Strategies for field collection of recalcitrant seeds and zygotic embryonic axes of the tropical tree, *Trichilia dregeana* Sond.

P. BERJAK, J.I. KIOKO, A. MAKHATHINI AND M.P. WATT

School of Life and Environmental Sciences, University of Natal, Durban, 4041 South Africa (E-mail: berjak@biology.und.ac.za)

(Accepted October 2003)

Summary

After aril and testa removal, high-quality *Trichilia dregeana* seeds dusted with a benzimadole fungicide were successfully stored for 91 d (13 weeks) in the hydrated condition at 16°C and when set to germinate, showed no associated fungal proliferation. In contrast in a poor harvest year, seeds with which fungal association was ubiquitous, had all lost viability and the associated fungi proliferated within 28 d. Use of systemic fungicides with or without other measures to curtail the mycoflora, was of only limited benefit; storage at 6°C was lethal and where fungal activity was curtailed, bacterial degradation was rife. As an alternative to seed storage, the investigation then focused on axis collection for cryopreservation. Two approaches were tested. In the first, three treatments were used involving 72 h storage of seeds in 1.6% solutions of potassium chloride, potassium sorbate or sodium benzoate, both with and without previous surface sterilization. The most successful treatment, potassium chloride storage following surface sterilisation, eliminated the fungus from 80% of the axes, but bacterial contamination was evident. The other involved an *in vitro* field collection of excised axes, followed by culture on medium with systemic fungicides and an antibiotic and it resulted in 85% germinated axes that were entirely free of both fungal and bacterial contaminants.

Introduction

Seed storage at low relative humidity (RH) and near- or sub-zero temperatures is the most convenient and widely-used practice for plant genetic resources conservation and for the maintenance of planting stocks. However, this practice is possible only for seeds that show orthodox post-harvest behaviour (Roberts, 1973), which is determined by the acquisition of desiccation-tolerance during development and the consequent ability of the seeds to survive in the dehydrated state (reviewed by Pammenter and Berjak, 1999).

However, seeds of many species are not able to withstand the degree of dehydration tolerated by orthodox types (Roberts, 1973). Such recalcitrant seeds are shed at elevated water contents (generally in the range 0.3 - >4.0 g g⁻¹), with the proportion of water loss tolerated being a species-specific characteristic, but differing markedly for individual species in relation to the drying rate and suspectedly, also the temperature during the dehydration period (Ntuli *et al.*, 1997; Pammenter and Berjak, 1999). While most of the species producing recalcitrant seeds are of tropical/sub-tropical provenance, this property characterises seeds of several temperate species also, e.g. *Quercus* spp. (Pritchard, 1991; Finch-Savage and Blake, 1994; Connor and Bonner, 1996).