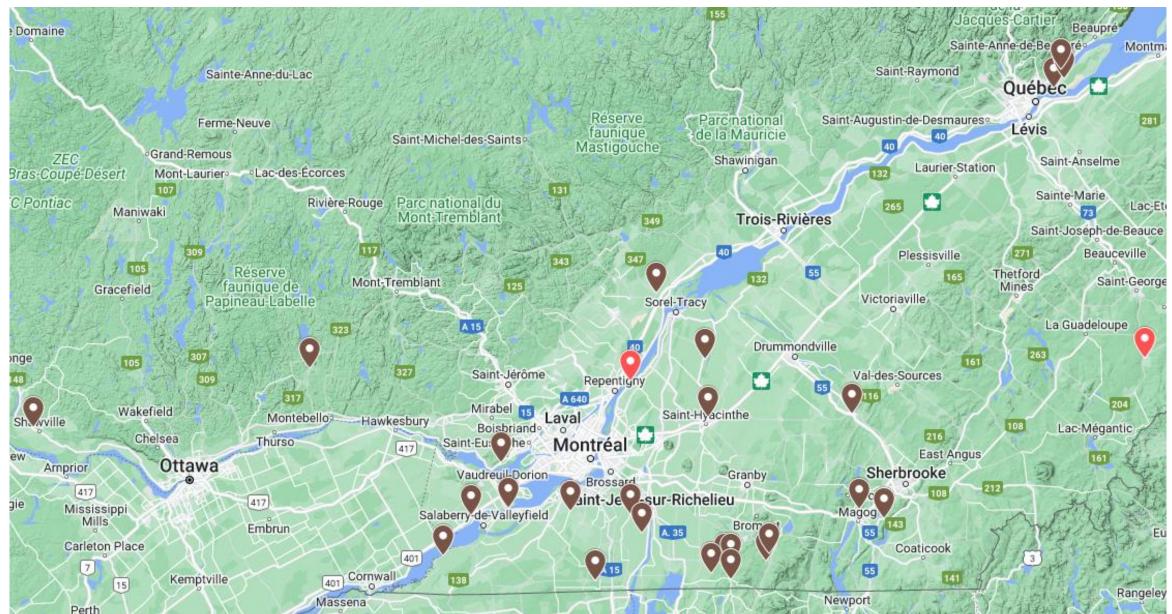




Figure 1: Distribution of Esca in Québec vineyards (*P. chlamydospora* (pink), *P. aleophilum* (brown)).

