



## **Towards microbiota-based disease management: analysis of grapevine microbiota in plots with contrasted levels of downy mildew infection**

Paola Fournier, Aarti Jaswa, Lucile Pellan, Jessica Vallance, Emilie Chancerel, Olivier Bonnard, Isabelle Masneuf-Pomarede, Guilherme Martins, Marc Raynal, Christian Debord, et al.

### **► To cite this version:**

Paola Fournier, Aarti Jaswa, Lucile Pellan, Jessica Vallance, Emilie Chancerel, et al.. Towards microbiota-based disease management: analysis of grapevine microbiota in plots with contrasted levels of downy mildew infection. 22nd GiESCO (Group of international Experts for Cooperation on Vitivinicultural Systems) 2023, Jul 2023, Ithaca, United States. hal-04180613

**HAL Id: hal-04180613**

**<https://hal.science/hal-04180613>**

Submitted on 13 Aug 2023

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



# Towards microbiota-based disease management: analysis of grapevine microbiota in plots with contrasted levels of downy mildew infection

P. Fournier<sup>1,2,3</sup> and A. Jaswa<sup>1,4</sup>, L. Pellan<sup>1</sup>, J. Vallance<sup>1</sup>, E. Chancerel<sup>2</sup>, O. Bonnard<sup>2</sup>, I. Masneuf-Pomarede<sup>4</sup>, G. Martins<sup>4</sup>, M. Raynal<sup>5</sup>, C. Debord<sup>5</sup>, S. Labarthe<sup>2</sup>, L. Deliere<sup>1</sup>, F. Delmotte<sup>1</sup>, P. This<sup>3</sup>, C. Vacher<sup>2</sup>

<sup>1</sup>INRAE, Bordeaux Sciences Agro, ISVV, SAVE, 33140 Villenave-d'Ornon, France. <sup>2</sup>INRAE, Univ Bordeaux, BioGeCo, 33610 Cestas, France. <sup>3</sup>INRAE, CIRAD, Univ Montpellier, Institut AGRO, AGAP institut, 34398 Montpellier, France. <sup>4</sup>Univ Bordeaux, UMR oenologie, INRAE, Bx INP, Bordeaux Sciences Agro, ISVV, 33882 Villenave d'Ornon, France. <sup>5</sup>IFV, 33290 Blanquefort, France.



Contact: [paola.fournier@inrae.fr](mailto:paola.fournier@inrae.fr)  
[aarti.jaswa@inrae.fr](mailto:aarti.jaswa@inrae.fr)

## 1. Context and hypotheses

Vineyards are colonized by a myriad of microorganisms that interact with each other and with grapevine plants<sup>1,2</sup>. Some microorganisms are plant pathogens, such as the oomycete *Plasmopara viticola* that causes grapevine downy mildew. Others have a positive influence on vine health, such as plant growth promoting bacteria and disease biocontrol agents. These beneficial interactions have gained more attention in recent years because they could represent an alternative to the use of fungicides in viticulture<sup>3</sup>.

