The storage of *Griselinia littoralis* seeds: a study of physiological deterioration in a recalcitrant seed

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Introduction

Recalcitrant seeds (desiccation-sensitive) cannot tolerate storage for extended periods. It is thought that such seeds do not undergo drying during maturation (Berjak *et al.*, 1990) and the continuation of active metabolism results in germination soon after release from the parent plant. Even though these seeds are fully hydrated, ageing events during storage result in inevitable damage.

Seed ageing can result in irreversible cellular deterioration that reduces viability and ultimately leads to an inability to germinate. Temperature, relative humidity and the gaseous environment all affect seed ageing during storage (Likhatchev *et al.*, 1984) and in combination with seed moisture content, influence the rate of biochemical processes associated with seed damage. Previous research in this area has largely been restricted to orthodox seeds (e.g., Madhava Rao and Kalpana, 1994).

The aim of this study was to determine whether the storage of native recalcitrant *Griselinia littoralis* seeds results in deterioration processes similar to those of aged orthodox seeds. Decline in seed germination after storage may be the result of plasma membrane damage, decreasing carbohydrate levels, or a combination of the two.

Materials and Methods

G. littoralis seeds were artificially aged by exposure to high temperatures and a high relative humidity (Delouch and Baskin, 1973). This method accelerates ageing, shortening storage time, and allows determination of any associated physiological changes. In this study, *G. littoralis* seeds were incubated at 44°C at relative humidities of either 100 % (water) or 5 % (silica gel) as a comparative desiccated condition. Specimens were collected for analysis at 0, 3, 5, 7 and 10 d after treatment and tested for germination, water content, carbohydrate status and electrolyte leakage (plasma membrane damage).

Results and Discussion

Germination of all treated seeds (24°C, 24 h light, 50-60 % RH) was less than 10 % in contrast to 70 % germination in untreated seeds. Seeds at low humidity lost a significant amount of water prior to day 3 compared to 100 % humidity where a water content similar to day 0 was maintained. The latter result indicates that deterioration in treated *G.littoralis* seeds was a result of a temperature effect rather than desiccation. Cellular reserves of sugar were significantly reduced under high humidity while mobilization of starch occurred at a greater rate during desiccating conditions (Figs. 1 and 2). The decline of carbohydrate reserves suggests that a relatively high rate of respiration was maintained and that the seed continued to mobilise sugars that protect membranes from damage or retain cellular water. Sugars have a strong interaction with water and bind to phospholipid heads which is thought to maintain head group spacing and preserve fluidity and stability of membranes (Berjak *et al.*, 1990). During desiccation, *Griselinia* seeds appeared to induce the mobilization of starch, and possible conversion to sugars may have aided in the preservation of cellular membranes. These observed processes of carbohydrate mobilisation did not however maintain seed viability.

G. littoralis seeds treated at 100 % humidity showed some indication of plasma membrane damage with a gradual increase in electrolyte leakage over time (Fig. 3). In desiccated seeds however, conductivity reached a high value by day 3 but by day 5 declined to a level similar to the day 0 value. This indicated a problem with the conductivity method where ions may have leaked out onto filter paper by day 3 and not into the solute for measurement.

In a later experiment, *G. littoralis* seeds stored at room temperature for 14 days resulted in 95 % germination at high humidity and 2 % in desiccation conditions. In future work, recalcitrant seeds treated at room temperature instead of 44°C may elucidate clearer physiological differences between the two storage conditions.

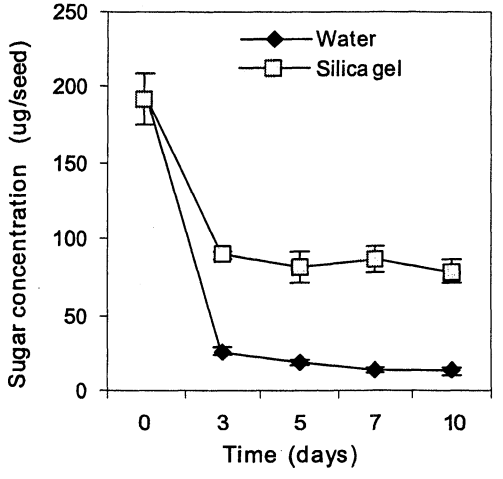

Figure 1. Time course analysis of sugar degradation of *G. littoralis* seeds over an ageing period of 10 days. Each data point represents the average of three replicates.

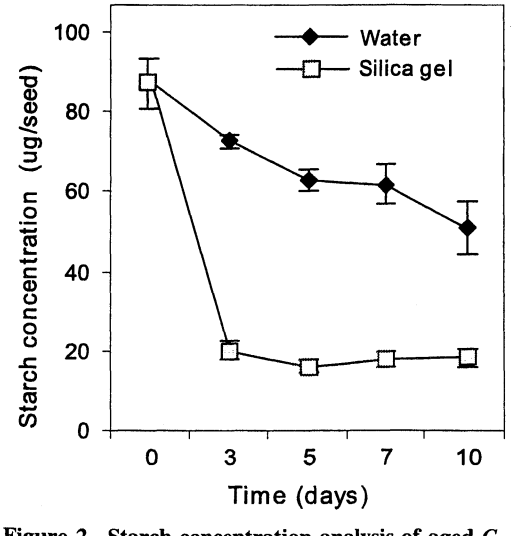

Figure 2. Starch concentration analysis of aged *G. Littoralis* seeds over a 10 d time course. Each data point represents three replicates.

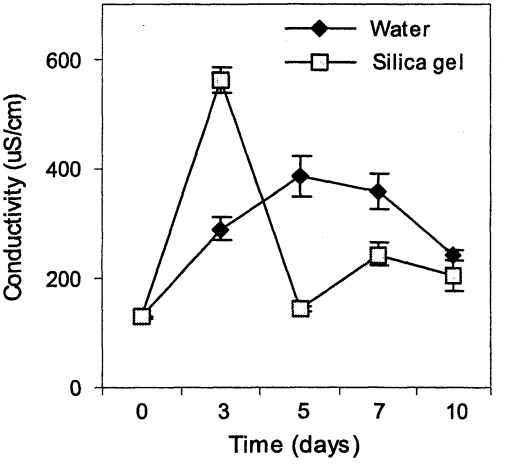

Figure 3. Analysis of electrolyte leakage from cellular plasma membranes of aged *G. littoralis* seeds. Each data point of conductivity represents three replicates.

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