Resistance to Rice Yellow Mottle Virus Disease in African Rice.

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RYMV-infected rice in West Africa

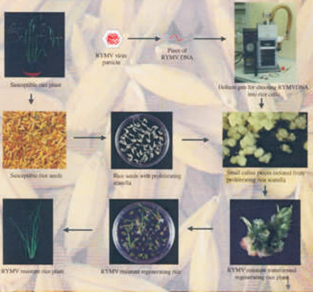
Introduction

Rice yellow mottle virus (RYMV) is a devastating viral disease of rice, endemic in East and West Africa and affecting mainly lowland rice ecosystems. This serious, highly infectious disease is spreading in those areas of Africa targeted for future increases in rice production. Some success has been achieved in the production of resistant interspecific varieties, and at the same time a pathogen-derived resistance mechanism, transgenically expressed in the host has been shown to be effective in halting the infection process (Pinto *et.al.* 1999). This approach was used here but using positive selection with mannose rather than antibiotic selection

Distribution of RYMV in Africa

The Transformation Process

Previous research has shown that inserting a small piece of viral RYMV DNA into the rice DNA can provide excellent protection against the virus (Pinto *et al.* 1999). Rice recognises and destroys completely all virus matching the piece that it contains. This protection is 100 % throughout the life cycle of rice and the inserted viral DNA accounts for only 0,004 % of the total rice DNA. This pathogen-derived resistance mechanism relies on a high level of homology between the expressed transgene and the incoming virus (Samford & Johnson,1985).

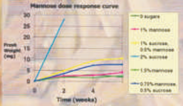


Constructs for Bombardment

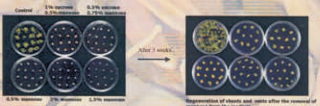
New constructs have been produced to replace the antibiotic hygromycin selection gene with a phosphomannose isomerase (PMI) gene for positive selection. A highly conserved region of the RYMV genome is used to maximize resistance to different strains of RYMV.

Mannose selection rather than antibiotic selection

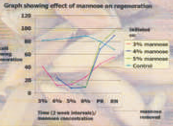
Most plants, except legumes are unable to use mannose-6-phosphate as a carbon source. Introducing the phosphomannose isomerase (PMI) gene during transformation makes this an ideal system for positive selection, whereby plants containing this gene are able to survive on a medium containing mannose. Mannose selection provides a metabolic advantage to transformed cells while the untransformed cells are starved and lose their viability (Joersbo *et.al.* 1998). This marker has been found to be effective in the selection of wheat and maize transgenics (Wright *et.al.* 2001), but there is limited information on selection in rice.



- Mannose at 10 or 15 g/l was lethal to the tissue within the first two weeks.
- The inclusion of even low quantities of mannose resulted in a significant decrease in callus weight over time.
- The inclusion of sucrose at 5 or 10 g/l resulted in some callus growth, although mannose present was still inhibitory



Adding mannose at the regeneration stage resulted in complete, but reversible, inhibition of regeneration and shoot formation (as seen above). Three weeks after removal of mannose from the medium, plant regeneration resumed.



It was necessary to remove mannose at the regeneration or germination phase to allow plantlet formation. It has been suggested that even transformed plants may be inhibited by mannose at this stage (Lucca *et.al.* 1999).

Future prospects

By combining multigenic partial resistance from conventional breeding, and the single gene total resistance from the transformed rice, we can provide strong, durable protection against RYMV for African rice farmers, to provide a rice yield free from the threat of RYMV.

References

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Pinto Y, Kok R & Baulcombe D (1999) Nature Biotechnology 17 (7): 702-707
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