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Chapter

Seed Characteristics and Germination Behaviour of *Bauhinia malabarica* Roxb.

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Abstract

Malabar *Bauhinia* (*Bauhinia malabarica*) is a native ornamental species belonging to the family Fabaceae, distributed throughout India in semievergreen and moist deciduous forests and in gardens. Information regarding seed characteristics and seed handling of the species is meagre. This study describes seed characteristics, germination behaviour and pretreatment for enhancing seed germination of *B. malabarica*. Treating the seeds with concentrated sulphuric acid for 30 min reduced germination duration up to 6 days and increased germination rate up to 100% against 22 days of germination duration and 10% germination in seeds without treatment. The results of this study are helpful for conservation and nursery practices of *B. malabarica*.

Keywords: maturity index, seed germination, germination value, pretreatment, sulphuric acid

1. Introduction

Bauhinia malabarica Roxb., commonly known as Malabar *Bauhinia*/mountain ebony, is a deciduous tree belonging to the family of Fabaceae, distributed all over the Indo-Malayan region. The species is found throughout Indian subcontinent and popularised in different vernacular names as Arampuli (Malayalam), Malayathi (Tamil), Basavana pada (Kannada), Amli (Hindi), etc. In native range, the species is distributed in the semievergreen and moist deciduous forests up to 600 m. It is used as an ornamental plant in homesteads and gardens of the native range.

Studies of the genus *Bauhinia* is very limited compared with other genus like *Caesalpinia*, *Cassia*, *Tamarindus*, etc., under the family Fabaceae. Primary documentation of the genus *Bauhinia* in India was conducted during the eighteenth century [1]. Neotypification of the species *Bauhinia malabarica* was recently done by the Central National Herbarium of Botanical Survey of India [2]. Micromorphological characteristics of selected species of the genus *Bauhinia* were studied earlier [3]. Variation in size and structure of selected species of *Bauhinia* was examined and recorded by different scholars [4, 5]. Analysis on mineral elements and nutritional and anti-nutritional contents in the seeds of *B. monandra* was conducted in an earlier study [6]. Phytochemical structure like polysaccharide and

proteins in seeds of *Bauhinia* was investigated earlier [7, 8]. Antibacterial nature of *B. acuminata* was reported in a previous study [9]. It was reported that seed oil of *Bauhinia* is a novel substance for the production of sophorolipids [10].

A few investigations were done on *B. malabarica* on foliar micromorphology, natural regeneration, chemical composition, phytochemical analysis, antimalarial activities and anti-oxidant potential [3, 11–15]. Studies related to seed germination in the genus *Bauhinia* was conducted only on selected species like *B. rufescens*, *B. thonningii*, etc. [16–18]. However, information on seed characteristics and germination parameters of *B. malabarica* is limited. The present study was done to fill the above-mentioned gaps.

2. Materials and methods

Periodical observations were carried out on mother plants of *B. malabarica* to identify optimum maturity index for seed collection. Mature pods (fruits) of *B. malabarica* were collected (March–April, 2016) from Peechi-Vazhani Wildlife Sanctuary (10°31′48″N; 76°20′50″E) in Thrissur District, Kerala, India. The pods were dried under sunlight (35–38°C) for 2 days, and seeds were extracted by splitting the pods. Extracted seeds were dried in shade, cleaned and stored in airtight containers. Seed characteristics such as morphology, moisture content and germination were evaluated. High constant temperature oven-dry method was used to determine seed moisture content [19]. The seeds were dried in hot-air oven for 1 h at 130°C. Seed moisture content (MC %) was estimated according to the formula of ISTA.

$$MC\% = \frac{fresh\ weight\ of\ seed - oven\ dry\ weight\ of\ seed}{fresh\ weight\ of\ seed} \times 100 \qquad (1)$$

Seeds were subjected to different pre-sowing treatments to enhance seed germination and reduce the germination period. The following were the pretreatments applied for the study:

