

NATURAL SPREAD OF MYCOVIRUSES A PUZZLING AND DAZZLING ISSUE FOR BIOLOGICAL CONTROL OF CHESTNUT BLIGHT BY CRYPHONECTRIA HYPOVIRUS (CHV1)

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Introduction

Mycoviruses are widespread viruses in filamentous fungi considered ubiquitous in all groups. *Cryphonectria hypovirus 1* (CHV1) of the Hypoviridae family is included in the genus Hypovirus. Hypovirus are well-studied biological control agent of chestnut blight a lethal disease of *Castanea sativa* Mill. Unlike many mycoviruses, which are cryptic or latent in their host, CHV1 reduce virulence, reduce pigmentation and sporulation and induce female sterility in its host the *Cryphonectria parasitica* fungus. Chestnut blight was introduced in Portugal and rapidly became a severe lethal disease in all chestnut regions in the 90's. Initial epidemics had a very rapid spread and no or few cases of hypovirulence was reported. Besides scientific issues are obtained therapeutic hypovirulence was introduced as a biological control to mitigate disease impact. Later, in 2013 chestnut blight appears in scattered stands of the coast north Minho region where natural spread is the dominant way of hypovirulence spread. In this study we investigate and specifically addressed the question of the massive natural spread of hypovirulence.

Materials and methods

We studied vegetative compatibility system (vic genes) of the host fungus, presence and characterization of CHV1 subtypes, hypovirus transmission capacity issues that play a key role in hypovirus transmission.

