the X. scabrida leaves was investigated with (+ petiole) or without (− petiole) petiolar involvement. This paper reports the reconstitution of the photosynthetic system in X. scabrida on rehydration with the exclusion of the rehydration through the petioles. Leaves in the + petiole treatment reached maximum water content in 8 h. The time course of rehydration in the − petiole leaves was identical to that in the + petiole ones. The reconstitution of the photosynthetic apparatus in the previously desiccated achlorophyllous X. scabrida leaves was complete in 72 h which was indicated by the values of photochemical activity, CO₂ assimilation, and stomatal conductance. There was no difference in the revival and the reconstitution of photosynthesis between the two treatments. That is the ‘petioles’ did not have a role in water uptake which occurred entirely through the leaf surface. In intact plants root and xylem transport are a prerequisite for petiolar uptake. During desiccation, the adventitious roots of X. scabrida die back and the new ones only develop after the completion of the revival of the leaves. In X. scabrida the fully functional revived leaves are a prerequisite for the development of new roots. There is no xylem function in the rehydrating X. scabrida and therefore petiolar water uptake can be of no importance for its revival. If petiolar uptake was of importance then the water relations and physiological revival could not be completed in the experimental leaves with their petioles prevented from taking up water.

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A gene of unknown function, specifically found in seeds, is upregulated during desiccation in vegetative tissues of Xerophyta humilis

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Xerophyta humilis is a desiccation tolerant angiosperm (resurrection plant) able to survive a loss of 95% of its relative water content and resume biological function upon rehydration. A small scale microarray analysis, using 424 annotated cDNAs from a X. humilis cDNA library, identified a novel gene (HC205) that is upregulated during desiccation. In this study we aim to characterise the gene and to determine its role in desiccation tolerance. The Arabidopsis thaliana ortholog of HC205 identified in a BLAST analysis was annotated as a glyoxalase I-like family protein. A βαβββ fold was identified in the HC205 protein sequence. This is a conserved structural feature of members of the vicinal oxygen chelae (VOC) super family, which includes the enzyme glyoxalase I. Although HC205 has low sequence similarity to known glyoxalase I genes and lacks the regions important for glyoxalase I activity, we tested its ability to detoxify methylglyoxal. Over-expression of HC205 conferred tolerance to E. coli only at low concentration levels of methylglyoxal. In addition, studies aimed to determine whether HC205 could complement a yeast glyoxalase I mutant showed that ectopic expression of HC205 is lethal in yeast. A BLASTn search against the TIGR gene indices database identified six plant HC205 orthologs in desiccation-sensitive plants. An analysis of representation of these orthologues in the TIGR EST libraries indicated that these orthologues were absent from EST libraries derived from vegetative tissue during abiotic stress but were present in seed EST libraries of desiccation-sensitive plants. These findings were confirmed by RT-PCR on desiccation-stressed vegetative tissues and seed of A. thaliana. In contrast, HC205 mRNA transcripts in X. humilis were shown to increase significantly in vegetative tissue during desiccation, in addition to being expressed at high levels in seeds. Western blot analysis and immunocytochemistry was used to characterise the temporal pattern and localization of HC205 protein during a time course of desiccation and rehydration in leaves and roots of X. humilis.

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Intact plasma membranes in dried cells

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Desiccation tolerance of a dried organism can be inferred only from tests after rehydration. It is virtually impossible to test viability in the dried state because dried cells lack metabolic activity. Under these conditions, the quintessence of life is restricted to structural organization. Freeze fracturing is the only available method to study intactness of membranes in dried cells at high resolution. So far, only desiccation-tolerant cells have been tested for membrane structural intactness in the dry state. With the use of high-resolution freeze-fracture scanning electron microscopy we now show that membrane structural intactness is maintained in dried human erythrocytes and other desiccation-sensitive cells. Our present observations using this technique indicate that, even in desiccation-sensitive cells, the native membrane structure and random distribution of IMPs can be maintained with drying without protective substances. The random IMP distribution indicates that drying did not cause large scale phase separations. The question remains whether such dried systems that are apparently intact on the nanometer scale and therefore ‘structurally organized’ can be considered as still viable. The massive leakage of solutes from desiccation-sensitive cells with rehydration would indicate that there is insufficient protection on a molecular scale to allow cells to become rehydrated without loss of life. A practical implication of this work might be that SEM
Evolutionary linkage between water-deficit responses and desiccation tolerance