T1: control (no pre-sowing treatment).
T2: soaked in water for 24 h.
T3: soaked in water for 48 h.
T4: soaked in hot water for 2 min.
T5: soaked in hot water for 5 min.
T6: soaked in hot water for 2 min + soaked in water for 24 h.
T7: soaked in hot water for 5 min + soaked in water for 24 h.
T8: soaked in GA$_3$ (500 ppm) for 2 h.
T9: soaked in GA$_3$ (1000 ppm) for 2 h.
T10: acid treatment (conc. H$_2$SO$_4$) for 10 min.
T11: acid treatment (conc. H$_2$SO$_4$) for 20 min.
T12: acid treatment (conc. H$_2$SO$_4$) for 30 min.

Tap water (≈35°C) was used in T2 and T3 treatments, whereas in the treatments T4–T7, hot water (85°C) was used. Different concentrations of gibberellic acid (GA$_3$)/gibberellin A3 (chemical formula: C$_{19}$H$_{22}$O$_6$) were used in T8 and T9 treatments. Concentrated sulphuric acid (98%) was used in the treatments T10–T12.

Seeds (n = 100 in 4 replications) were sown in germination trays having a size of 25 × 20 × 5 cm filled with vermiculite and kept in germination room (30 ± 2°C and 90% RH) under laboratory condition. Randomised block design was adopted for the experiment. Data on seed germination were recorded starting from seed germination till culmination and computed germination-related parameters. Germination initial time (GIT), germination percentage (GP), germination duration (GD), mean germination time (MGT), mean daily germination (MDG),

germination energy (GE), peak value (PV) and germination value (GV) were calculated [20–24].

Germination-related parameters were determined as follows:

$$\text{Germination initial time (GIT)} = D_g - D_s \qquad (2)$$

where D_g = first germination day and D_s = seed sowing day.

$$\text{Germination percentage (GP)} = (G/T) \times 100 \qquad (3)$$

where G = no. of germinated seeds and T = no. of seeds sown.

$$\text{Germination duration (GD)} = G_f - G_i \qquad (4)$$

where G_f = final day of germination and G_i = initial day of germination.

$$\text{Mean germination time (MGT)} = (G_t \times D_t)/G \qquad (5)$$

where G_t = no. of germinated seeds at day-t, D_t = no. of days at 't' from the day of sowing and G = total no. of germinated seeds.

$$\text{Mean daily germination (MDG)} = GP/G_d \qquad (6)$$

where GP = germination percentage and G_d = no. of days to complete germination.

$$\text{Germination energy (GE)} = X_1/Y_1 + (X_2 - X_1)/Y_2 + \ldots\ldots + (X_n - X_n - 1)/Y_n \qquad (7)$$

where X_n = no. of germinants on the nth counting date and Y_n = no. of days from sowing to the nth count.

$$\text{Peak value (PV)} = \text{Highest number of seeds germinated}/ \text{no.of days required to the peak germination} \qquad (8)$$

$$\text{Germination value (GV)} = PV \times MDG \qquad (9)$$

where PV = peak value and MDG = mean daily germination.

2.1 Statistical analysis

Each trait was analysed using mean values under the various pretreatments. The variation on mean values between these treatments were performed through analysis of variance (ANOVA) done by statistical software SPSS version 22.

3. Results

3.1 Seed weight and moisture content

The study recorded 7092 ± 50 seeds per kilogram. Moisture content (MC %) of fresh seeds was 5.35%.

3.2 Maturity index

The optimum maturity indices for seed collection of *B. malabarica* were presented in **Table 1**. The maturity indices identified for determining optimum period for seed collection were colour of pods (yellowish-green turned to blackish-green), leaf number (minimum number of leaves), dehydration (pods become dehydrated) and hardness (pods and seeds become hardest).

3.3 Pod/seed characteristics

Colour, shape, size, type, weight, number of seeds per pod and per kg, type of germination, etc. are presented in **Table 2**.

3.4 Seed germination

Figure 1 depicts germination pattern of seeds under various pretreatments. Seed germination among treatments was significantly different (**Table 3**).

