# EFFECTS OF IMBIBITION, SCARIFICATION AND MOISTURE CONTENT ON SEED GERMINATION OF BACCAUREA SAPIDA

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Received June 2000

PRASAD, P. 2004. Effects of imbibition, scarification and moisture content on seed germination of Baccaurea sapida. The germinability of freshly collected Baccaurea sapida seeds improved with scarification. The seeds exhibited hard seed coat with only 40% germination in the controls. Seeds scarified mechanically by rubbing against sand paper at the micropylar end had high germination (99%), with higher germination index and lower mean germination time compared with the control. Germinability of seeds was improved by soaking seeds in hot water and through mechanical scarification opposite the micropylar end (70 and 75% germination respectively). Seed coat did not act as a mechanical barrier to emergence of embryo but prevented water entry for initiating germination. Therefore, B. sapida seeds should be sown immediately after collection and mechanical scarification.

Key words: Mean germination time – germination index – emergence – viability – Himalaya

PRASAD, P. 2004. Kesan pemedapan, pelelasan dan kandungan lembapan terhadap percambahan biji benih Baccaurea sapida. Percambahan biji benih Baccaurea sapida yang baru dikutip diperbaik melalui pelelasan. Kulit biji keras dan biji benih kawalan cuma menunjukkan percambahan 40%. Biji benih yang dilelas dengan menggosok hujung mikropil menggunakan kertas pasir menunjukkan percambahan yang tinggi (99%). Indeks percambahannya tinggi dan masa percambahan singkat berbanding kawalan. Percambahan biji diperbaik dengan merendam biji benih di dalam air panas dan melelas biji benih pada hujung bertentangan dengan mikropil (masing-masing percambahan 70% dan 75%). Kulit biji tidak berfungsi sebagai penghalang mekanik kepada kemunculan embrio tetapi berfungsi sebagai penghalang kepada kemasukan air untuk memulakan percambahan biji benih. Justeru, biji benih B. sapida patut dicambah serta-merta selepas dikutip dan selepas dilelas.

#### Introduction

Baccaurea sapida, family Euphorbiaceae, is a medium-sized tree, which occurs wildly or is cultivated in the sub-Himalayan tracts, Sikkim, Assam, Myanmar and the Andamans. It prefers a deep moist soil and wet climate. The yellow glabose fruits, about an inch in diameter, posses a pleasant acid taste. In Thailand, the flowers and leaves are said to be eaten (Anonymous 1969). Four seeds have been recorded from each fruit. In general, the seed of B. sapida is heart-shaped, bearing parchment hairs, round at one end (posterior end) and sharply pointed at the micropylar end.

The genus, comprising about 100 species, is mostly of Indo-Himalayan origin;

some 29 species are found in India and a few extend to tropical Africa and Polynesia. The trees often produce fruits in great abundance. The pulp and the juicy aril of some are edible. The fruits are generally acidic but those from Malaysia, Sumatra and Java are sweet. In Malaysia, fermented liquor is made from the fruits of some species.

Sikkim Himalayan region stands out as one of the very few remaining areas throughout the Hindukush Himalaya where environmental degradation has yet to reach alarming dimension. The genetic diversity of both forest vegetation and agricultural crops is enormous. The state has about 600 plant species and more than 4000 species of flowering plants (Sundriyal *et al.* 1992). The availability of the wild fruit plant is rich in this tiny state of India due to its wide range of climatic conditions, from subtropical to temperate and alpine. Sundriyal and Sundriyal (1998) have reported 189 wild fruit plant species from the wild of the Sikkim Himalaya.

Germination of the seeds, as in many other species, is hindered by dormancy (Burkart 1952, Bewley & Black 1982, Popinigis 1985), which is mainly associated with water-impermeable seed coats (Lopez & Aviles 1986). Baccaurea sapida seeds exhibit maximum germination after removing the hard seed coat of freshly collected seeds but died shortly after one month of storage. This shows that either they are short-lived or recalcitrant in nature. Due to the very high consumption rate of fruits, the dispersal of the seed is important for the regeneration of such valuable tree species. In an attempt to standardise its nursery technology, seeds of this species were studied to obtain maximum germination.

