Virome characterization of *Diplodia sapinea* isolates in France

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CONTEXT & OBJECTIVES

Diplodia sapinea is a fungus, widely distributed in the world, which can colonize as an endophyte different organs of several pine species. However, it can become pathogenic under different stresses and induce an emerging disease called Diplodia shoot blight (Fig.1). Two mycoviruses, *Sphaeropsis sapinea* RNA virus 1 & 2 (SSRV1 & 2), have been described in *D.sapinea* in a few isolates [1]. Their prevalence in French populations of *D.sapinea* is not known, nor is it known whether they can decrease disease severity.

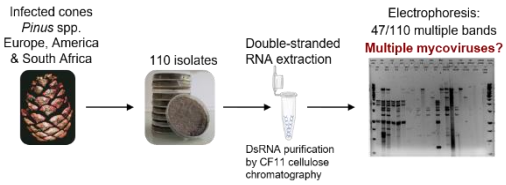
Our objective:

Screening a D. sapinea collection for mycoviruses to identify biological control candidates.

Fig.1: Symptoms on Salzmann pine following infection by *D. sapinea*

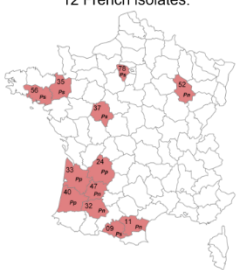
METHODS

1 Screening collection for mycoviruses



2 Selection for sequencing

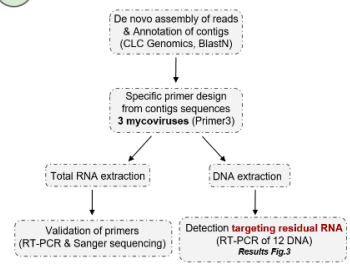
12 French isolates:



Illumina Novaseq 2x150 bp
10M reads / sample
Results Fig.2

Host species:
Pn: Pinus nigra
Pp: Pinus pinaster
Ps:Pinus sylvestris

3 Design of primers for detection



RESULTS

- Confirmation of multiple infections except for one isolate (Ds-24).
- SsRV1 & 2 detected in 4 isolates.
- **More than 20 new mycoviruses:** dsRNA, ssRNA+ and ssRNA- genomes.
- Mycovirome variability (between and within isolate).

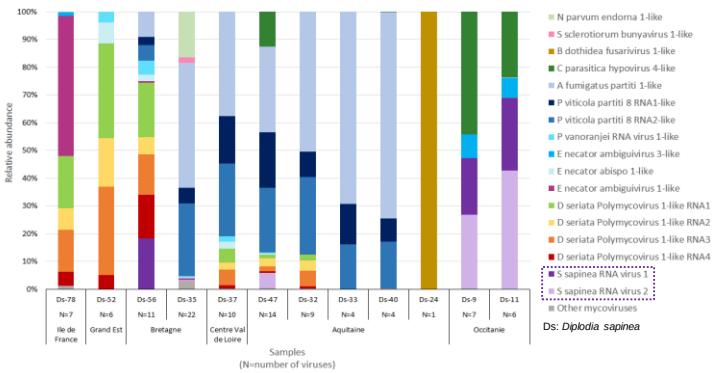


Fig.2: Relative abundance of each mycovirus in the 12 isolates
(abundance= number of specific viral reads / total number of viral reads)

- Successful amplification of targeted viral sequences with specific primers allows detection of mycoviruses from fungal DNA extractions.

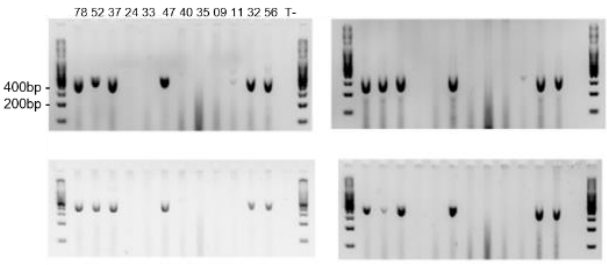


Fig.3: RT-PCR of 12 isolates
with primers targeting *D.seriata Polymycovirus* RNA1 to RNA4
(cDNA from DNA samples)

PERSPECTIVES

- Screening the rest of the collection with specific primers to study prevalence and diversity of mycoviruses.
- Studying phenotype and virulence of these mycoviruses.

Reference: [1] E. T. Steenkamp, B. D. Wingfield, W. J. Swart, and M. J. Wingfield, "Double-stranded RNA and associated virulence in South African isolates of Sphaeropsis sapinea," Can. J. Bot., vol. 76, no. 8, pp. 1412–1417, 1998, doi: 10.1139/cjb-76-8-1412.

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