Germination-related parameters under different pretreatments such as germination initial time, germination percentage, germination duration, mean germination time, mean daily germination, germination energy, peak value and germination value are given in **Table 4**.

Character	Variable	Nature
Fruit	Colour	Yellowish-green turned to blackish-green
	Water content	Very less
	Hardness	Hardened
Seed	Colour	Dark brown
	Hardness	Hardened
Leaf	Number	Minimum

Table 1.
Pod/seed maturity indices of B. malabarica.

Character	Variable	Nature
Fruit	Type	Pod
	Colour (young)	Green
	Colour (mature)	Blackish brown
	Size (cm)	15.25 ± 4.75 × 2.15 ± 0.25
	Wall type	Dry, semihard
Seed	No. of seeds/fruit	10 ± 2
	Colour	Dark brown
	Shape	Broad elliptic/oblong
	Size (mm)	.02 ± 1.11 × 6.14 ± 0.88
	Wall type	Dry, hard
	Weight	7092 ± 50
Germination	Type	Epigeal

Table 2.
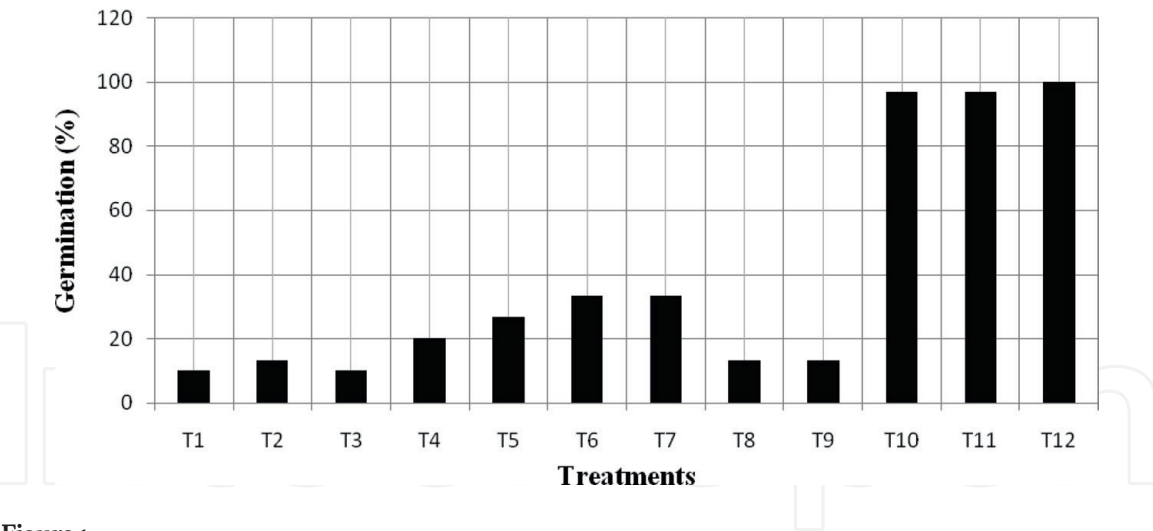*Seed characteristics of B. malabarica.*

Figure 1.
Seed germination pattern under different treatments. Note: T1, control; T2, soaked in water for 24 h; T3, soaked in water for 48 h; T4, soaked in hot water for 2 min; T5, soaked in hot water for 5 min; T6, soaked in hot water for 2 min + soaked in water for 24 h; T7, soaked in hot water for 5 min + soaked in water for 24 h; T8, soaked in gibberellic acid (GA₃—500 ppm) for 2 h; T9, soaked in gibberellic acid (GA₃—1000 ppm) for 2 h; T10, acid treatment (conc. H₂SO₄) for 10 min; T11, acid treatment (conc. H₂SO₄) for 20 min; T12, acid treatment (conc. H₂SO₄) for 30 min.