## Materials and methods

Ripe fruits of *B. sapida* were collected from the natural habitat from a mature tree stand at Sikkim Himalayas. The skin of the fruit was broken to separate the seeds, which were thoroughly washed in water. Seeds were mixed and divided into four groups. The first group of seeds served as control (untreated seeds), while those in the second group were immersed in a beaker containing hot distilled water (80 °C), then left to cool to room temperature and kept in water for 24 hours. In the third group, seeds were scarified from the micropylar end, while those of the last group were scarified from the opposite end of the micropyle. Seeds were mechanically scarified by abrasion with a fine-grained sand paper to avoid injury to any internal tissue. A total of 120 seeds replicated four times (with 30 seeds each) were placed for germination in Petri dishes on top of a layer of filter paper and kept in an incubator at 25 °C. The filter papers were regularly moistened with distilled water. Daily observation for seed showing radical emergence was recorded as germinated. Percentage germination was recorded every day for 50 days until no further germination was observed.

For water imbibition, 10 seeds from each treatment were initially weighed dry, imbibed with water at room temperature for 24 hours and weighed again. For the hot water treatment, seeds soaked for 24 hours in hot water were weighed before and after soaking to determine the water uptake. The germination index (GI) and mean germination time (MGT) were calculated according to Kendrik

and Frankland (1969) as well as Ellis and Roberts (1981) respectively. Data were analysed using Duncan's test. Treated seeds stored for one month at room conditions were tested for their germination capacity. Data of germinability were also analysed using analysis of variance. The objective of this study was to investigate ways of improving the germination of *B. sapida* seeds.

## Results and discussion

The seeds of *B. sapida* had low percentage of germination in the control with only 40% germination after 25 days (Table 1). However, mechanical scarification at the micropylar end, posterior end and hot water treatment significantly improved the germinability of seeds.

Table 1 Percentage germination after 25 days, mean germination time (MGT), germination index (GI) and water imbibition in seeds of Baccaurea sapida

Baccaurea sapiaa				
Per cent germination	MGT	GI	Water imbibition	
40 ± 3.8 a 70 ± 3.1 b 99 ± 1.9 c 75 ± 3.2 bd	$14.0 \pm 0.41$ $7.6 \pm 0.53$ $4.1 \pm 0.21$ $3.6 \pm 0.23$	$2.5 \pm 0.31$ $13.2 \pm 2.6$ $40.0 \pm 3.8$ $24.5 \pm 2.05$	2 12 20 30	
	germination 40 ± 3.8 a 70 ± 3.1 b 99 ± 1.9 c	germination  40 $\pm$ 3.8 a 14.0 $\pm$ 0.41  70 $\pm$ 3.1 b 7.6 $\pm$ 0.53  99 $\pm$ 1.9 c 4.1 $\pm$ 0.21  75 $\pm$ 3.2 bd 3.6 $\pm$ 0.23	Per cent       germination $40 \pm 3.8 \text{ a}$ $14.0 \pm 0.41$ $2.5 \pm 0.31$ $70 \pm 3.1 \text{ b}$ $7.6 \pm 0.53$ $13.2 \pm 2.6$ $99 \pm 1.9 \text{ c}$ $4.1 \pm 0.21$ $40.0 \pm 3.8$ $94 \pm 0.0 \pm 0.00$ $94 \pm 0.00 \pm 0.00$	

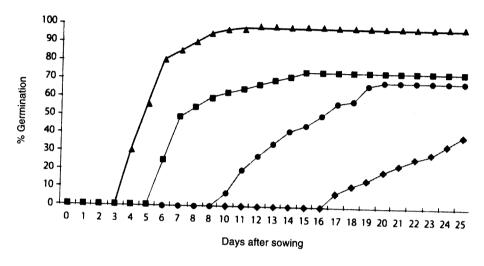
Values followed by the same letter in the same column are not significantly different at the 0.05 probability level.

The earliest onset (Figure 1) of germination (30% by day 4), highest germination percentage (99%), highest germination index (40) and second lowest germination time (4.1) were recorded in seeds scarified at the micropylar end (Table 1). The onset (Figure 1) of germination for seeds scarified opposite the micropylar end was on day 6 with 25% germination. These seeds showed the second highest germination index (24.5) and lowest germination time (3.6) (Table 1).