Results and Conclusions

Isolates

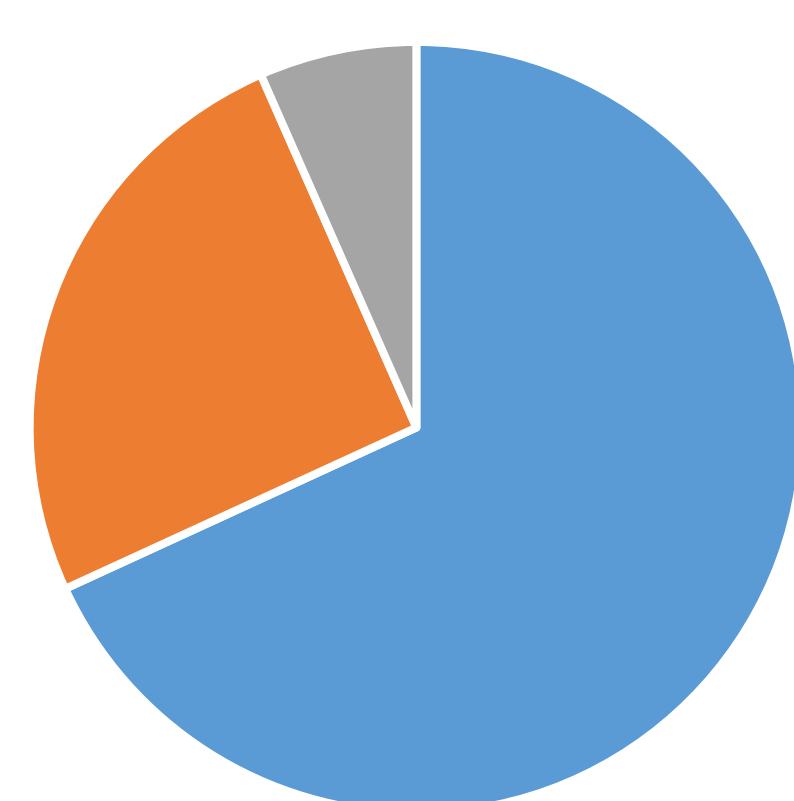


Felgueiras orchard

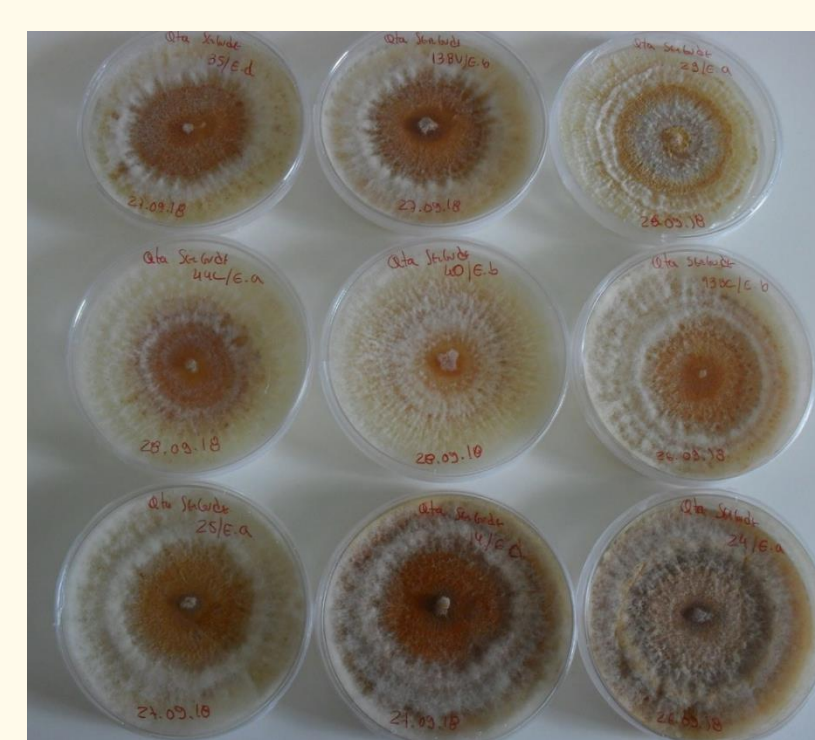


Felgueiras Healed canker

Isolates of *Cryphonectria parasitica*



Isolates of *Cryphonectria parasitica* obtained in Felgueiras



C. parasitica isolates



Orange isolates Intermediate isolates White isolates

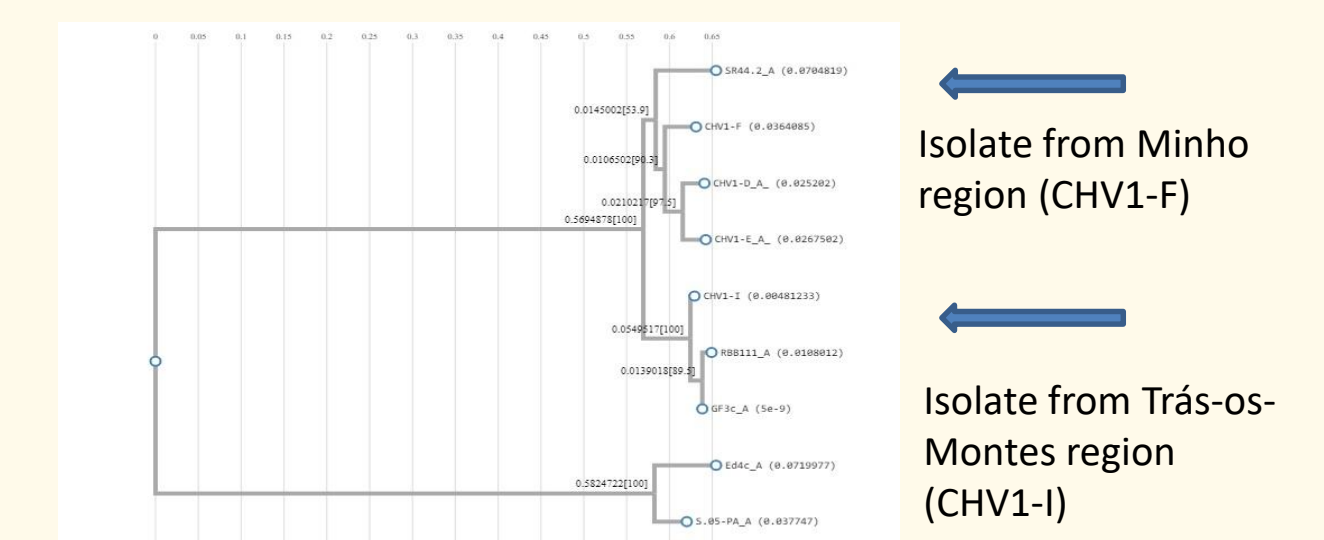
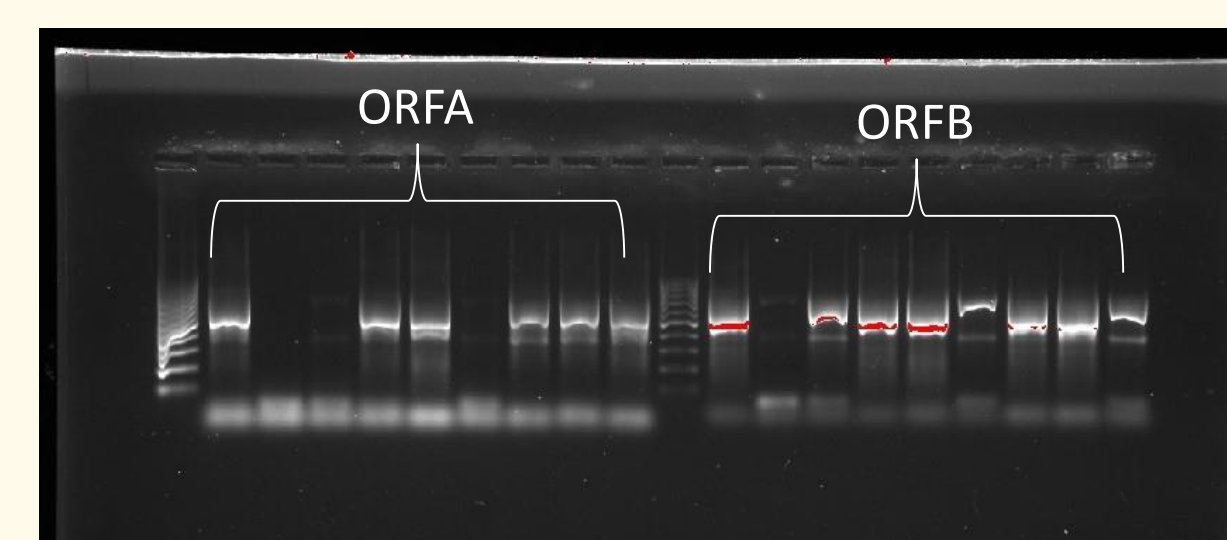
Vc types determination



Vc types	EU01	EU02	EU11	EU12	EU66
Isolates	0	0	91	0	0

All isolates belonged to vc type EU11

CHV1 hypovirus detection



Sequencing of the PCR product of the isolates indicates that CHV1 is of the French type (CHV1-F), and is different from the hypovirulent isolates found in the Trás-os-Montes region (Italian type).

The success of Hypovirulence is determined by the fitness of both members of the system (*C. parasitica* and hypovirus CHV1) as well as by diversity of fungal vegetative compatibility (vc) types, which impedes the spread of the virus through *C. parasitica* populations. Vegetative compatibility types in the studied site (Minho) are included in the same group (EU11) and the hypovirus French subtype F1 was identified. In Trás-os-Montes, where natural spread of hypovirulence is not frequent, Italian subtype I is dominant even although the dominant vc type is also the EU11.

Major key determinants and driven forces to improve natural hypovirus transmission are still unclear and new approaches as (HTS) high throughput sequencing will be valuable to understand the driven forces of natural dissemination that will greatly increase field sustainability of therapeutic applications of hypovirulence and chestnut recovered.

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