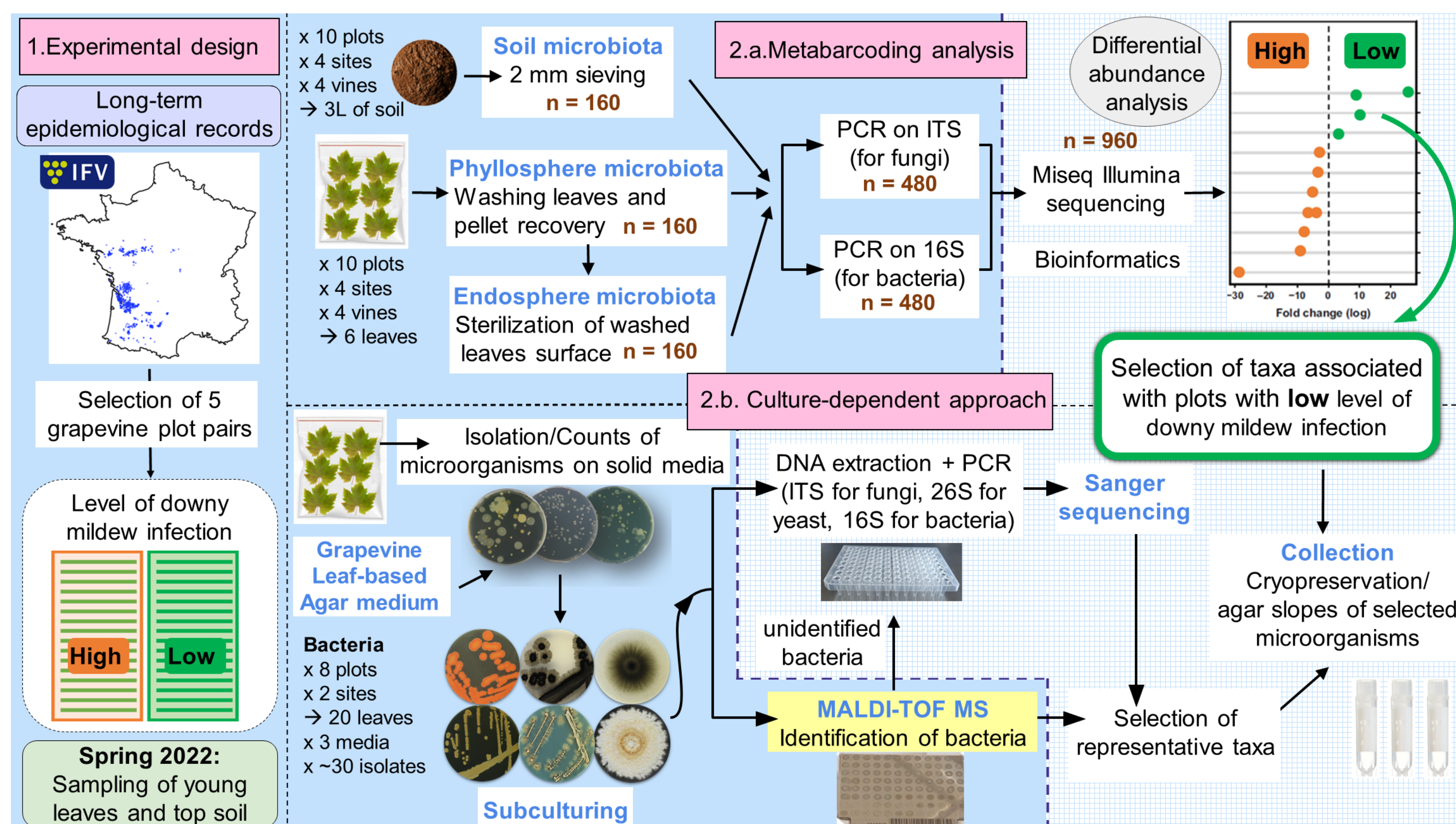
To develop microbiota-based disease management, we aim to cultivate the myriad of microorganisms that colonize grapevine leaves and then assemble multi-strain microbial consortia that could protect vines against *P. viticola*. To increase the probability of isolating microorganisms of interest, we developed a plant-based culture medium. We tested the hypothesis that it increases the cultivable microbial diversity in comparison to standard culture media.



Downy mildew  
*Plasmopara viticola*  
Oomycete

## 2. Materials and methods

We combined metabarcoding (Fig.1(2.a)) and culture-dependent (Fig.1(2.b)) approaches to analyze the microbiota of soil and leaf samples collected in plots with contrasted levels of downy mildew infection.



**Fig.1:** Workflow to characterize and isolate bacterial and fungal communities naturally colonizing vineyard soil and leaves in plots with contrasted levels of downy mildew infection. Media used for bacteria: Tryptic Soy Agar (TSA), Luria Bertani Broth Agar (LBA) and a Grapevine Leaf-based Agar medium (GLA).

### Bibliography

1. Bettenfeld *et al.*, 2021. *J. Adv. Res.*
2. Fournier, Pellan *et al.*, 2022. *Adv. Ecol. Res.*
3. Busby *et al.*, 2017. *PLOS Biol.*

