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Desiccation Sensitivity of *Trichilia dregeana* Axes and Antioxidant Role of Ascorbic Acid

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Abstract: Recalcitrant seeds of *Trichilia dregeana* Sond. were used as experimental material, and desiccation sensitivity of *T. dregeana* axes and antioxidant role of ascorbic acid were studied. Desiccation tolerance of *T. dregeana* axes progressively declined with dehydration, water content at which 50% of axes has been killed by dehydration (W_{50}) was about 0.16 g H_2O/g DW. During dehydration, electrolyte leakage rate of axes gradually increased, the activities of superoxide dismutase (SOD), ascorbate peroxidase (APX), catalase (CAT), glutathione reductase (GR) and dehydroascorbate reductase (DHAR) declined, and content of thiobarbituric acid (TBA)-reactive products increased. Two point five-10 mmol/L ascorbic acid (AsA) treatment could significantly increase desiccation tolerance and activities of SOD, APX, CAT and GR in axes, and decrease electrolyte leakage rate and content of TBA-reactive products in axes. The results showed that desiccation tolerance of *T. dregeana* axes was strongly correlated with the increase in the activities of antioxidant enzymes and the decrease in lipid peroxidation.

Key words: ascorbic acid; ascorbate peroxidase; catalase; dehydroascorbate reductase; glutathione reductase; lipid peroxidation; recalcitrant seed; superoxide dismutase; *Trichilia dregeana*

Recalcitrant seeds are shed at high water contents and are intolerant of dehydration and often also of chilling. A number of processes or mechanisms have been suggested to confer, or contribute to desiccation tolerance. Different processes may confer protection against the consequences of loss of water at different hydration levels, and the absence or ineffective expression of one or more of these could determine the relative degree of desiccation sensitivity of seeds of individual species (Pammenter and Berjak, 1999; Song *et al.*, 2003a; 2003b). Free-radical generation as a consequence of uncoordinated metabolism may well be a major injurious factor during relatively slow dehydration of recalcitrant seeds, underlying a spectrum of lethal lesions (Smith and Berjak, 1995; Côme and Corbineau, 1996). Recalcitrant seeds (or their embryos) do appear to possess antioxidant mechanisms (Hendry *et al.*, 1992; Finch-Savage *et al.*, 1994). However, these protective mechanisms may become impaired under conditions of water stress (Smith and Berjak, 1995; Walters *et al.*, 2002); certainly, they are ineffectual in terms of protecting against desiccation damage.

A peculiar difference between orthodox and recalcitrant seeds concerns the ascorbate recycling enzymes, AFR redutase and DHA reductase. The DHA reduction capability is low in recalcitrant seeds, but is high in the orthodox ones. In contrast, AFR reductase activity is high in recalcitrant seeds and low in the orthodox ones (Tommasi *et al.*, 1999).

AsA is a major primary antioxidant, reacting directly with hydroxyl radicals, superoxide and singlet oxygen (Foyer, 1993; Buettner and Jurkiewicz, 1996). In addition to its importance in photoprotection and the regulation of photosynthesis (Forti and Elli, 1995), AsA plays an important role in preserving the activities of enzymes that contain prosthetic transition metal ions; and is also a powerful secondary antioxidant, reducing the oxidized form of α-tocopherol, an important antioxidant in nonaqueous phases (reviewed by Noctor and Foyer, 1998). *Trichilia dregeana* seed is a recalcitrant seed, and is highly desiccation sensitive (Han

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Abbreviations: AFR, ascorbate free radical; APX, ascorbate peroxidase; AsA, ascorbic acid; BSA, bovine serum albumin; CAT, catalase; DHA, dehydroascorbate; DHAR, dehydroascorbate reductase; EDTA, ethylenediaminetetra-acetic acid; GR, glutathione reductase; GSH, reduced glutathione; GSSG, oxidized glutathione; MDA, malonylaldehyde; NADPH, reduced nicotinamide adenine dinucleotide phosphate; NBT, nitroblue tetrozulium; PVPP, polyvinylpolypyrrolidone; SOD, superoxide dismutase; TBA, thiobarbituric acid.