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Mycoviruses: Potentiality to Use as Bio-control Agents

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SUMMARY

Pesticidal resistance and environmental pollution have arisen as a result of chemical pesticides in modern agriculture. Biocontrol is the alternative strategy that provides substantial and consistent disease management. Mycoviruses from plant pathogens have attracted a lot of scope by inducing hypovirulence (lower virulence) in their host fungi. The application is greatly dependent on their ability to spread in the natural populations of the pathogen. Mycovirus are transmitted horizontally via. protoplastic fusion (anastomosis) and vertically via sporulation. During virus and fungus interaction reprogramming of host metabolism, signalling pathways, cellular processes and RNA silencing takes place to overcome the virus infection. Contrarily, virus-encoded RNA silencing suppressors (RSS) employ distinct mechanisms to suppress host defense. Much more research is needed to understand the molecular interactions between mycovirus and fungi to be used successfully in the field.

INTRODUCTION

The word Mycovirus is derived from ancient Greek word 'mykes' (mushroom/ fungus) and Latin word 'virus'. These are the viruses that infect fungi or fungi-like organisms. Fungi belonging to Ascomycota, Basidiomycota, Chytridiomycota, Deuteromycota, and Zygomycota have all been reported to harbour mycoviruses (Ghabrial et al., 2015). The first instance of a mycovirus infecting a cultivable mushroom (*Agaricus bisporus*) and causing a La France illness or X disease was described by Hollings in 1962. Disease is characterised by slow and aberrant growth of mycelia, water-logging of tissues, malformation, shortened shelf-life and decreased sporulation. Most of the reported mycoviruses consist of linear double-stranded RNA (dsRNA~70%), while negative single-stranded RNA (-ssRNA) or positive single-stranded RNA (+ssRNA), rarely mycoviruses with circular single-stranded DNA (ssDNA) were also found (Yu et al. 2010; Jiang et al. 2013; Li et al. 2020).

There have also been reports of mycoviruses that induce both hyper- and hypo-virulence. In recent years, viro-control, or the biological management of phytopathogens employing viruses that cause hypovirulence, has drawn more attention. In Europe, *Cryphonectria parasitica*, a chesnut blight pathogen has been successfully managed under field conditions. Several mycoviruses have been shown to cause hypo-virulence to several plant pathogenic fungi. Mycoviruses such as MoV1, MoV2 and MoCV1 (*Magnaporthe oryzae*), Rhizoctonia virus M2 (*Rhizoctonia oryzae*), FgV-1, 2, 3, 4 and ch9 (*Fusarium graminearum*) and PiRV-1, 2, 3 and 4 (*Phytopthora infestans*) were found effective in reducing the virulence of pathogens.

Transmission

In addition to inducing hypo-virulence, mycoviruses requires an ability to infect a large proportion of the pathogen in order to be suitable biological control agent. This is depending on how efficiently they are transmitted to uninfected isolates through hyphal anastomosis, converting them from virulent to hypovirulent. They follow different modes of transmissions. In horizontal transmission, the viruses are transmitted among individuals of same generation either through anastomosis or through protoplasmic fusion. But it has few limitations like, 'Vegetative in-compatibility' among different strains of same fungal species. Where vegetative compatible isolates are able to establish successful hyphal anastomosis and the interaction between vegetative in-compatible isolates triggers localized programmed cell death (PCD), which hampers virus transmission (Glass and Dementhon 2006).

The viruses transmitting among different vegetative groups have been recorded whereas, the mechanism is not clear. This type of transmission has been observed in *C. parasitica* infected with *Cryphonectria parasitica* hypovirus-1 (CHV-1). Another way of transmission is vertical transmission**,** where the viruses are transmitted through asexual spores or through sexual spores. The transmission through sexual spores is less effective. Majority of sexual spores are virus-free, while asexual spores of the same species proven to be infected. The exclusion of mycovirus from sexual spores is in line with the selection arena hypothesis, this is to exclude deleterious elements and mutations from the germ line of fungus (Bruggeman et al. 2004). The exclusion of

dsRNA segments from sexual spores of *Ophiostoma nova ulmi* has been reported. Extra-cellular transmission by mycoreovirus and mycophagous insect transmissions (SsHADV by *Lycoriella ingenue* and CHV-1 by *Aphelenchoides spp*) are also recorded.