Presoaking of seeds in hot water improved germination (30% over the control) (Table 1) although seeds started germination quite late in day 10. These values were very close to those of seeds scarified at the posterior end. The GI and MGT of hot water soaked seeds were significantly different from those of control seeds. However, no significant difference was recorded in germination percentage in comparison with the seeds scarified at the posterior end.

Seeds scarified at the micropylar end showed highest germination. Mechanical scarification improves germination, suggesting that dormancy in seeds may be due to hardness or impermeability of the seed coat. The covering of seed can restrict germination through interfering with water uptake, gaseous exchange, escape of inhibitor, light penetration or presence of inhibitor (Bewley & Black 1982). Prasad and Nautiyal (1996) reported that mechanical scarification (through micropylar end) enhanced germinability of *Bauhinia racemosa* seeds and considerably decreased germination time. Scarification treatments in *Prosopis* sp. had been reported to improve the germination percentage. Pimente (1982) and Santos

(1985) reported improved germinability by mechanical scarification of *P. juliflora* seeds, while Leonorana and Rauledgardo (1990) reported for *P. flexuosa* and *P. alba* seeds. This study indicates that germination in seeds subjected to mechanical scarification (at micropylar end) was more homogeneous than hot water and scarification at the posterior end.



- ◆ Control
- Hot water imbibed
- Scarified at micropylar end
- Scarified at opposite to micropylar end

Figure 1 Germination of Baccaurea sapida seeds under different treatments

Failure of B. sapida seeds to attain full germination seems to be caused by interference from the seed coat. The seed coat's role as a mechanical barrier (Bewley & Black 1982) for radical emergence is excluded because of high germination percentage of seeds scarified at the micropylar end. The radicle always emerges through the micropylar end without pushing away the seed coat. Normal radicle emergence clearly shows that the seed coat prevents water from entering the seeds, thus preventing emergence of radicle because the micropylar end is covered with hard seed coat. Hard seed coat prevented radicle emergence in B. sapida seeds and delayed germination up to 17 days in the control (Figure 1). Almost no water was imbibed by the control, while hot water soaked seeds gained 12% weight and seeds scarified at the micropylar end gained 20% (Table 1). This shows that sufficient water is needed for initiating germination early as well as for maximum gain. Scarification at the micropylar end makes possible both radicle emergence and water uptake by the embryo tissues (Rolston 1978, Lopez & Aviles 1986, Rana & Nautiyal 1989, Prasad & Nautiyal 1996). However, 30% increase in seed weight was recorded in seeds scarified at the posterior end. However, the seeds failed to attain a high percentage of germination. This may be due to high percentage of water imbibition which may

have caused damage of the soft embryo cells. Bursting of the seeds may be due to high osmotic water pressure towards the micropylar end. Seeds at the beginning of the experiment had 60% moisture content.

The same set of experiment was repeated on stored (one month at room conditions) seeds, when the moisture content of seed was 10 to 15%. The water uptake by the stored seeds was around 70% in scarified seeds. About 40 to 50% of water imbibition was recorded in other treatments. However, no germination was recorded in seeds scarified at the micropylar end. This shows that the germination of *B. sapida* seeds is directly dependent on their high moisture content and possibly their recalcitrant behaviour.

Maintenance of seed viability during storage is important for the conservation of plant genetic resources not only under *ex situ* methods but also in soil seed banks in their natural habitat. Unfortunately, seeds of all plant species do not have long life span and storability. On the basis of their viability behaviours, seeds have been categorised as orthodox and recalcitrant (Roberts 1973). Seeds of other tree species such as *Quercus robur* (Finch-Savage 1992) and *Aesculus hippocastanum* (Tompsett & Pritchard 1993) were reported as desiccation sensitive and short-lived. Due to hard seed coat, seeds have low germination. Seeds of *B. sapida* should be sown within a fortnight of collection of their fruits and mechanical scarification for germination. This technique could be useful for nursery men.

## Acknowledgement

The author is grateful to E. Sharma for providing the facilities for the study.

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