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Drought tolerance and desiccation tolerance have often been cited as manifestations of the same mechanism; desiccation tolerance being the extreme form of drought tolerance. However, there is a fundamental difference between drought and desiccation tolerance: drought tolerance mechanisms include ways of maintaining cell water content, such as osmotic regulation and stomatal closure, whereas desiccation tolerance consists of ways to survive the almost complete loss of water. It is clear that an evolutionary understanding of the relationship between drought and desiccation tolerance is necessary to determine which genes are adaptive in nature and which simply respond to secondary events such as cell injury. Our approach is to compare the expression profiles for genes in response to water deficits in drought sensitive species with their orthologues in desiccation-tolerant species during desiccation and within a phylogenetic framework. Our comparisons encompass a dicot to dicot pairing, a monocot to monocot pairing, and the comparison of both to the most primitive form of vegetative desiccation tolerance as manifested in the desiccation tolerant bryophyte Tortula ruralis. Initial comparisons between the water stress response of Arabidopsis and the desiccation response of Tortula, have generated a solid baseline of similarities and differences that have generated the necessary hypotheses for our pair-wise comparisons. Our comparisons between Arabidopsis and Craterostigma and Sporobolus stapfianus and S. pyrimidalis (aligned with maize) have given us an insight into the evolution of the response to dehydration in vegetative tissues. These data will allow us to focus attention on genes and gene networks that are truly central to cellular dehydration tolerance and may enable a more rational approach for the improvement of drought tolerance in crop species.

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A comparison of the responses of Eucalyptus grandis and E. grandis × camaldulensis to ABA pretreatment and desiccation in preparation for cryostorage

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Cryopreservation offers an attractive option for the maintenance of potentially useful, clonally propagated genotypes of Eucalyptus while field testing is conducted. The most suitable explants for this purpose are in vitro axillary buds. However, a major limitation of axillary buds is their susceptibility to freezing injury, as a consequence of being highly hydrated. Reduction of the water content to levels low enough to minimize ice crystal formation may nevertheless, overcome this limitation. The extent to which Eucalyptus buds could be dried however, was determined to be a factor of the natural tolerance of the species to water loss. Eucalyptus grandis, a drought-sensitive, sub-tropical eucalypt, was found to be less tolerant of water loss when dried over silica gel for varying periods (20, 40 and 60 min), while a more drought-tolerant hybrid, E. grandis × camaldulensis, was more resistant to water loss, maintaining a significantly higher water content than E. grandis at all the drying times tested. This was also evident at the ultrastructural level, where E. grandis buds displayed a greater degree of subcellular damage than those of E. grandis × camaldulensis. The difference between the E. grandis and the hybrid was further emphasized by the responses of the buds to exogenously applied abscisic acid (ABA). While ABA-pretreated E. grandis buds maintained a significantly higher water content and viability at each drying time tested, and displayed a marked improvement in the maintenance of cellular integrity after 20 min drying, E. grandis × camaldulensis did not respond to exogenous ABA. The beneficial effects of ABA on E. grandis was also demonstrated by the comparison of the responses of encapsulated buds precultured on progressively increasing concentrations of sucrose and glycerol (0.4 M, 0.7 M and 1.0 M) and encapsulated buds precultured on similar media but supplemented with 5 mg l⁻¹ ABA. Buds precultured on media containing sucrose, glycerol and ABA were able to resist water loss and maintain viability for a significantly longer period than those precultured on media without ABA. The results therefore demonstrated 1) the impact of genotype on the responses of Eucalyptus in vitro buds to desiccation and 2) the significant effect of ABA on the desiccation tolerance of E. grandis buds.

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The maturation of recalcitrant seed biology

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