Source	Sum of squares	Degrees of freedom (df)	Mean square	F-value	Sig.
Between groups	4332.775	11	393.889	29.804	0.005
Within groups	2669.618	202	13.216		
Total	7002.393	213			

Table 3.
ANOVA table—level of significance on mean values of seed germination under different pretreatments.

Germination Initial Time (GIT): Germination initial time was 2 days after sowing in the treatments T4, T10, T11 and T12, whereas it was 5 days after sowing in T1, T2, T5, T8 and T9 treatments.

Germination Percentage (GP): 100% germination was achieved in T12 treatment (acid scarification for 30 minutes) followed by 97% in T11 and T10 (acid scarification for 20 and 10 minutes). However, 10% was noticed in TI (no treatment) and T3 (soaked in water for 48 h) followed by 13.3% in T2 (soaked in water for 24 h), T8 (500 ppm GA₃) and T9 (1000 ppm GA₃). Hot water treatments followed by water soaking (T6 and T7) exhibited about 33% germination, which was better than mere hot water treatment (T4, 20%, and T5, 27%).

Germination Duration (GD): Seed germination started 2 days after sowing and completed in 28 days. The least GD noticed in treatment T12 (06 days) followed by T11 (09 days) and T10 (11 days) and the highest in T2 (26 days).

Mean Germination Time (MGT): Mean germination time was more in T12 treatment (21.67) where the treatment gave 100% germination, whereas the least value was in seeds without any treatment (T1).

Mean Daily Germination (MDG): Mean daily germination was highest in the treatment T12 (16.67), and the lowest value was in T3 treatment (0.43).

Germination Energy (GE): Germination energy in various pretreatments showed that the highest GE was noticed in T12 (6.85), followed by T11 (5.81) and T10 (4.48). The least GE is in T3 (0.29) followed by T1 (0.30).

Peak Value (PV): Peak value was highest in T12 treatment (12.50), and least value (0.56) was noticed in treatments T1, T3, T8 and T9.

Germination Value (GV): The highest germination value was also observed in T12 (208.36), followed by T11 (116.31) and T10 (58.63).

Treatment	GIT	GP	GD	MGT	MDG	GE	PV	GV
T1	5	10	22	1.77	0.45	0.30	0.56	0.25
T2	5	13.3	26	2.23	0.51	0.42	1.11	0.57
T3	4	10	23	1.83	0.43	0.29	0.56	0.24
T4	2	20	25	3.28	0.80	0.59	1.11	0.89
T5	5	26.7	25	4.08	1.07	0.81	1.11	1.19
T6	4	33.3	25	6.56	1.33	0.78	0.67	0.89
T7	4	33.3	24	5.54	1.39	0.92	1.11	1.54
T8	5	13.3	21	2.57	0.63	0.37	0.56	0.35
T9	5	13.3	21	2.57	0.63	0.37	0.56	0.35
T10	2	96.7	11	18.27	8.79	4.48	6.67	58.63
T11	2	96.7	9	17.56	10.74	5.81	10.83	116.31
T12	2	100	6	21.67	16.67	6.85	12.50	208.36

Note: *T1, control; T2, soaked in water for 24 h; T3, soaked in water for 48 h; T4 soaked in hot water for 2 min; T5 soaked in hot water for 5 min; T6, soaked in hot water for 2 min + soaked in water for 24 h; T7, soaked in hot water for 5 min + soaked in water for 24 h; T8, soaked in GA_3 (500 ppm) for 2 h; T9, soaked in GA_3 (1000 ppm) for 2 h; T10, acid treatment (conc. H_2SO_4) for 10 min; T11, acid treatment (conc. H_2SO_4) for 20 min; T12, acid treatment (conc. H_2SO_4) for 30 min; GIT, germination initial time; GP, germination percentage; GD, germination duration; MGT, mean germination time; MDG, mean daily germination; GE, germination energy; PV, peak value; GV, germination value.*

Table 4.
Seed germination-related parameters under different treatments.