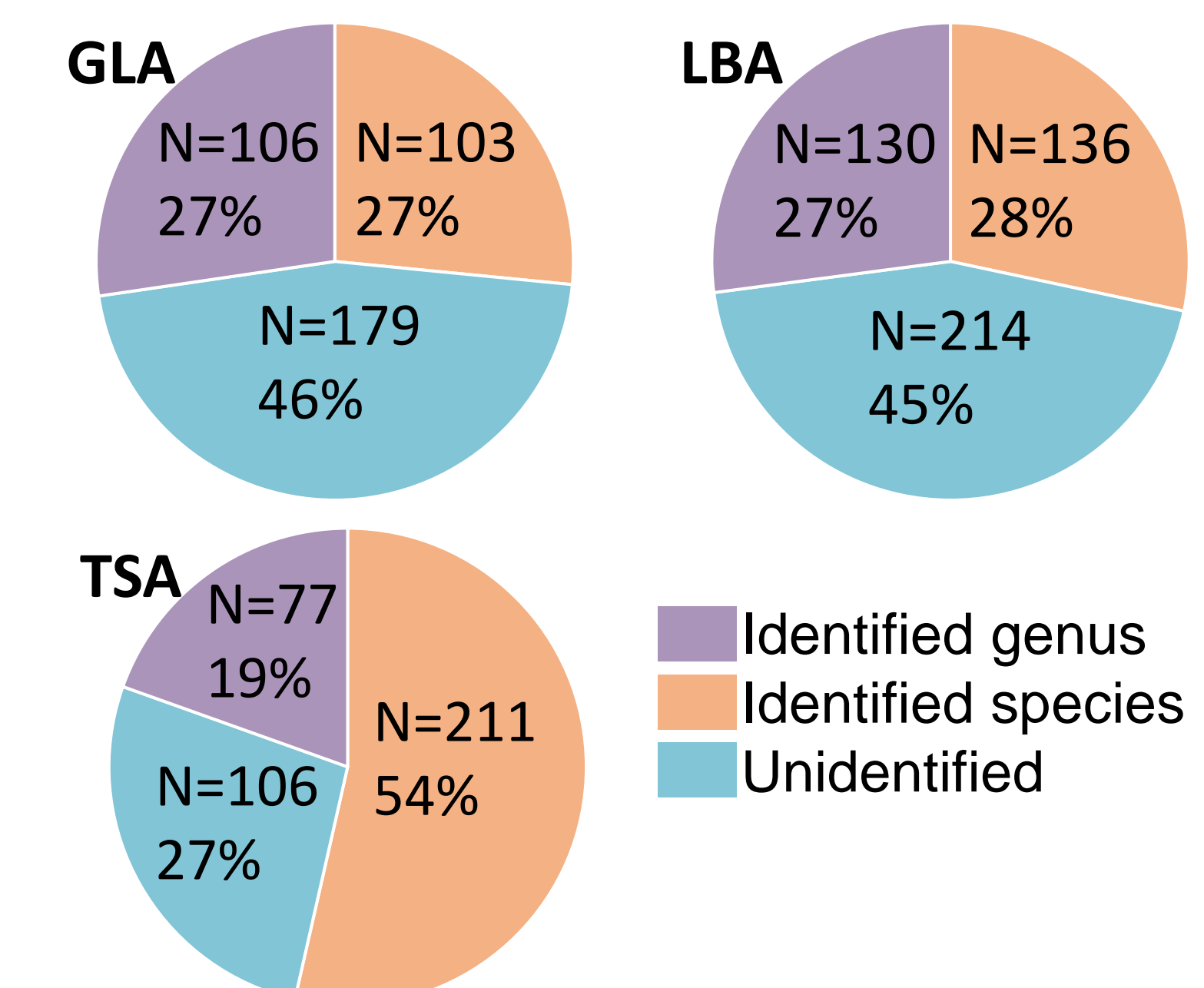
### Funding

This work is part of the VITAE project ('Cultivating grapevine without pesticide') funded by the French National Research Agency (ANR, 20-PCPA-0010).

