Pre-Treatment of Bulk Samples of Certain *Eucalyptus* Species to Enhance Germination

David O. Cliffe
Narromine Transplants, PO Box 123, NARROMINE NSW 2821

Increases in germination percentages were achieved when seeds of several *Eucalyptus* species were subjected to a pregermination treatment and then separated in sugar solutions. In one species, *E. pilularis* Smith, an increase in germination percentage of 29% was achieved.

INTRODUCTION

With an upsurge in demand for seedlings of *Eucalyptus* for the establishment of forestry plantations, there has been a need to develop techniques to increase seed viability. A number of species, which have in the past been propagated via broadcast and pricking in techniques, do not lend themselves to the single seeding practices needed for the economic production of cell-raised seedlings. The more commonly propagated species, such as *E. globulus* Labill. ssp. *globulus* and *E. nitens* (Deane & Maiden) Maiden, belong to the subgenera *Symphyomyrtus*. This group has seed that is usually of high viability, 780 and 2710 germinants per 10 g, respectively (Langkamp, 1987). Two species that are currently in demand for planting along the New South Wales north coast are *E. pilularis* Smith and *E. cloeziana* F. Muell., both of which belong to the subgenera *Monocalyptus*. These species have been tested at 550 and 1210 germinants per 10 g, respectively (Langkamp, 1987).

Seed of *Eucalyptus* is collected by harvesting mature capsules from the tree. Once the capsule has opened and its contents expelled, the spent capsules and opercula are sieved from the bulk and the remaining sample is used for sowing. This sample contains not only seed but large amounts of chaff comprising aborted seed and other small parts of the inner capsule. In many species, in particular those associated with the subgenera *Symphomyrtus*, it is possible to separate the chaff from the seed by mechanical means. Fractionating aspirators, winnowers, gravity tables, and sieves will give a clean sample that can be single seeded using needle type vacuum seeding equipment.

With the subgenera *Monocalyptus*, there are often large amounts of aborted seed present in the sample, even after a lengthy cleaning process. Microscopic examination reveals that the differential of mass, shape, and size between the viable and nonviable portions are so small as to be almost unmeasurable. Part of the technique of fluid drilling (Currah et al., 1974) employs the process of imbibition and germination of seed to a point where the radical just emerges. The differences in weight between those seeds that have imbibed and those that have not, can be exploited by introducing the seedlots into a solution of approximately the same relative density (Darby and Salter, 1976). Those seeds that have imbibed and have a changed specific gravity will float, those that have not, because they lack endosperm and therefore the ability to absorb moisture, will sink (Salter, 1978). This process can be utilised to separate imbibed seedlots of *E. pilularis* and *E. cloeziana*

but before radical emergence. The heavier portion can then be surface dried and sown immediately using vacuum type sowers.

MATERIALS AND METHODS

Seed capsules of *E. pilularis* were collected from mature trees and the seed extracted from the capsules after drying in direct sunlight. The bulk sample was sieved to eliminate expelled capsules, opercula, and other large material. Gravity table and fractionating aspiration separation was then employed to eliminate the lighter fraction of the seed sample, containing chaff, dust, and small aborted seed. Immediately before the start of the imbibition process the seed was immersed in a solution of 10 ml of 12% commercial bleach, one litre of tap water and 1 ml of detergent, for 3 min. The sample was then rinsed with tap water to remove the bleach and detergent.

Approximately 30 g of the sample was then placed into stainless steel baskets each 230 mm long, 80 mm wide, and 15 mm deep with a fine-mesh bottom small enough to retain the seed and let excess water pass through. Eighteen of these baskets were then placed on a frame inside a germinator constructed from a domestic chest freezer. The freezer was connected to the water supply and filled to a depth of 300 mm. An overflow at the 300 mm level allowed water to flow through the germinator. A heating element and thermostat regulated water temperature and the water reservoir was circulated by a small pump to ensure adequate aeration. A misting system using a timer and commercially available propagating mist nozzles was placed over the top of the baskets. The freezer itself was not operational for this experiment. Water flowed through the germinator at 500 ml min^{-1} and the reservoir for the treatment of *E. pilularis* was heated to 25C. Misting frequency was 10 sec every 20 min from two nozzles.

Treatment time for *E. pilularis* is usually 40 h. The testa of the seed starts to become translucent at this stage indicating that it is imbibing and that radical emergence is imminent. Germination must not proceed beyond this point or desiccation will occur during the surface drying process if the radical is allowed to emerge.

A separating solution was made by dissolving 1 kg of sugar in 1 litre of water. Five litres of this solution was then placed in a bell jar fitted with a large tap at its base. The imbibed seed was then emptied into the bell jar and lightly agitated. After a period of about 1 min, seed starts to separate into two fractions. There should be a clear delineation between the denser fraction on the surface and the lighter fraction which falls to the bottom. The solution was then decanted through a sieve taking first the bottom fraction, either discarding it or placing it back in the germinator for a further period, and then the top fraction.

The seed from the top fraction was then washed thoroughly with fresh water prior to storage or surface drying. Separated seed from the top fraction can be stored for 4 to 5 days in containers with fresh water which must be sealed and kept in a refrigerator at a temperature of 3 to 4C. It is then placed into small net bags containing 100 g of seed each, placed in an air stream at ambient temperature for approximately 15 min and then sown.

RESULTS

Germination results for seed of *E. pilularis* treated by this method shows a significant improvement (McLeod pers. comm.).

Table 1. Comparison of germination tests for seed of *Eucalyptus pilularis* Smith before and after treatment (3 replicates).

Treatment	Germination (%) (after 14 days)
Gravity separated and aspirated	22
Manually separated (pure seed)	69
Aspirated, bleached and washed	29
Imbibed and separated in sugar solution	51
Residual seed left after separation	0
Squash test after separation	52

DISCUSSION

A significant increase in germination percentage has been obtained as a result of the imbibition and sugar separation treatment. However, it has been observed that there is a secondary phase of germination taking place approximately 30 days after sowing bringing the percentage of germinants into line with the results shown in Table 1 for manually separated, i.e. 69%. This phenomenon seems to occur with a number of the subgenera *Monocalyptus*; in particular, *E.pilularis, E. cloeziana, E. obliqua,* and *E. laevopinea* when they are subjected to this treatment. From a nurseryman's point of view, an increase in the percentage of germinants of 29% for a given sample, allows for single or double seeding. Using specially designed seeders for this purpose, there are considerable savings to be had in the cost of production. Our results to date have been encouraging although a point has been reached, where we believe it may not be possible to further increase the percentage of germinants in samples of *E. pilularis*, via this process (McLeod pers. comm.).

It is quite possible that the species we are dealing with experience inherent problems during the reproduction process, particularly at pollination. Viability may also be affected by environmental factors prevailing during that period and timing of harvest (McLeod and Schoer pers. comm.).

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