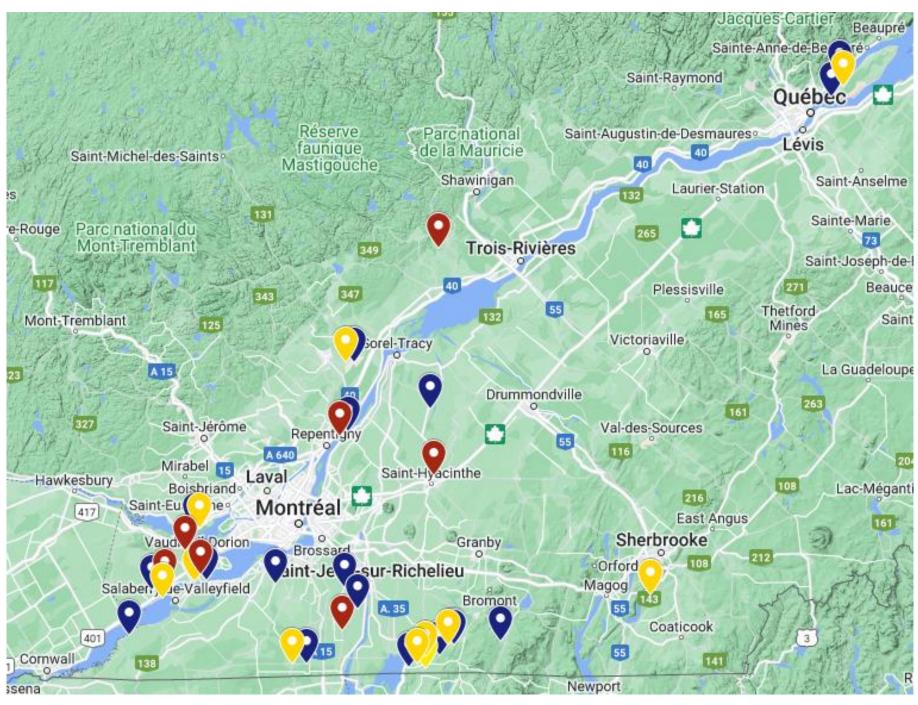


Figure 2: Distribution of Botryosphaeria dieback in Québec vineyards (*B. dothidae* (yellow), *N. parvum* (red), *D. seriata* (blue)).

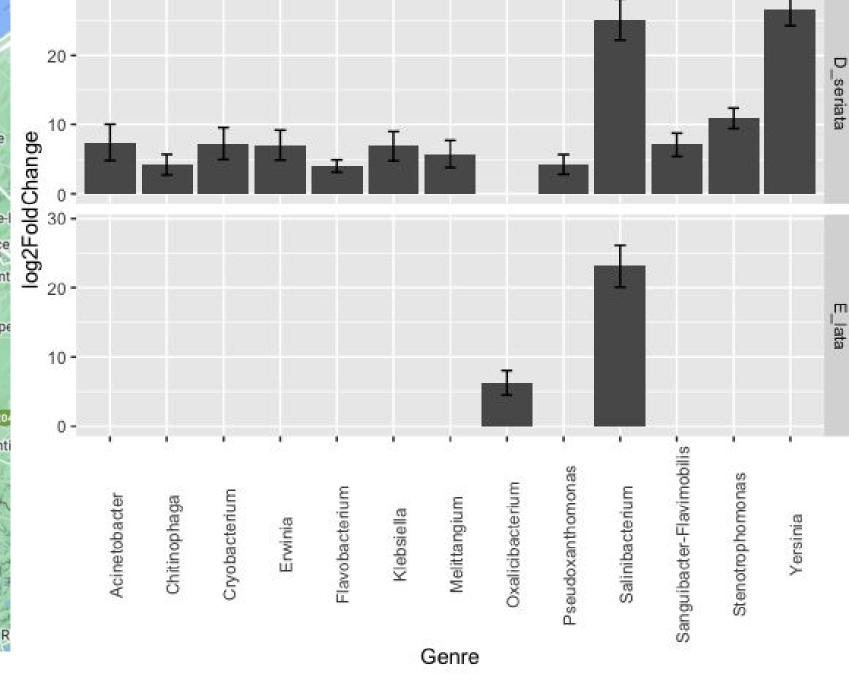


Figure 3: Phylotypes of bacteria grouped at the taxonomic rank of the genus for which the relative abundance was higher in healthy vines compared to vines having demonstrated a positive diagnosis for *D. seriata* and *E. lata*.

CONCLUSIONS

- This portrait allows us to mention that GTD are indeed present in Quebec wine-growing territory and must be considered in the context of cultural practices.
- The presence and distribution of GTD in vineyards demonstrate that their presence is linked to sources external to the vineyard.
- The microbiome of grapevine offers new opportunities to predict the distribution and control GTD.

Acknowledgement

The authors wish to thank Stefano Campagnaro and Pascale Boulay for the technical support, and Louis Thomas, Conseil des vins du Québec. This project is funded through the Innov'Action Agroalimentaire program, under the Canadian Partnership for Agriculture, an agreement between the governments of Canada and Quebec.



References

Bertsch, C., Ramírez-Suero, M., Magnin-Robert et al. (2013). Grapevine trunk diseases: complex and still poorly understood. *Plant Pathology*, 62(2), 243-265.

Gramaje, D., Urbez-Torres, J. R., & Sosnowski, M. R. (2018). Managing grapevine trunk diseases with respect to etiology and epidemiology: current strategies and future prospects. *Plant disease*, 102(1), 12-39.