Mycovirus-fungal interaction

Mycoviral accumulation levels are expected to depend primarily on the host antiviral defense and how virus counteracts them. During virus infection, RNA silencing (RNAi) through post-transcriptional gene silencing (PTGS) system plays a vital role. The process is triggered by the presence of dsRNA that is processed by Dicer or Dicer-like (DCL) proteins into small interfering RNAs (siRNAs). One strand of the siRNA is loaded into an RNA-induced silencing complex (RISC), where the Argonaute or Argonaute-like (AGL) proteins play a central role. The siRNAs guide the RISC to the target RNAs, cleaving them. RNA-dependent RNA polymerases (RDRPs) convert single-stranded RNA (ssRNA) into dsRNA thus amplifying the siRNA signals.

The viruses counteract by encoding RNA silencing suppressors (RSS), they employ a variety of mechanisms to suppress RNA silencing pathways. The mechanism is well-studied in viruses infecting *C. parasitica.* Mycovirus encoded RSS p29 is structurally and functionally similar to HC-Pro suppressing the hairpin RNA-mediated gene silencing in *C. parasitica.* Similarly, in *Rosellinia necatrix* Mycoreovirus-3 encodes s10, and interacts with dicing of dsRNA thereby suppressing RNAi mechanism.

Effect of mycovirus on fungal host

The majority of mycoviruses don't show symptoms on their host. They induce beneficial effects like hyper-virulence, which is the opposite of hypo-virulence. It causes increased growth rate, sporulation and virulence of its host. This has been observed in *Alternaria alternata* infected by AaCV-1 induced enhanced pathogenicity and *Phytophthora infestans* infected by PiRV-2 exhibited enhanced ecological fitness and virulence. Another beneficial effect is Killer-phenomenon associated with certain dsRNA, *Totivirus* species in yeasts such as *Saccharomyces*, *Hanseniaspora* and *Zygosaccharomyces* and also in *Ustilago maydis*. The killer strains excrete a proteinaceous toxin, to which they are immune but which is lethal to non-killer cells. The killer trait will help beneficial yeasts by eliminating undesirable competitors.

Mycoviruses also induce harmful effects like Hypo-virulence where, virus infected fungal strains exhibit reduced growth, sporulation and virulence. It has greater importance in fungal disease management. During hypo-virulence mechanism re-programming of host-metabolism, signalling pathways, virulence factors and gene silencing occurs, where the viruses target various virulence factors of the fungus and alters their expression. The best example is the hypovirulence caused by mycovirus, SsHADV-1 on plant pathogenic fungi, *Sclerotinia sclerotiorum.*

Engineering of mycoviruses or their fungal host to enhance their potentiality as a bio-control agent

The potentiality of mycoviruses can be enhanced by the nuclear integration of a full-length cDNA copy of mycovirus. It provides a strategy to enhance the horizontal and vertical transmission of viruses. This results in the transmission of the virus to ascospores, which in turn allows horizontal transmission through sexual crosses regardless of vegetative incompatibility. The 'SUPER DONAR' approach also can be used where, *C. parasitica* strains are able to transfer CHV-1 to isolates from a variety of vegetative compatible groups (VCG). The super donors are produced by disrupting vic loci, which is known to restrict mycovirus transmission, they are expected to enhance the effective delivery of hypoviruses in *C. parasitica* (Zhang and Nuss 2016).

Further, chimeric mycoviruses with enhanced properties can be used as a bio-control tool. CHV1-EP13 showed that there is a possibility to fine-tune the effect on the host through molecular manipulation of mycoviruses, uncoupling hypo-virulence from alteration in growth, as less impact on growth is indeed advantageous to the dispersion of mycoviruses. But the key problem here is to achieve a proper balance between factors that makes mycoviruses successful in preventing diseases and contribute to their ecological fitness allowing them to be prevalent in the pathogen population. Genetic manipulation of mycoviruses is also one of the approaches, but it is difficult and requires a huge understanding of complex mycovirus molecular biology and needs extensive field trials. In all these ways, the potentiality of mycoviruses can be enhanced to use as a successful biological control tool.

Applications

Mycoviruses have wider applications in agricultural and other medical sciences to control many fungal diseases of plants and humans. It has both protective and therapeutic action against inducing hypo-virulence.