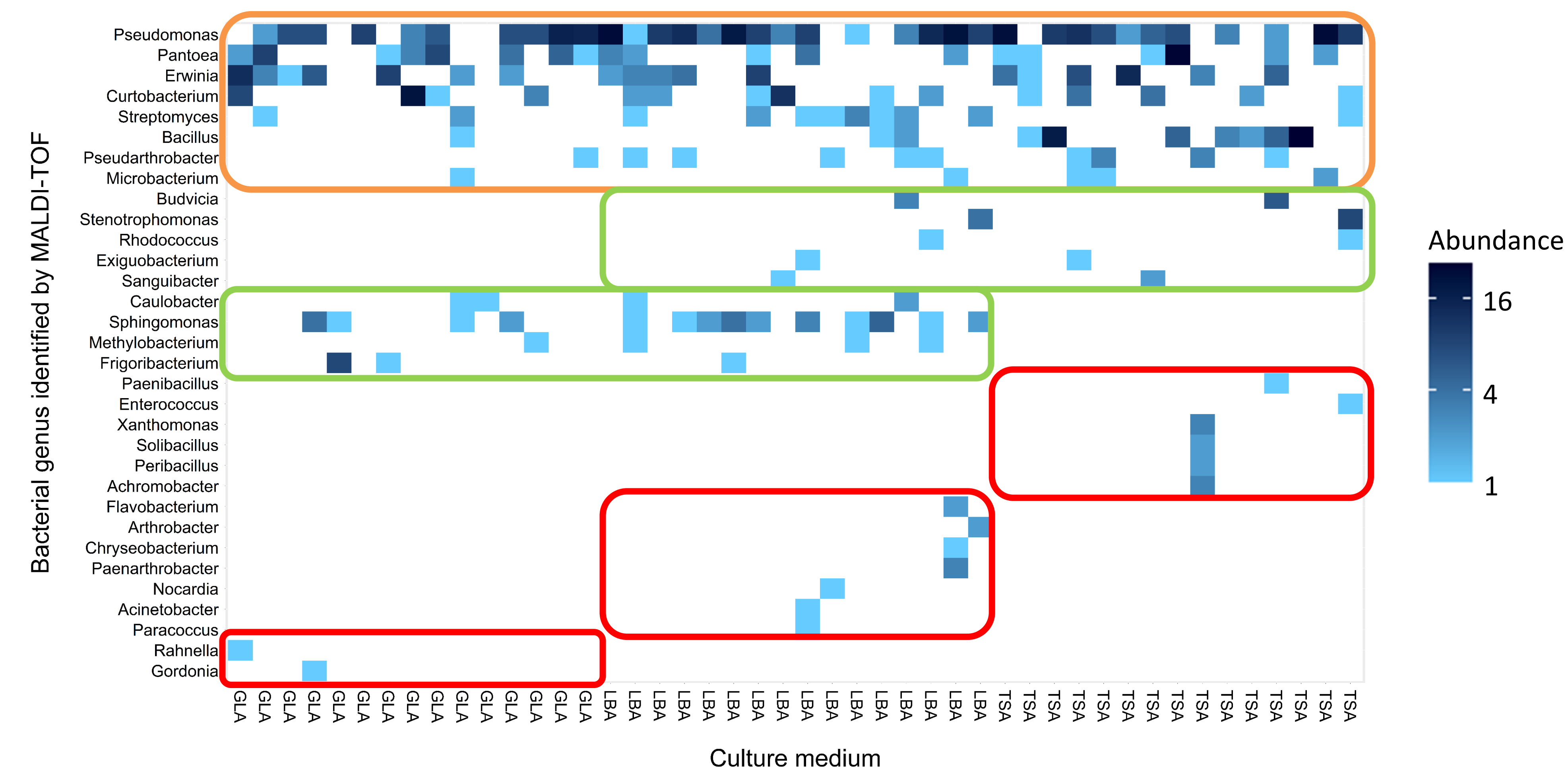
## 3. Results and discussion

### MALDI-TOF MS Identification of bacteria

- A total of 1262 bacterial isolates were analyzed by MALDI-TOF, of which 60% could be identified at least to genus level (Fig.2).
- Strains isolated from GLA and LBA were less well assigned than those isolated from TSA (higher % of non-identification and lower % of assignment to the species level) (Fig.2).
- A total of 32 bacterial genera were identified on the three culture media. Eight genera were isolated on all three media (Fig.3).
- The genera *Gordonia* and *Rahnella* were isolated only on GLA (Fig.3).



**Fig.2:** Number of bacterial strains isolated on each culture media and percentage of taxonomic assignments by MALDI-TOF MS.



**Fig.3:** Bacterial genera identified using MALDI-TOF MS in grapevine leaf samples, depending on the culture media (GLA, LBA and TSA). Abundance is the number of bacterial isolates belonging to each genus. Bacterial genera identified in only one, two or three culture media are circled in red, green and orange, respectively.

## 4. Conclusion and perspectives

We were able to cultivate and isolate >1000 bacterial strains from grapevine leaves. Diversifying the culture media allowed us to isolate more strains. After completing the Sanger sequencing of all microbial isolates (including yeasts and filamentous fungi; Fig. 1 (2.b)), we will analyze the complementarities between the grapevine leaf-based culture medium and standard culture media. We will test the hypothesis that the vine-based medium selects for microbial taxa that are different from those selected on standard media and/or poorly referenced in the current version of the MALDI-TOF MS database. Statistical analysis of metabarcoding data (Fig. 1 (2.a)) will provide an estimate of the % of cultivable taxa and will allow us to identify microbial taxa that are significantly more abundant in vineyard plots with low levels of downy mildew infection. Those microorganisms will be retrieved from our microbial collection. Their effect on *P. viticola* will be tested in vitro and in planta, as single strains and as consortia. Modes of action will be elucidated for the most promising microbial consortia (those that thrive on grapevine leaves, are resilient to disturbances and have a biocontrol activity against *P. viticola*).