

Biocontrol of root diseases of fruit trees with fungal viruses

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Helicobasidium mompa Tanaka and *Resellinia necatrix* Prillieux cause violet root rot and white root rot of various crops, respectively. Intensive cultural practices, such as the use of dwarf stock, glasshouse cultivation, etc., predispose plants to the diseases. The diseases can be controlled only by biennial drench of 50100L of chemicals for each tree. Biocontrol with soil microorganisms proved ineffective under field conditions. Long-term control may be hampered by the perennial growth of hosts and by the difficulty in the establishment of antagonists in soil. Crop rotation or soil amendment is not applicable, either. Fungal viruses with dsRNA genome (Buck 1986) are promising against root diseases of fruit trees since they exist within the cytoplasm of fungal hyphae and need no effort to help them persist in the field. The viruses are considered to spread through the network of fungal mycelia in the soil once they enter the fungal cytoplasm. Here, we present preliminary results from a project to control the root diseases of fruit trees with dsRNA (Matsumoto 1998).

Six-11 isolates for each diseased trees were first obtained from underground parts. Pairing in all possible combinations within each tree revealed that, with an exception of a tree with white root rot located on the junction of disease patches, isolates from the same trees belonged to the same mycelial compatibility groups (MCGs) in both *H. mompa* and *R. necatrix*. Subsequent analysis on population structure indicated that each orchard had a few disease patches which were occupied by single MCGs. Large patches occupied by prevalent MCGs of *H. mompa* spread more extensively than those with less prevalent MCGs in 4 years. These observations indicate that, on starting biocontrol, one should determine which patch to control since inoculum containing dsRNA is custom-made to apply to specific MCGs.

A variety of dsRNA elements differing in number and size was detected from 464 out of 608 MCGs (76.3%) for *H. mompa* and 82 out of 413 MCGs (19.9%) for *R. necatrix*. Isolates of the same MCGs from single patches often had different dsRNAs. These findings may not preclude the possibility that indigenous dsRNAs interfere with novel dsRNAs introduced as biocontrol agents and reduce their effect. Fitness of *R. necatrix* MCGs was compared in terms of virulence, competitive saprophytic ability, and the presence or absence of dsRNA. MCGs that were found both from roots and soil were generally more virulent than satellite MCGs found exclusively from soil. Satellite MCGs tended to be less competitive for colonized substrate and to have dsRNA.

Hyphal tip isolation was made from a weakly virulent strain of *R. necatrix* to remove dsRNA. Whereas, less than 20% of plants were killed 14days after inoculation with a parental strain, its hyphal tip isolates without dsRNA recovered virulence, killing almost 100% of plants. Hyphal tip

isolation to recover virulence was successful in some other strains as well as in *H. mompa*. Other methods to introduce dsRNA will be presented.

dsRNA is transmitted through hyphal anastomosis between somatically compatible strains, and simple population structure of pathogens is prone to the dissemination of dsRNA. Surveys on population structure of the two soilborne pathogens of fruit trees demonstrated the feasibility of biocontrol with dsRNA along with the economic value of individual trees. Methods to transfect dsRNA to different MCGs should be developed so that any MCGs may be infected. Our ultimate goal is to breed strains that are compatible with any MCG and to formulize them for field application.