Gene vector:

As many mycoviruses have minimal effects on their host fungus, a complementary approach to biological control might be the use of gene vectors. ssRNA virus belongs to the family *Flexiviridae*, BVX, BCVF and ssDRV are prime components of this approach. PVX has been successfully used as a vector for the expression of genes from a range of different sources in plants.

Plant vaccine:

Here the plant vaccination strategy may enhance the plant resistance to a broad spectrum of pathogens and promote plant growth as endophytes, PGPR and arbuscular mycorrhizal fungi (Dutta and podile 2010). Hypervirulence-inducing mycoviruses can be used as an efficacy enhancer of some of the fungal bio-control agents. Mycoviruses have huge scope in forest and horticultural crops, where repetitive chemical management is difficult and non-economical. As mycoviruses can transmit independently, persist in nature exerting continuous control over fungi (but vegetative incompatibility is a limiting factor in some of the cases).

Killer phenomenon:

The phenomenon is associated with certain dsRNA *Totivirus* species in yeasts such as *Saccharomyces*, *Hanseniaspora* and *Zygosaccharomyces* and also in *Ustilago maydis*. The killer strains excrete a proteinaceous toxin to which they are immune but which is lethal to non-killer cells.

Challenges

The use of mycoviruses is associated with certain challenges like, vegetative incompatibility, co-infection and efficacy.

Vegetative incompatibility:

There is a lack of independent transmission within pathogenic population due to vegetative incompatibility, which limits transmission and effect of mycovirus infection.

Co-infection of mycoviruses:

It is common in natural conditions, where mixed infection of mycoviruses has an antagonistic effect on themselves. Hence, they reduce the efficiency, stability and transmission of mycoviruses.

Effectiveness:

Efficacy is questionable in some cases along with the stability of mycovirus (in extracellular life) under influence of the environment.

CONCLUSION

Since mycoviruses are infecting many fungal species, there is a scope to exploit them to a greater extent. Prerequisite qualities of inducing hypo-virulence and effective transmission are needed in order to be used in the management of fungal diseases. Information regarding the role of *vic* genes in restricting horizontal transmission, and the characterization of mycovirus that lower vegetative incompatibility barriers, can be exploited to improve the ability of biocontrol viruses. RNA-Seq will give a comprehensive transcriptome analysis of *fungus* when infected with different mycoviruses that can be used to determine transcriptional regulation in fungus by mycovirus. Mycoviruses with the ability to manage a wide range of diseases are already in existence, and strategies to increase their properties by manipulating virus–fungus interactions are being developed. Field experiments and research on the effectiveness of different phytopathogens are required.

REFERENCES

Bruggeman, J., Debets, A. J., & Hoekstra, R. F. (2004). Selection arena in *Aspergillus nidulans*. *Fungal Genetics and Biology*, *41*(2), 181-188.

Dutta, S., & Podile, A. R. (2010). Plant growth promoting rhizobacteria (PGPR): the bugs to debug the root zone. *Critical reviews in microbiology*, *36*(3), 232-244.

Ghabrial, S. A., Castón, J. R., Jiang, D., Nibert, M. L., & Suzuki, N. (2015). 50-plus years of fungal viruses. *Virology*, *479*, 356-368.

Glass, N. L., & Dementhon, K. (2006). Non-self-recognition and programmed cell death in filamentous fungi. *Current opinion in microbiology*, *9*(6), 553-558.

Jiang, D., Fu, Y., Guoqing, L., & Ghabrial, S. A. (2013). Viruses of the plant pathogenic fungus *Sclerotinia sclerotiorum*. *Advances in virus research*, *86*, 215-248.

Li, P., Wang, S., Zhang, L., Qiu, D., Zhou, X., & Guo, L. (2020). A tripartite ssDNA mycovirus from a plant pathogenic fungus is infectious as cloned DNA and purified virions. *Science advances*, *6*(14).

Yu, X., Li, B., Fu, Y., Jiang, D., Ghabrial, S. A., Li, G., ... & Yi, X. (2010). A geminivirus-related DNA mycovirus that confers hypovirulence to a plant pathogenic fungus. *Proceedings of the National Academy of Sciences*, *107*(18), 8387-8392.

Zhang, D. X., & Nuss, D. L. (2016). Engineering super mycovirus donor strains of chestnut blight fungus by systematic disruption of multilocus *vic* genes. *Proceedings of the national academy of sciences*, *113*(8), 2062-2067.