4. Discussion

Maturity indices help to collect seeds with maximum viable seeds. The optimum maturity indices of *B. malabarica* identified in the present study were the colour of pod turned from yellowish-green to blackish-brown, minimum number of leaves, pods become dehydrated and the pods and seeds become hardest. A previous study reported that seed germination of *Albizia lebbeck* significantly influenced by date of pod collection [25]. Seed weight of *B. malabarica* in the present study showed 7092 ± 50 seeds per kilogram. However, in an earlier report, it is 1100–2600 seeds per kilogram [26]. Seed size is usually related with its vigour and a measure of potential performance; hence, seed weight is significant. Information regarding seed weight and moisture content is helpful for nursery practices and research-oriented studies and also an updating of the earlier information regarding seed weight and moisture content and seed characteristics.

The present study indicated that the highest germination was recorded in acid treatments (acid scarification for 30, 20 and 10 min). All other treatments exhibited poor performance in germination. Germination initiation period was minimum in the treatments T4, T10, T11 and T12, whereas maximum was in T1, T2, T5, T8 and T9 treatments. The lowest germination initial time shows speedy initiation of germination among pretreatments.

Estimation of germination percentage is the best tool to explain seed viability of a particular lot. Seeds treated with concentrated sulphuric acid for 30 minutes (T12) resulted in very high germination rate (100%) than that of other treatments. The study also revealed that the hormonal treatment had no significant role on seed germination of *B. malabarica*. A similar result was reported in an earlier study on *B. rufescens* after acid treatment [17]. Better performance in acid scarification on seed germination was reported in seeds of many species having hard seed coat [27–29]. Germination percent

is useful for computing seed requisite for desired number of plants. Pretreatment with high germination value indicates the germination power of seeds.

Germination duration (GD) is helpful to understand the duration required for completing the process of germination. Germination duration in the present study varied with treatments (06–26 days). The study showed that the seeds scarified with concentrated sulphuric acid for 30 min helped to reduce germination period into 6 days compared to 26 days in seeds soaked in water for 24 h. The lowest GD shows the minimum period required to complete germination among pretreatments. Mean germination time (MGT) is the indicative of emergence performance of seed lots. Mean germination time (MGT) was highest in seeds treated with concentrated sulphuric acid for 30 min (21.67) where the treatment gave 100% germination, whereas the least value was in seeds without any treatment (T1). Similarly, mean daily germination (MDG), germination energy (GE), peak value (PV) and germination values were also highest in the treatment T12.

Mean germination time and mean daily germination are used as a gauge of the rate and time spread of germination. High MGT and MDG values indicate high germinability of seed lots due to pretreatments. Peak value indicates the maximum germination rate in a particular day, and germination value is the expected seedlings in the field or nursery. Germination energy and germination value are the easier way to understand the rate of germination and period of germination. Highest GE and GV show the enhanced germination and reduced duration.

The study resulted in scarification of seeds by concentrated sulphuric acid for 30 min which was the best pre-sowing treatments for enhancing seed germination and reduce germination period in *B. malabarica*. Previous studies showed that acid scarification is the best pretreatment to improve germination of seeds with hard seed coat [27–29]. Similarly, parameters like germination percentage, germination energy, mean germination time, mean daily germination, peak value and germination value were also highest in the acid treatment for 30 min compared to other treatments. High values of parameters indicate the germination potential of the seeds. Germination energy/germination value is the tool for indexing the speed and completeness of seed germination [22]. High germination energy and germination value show the effects of pretreatment on seed germination. Period of seed germination in *B. malabarica* is reported in an earlier study with 6–30 days and hot water treatment for 1 min followed by soaking in cold water for 24 hours as the best pretreatment [26]. Similarly, a high rate of germination observed in seeds of *Hippophae salicifolia* treated with thiourea [30]. The Forest Research Institute, India, reported only a low rate of seed germination (14–18%) in *B. malabarica* [31]. However, the present study revealed that the concentrated sulphuric acid treatment for 30 min shall reduce germination period (6 days) and increase germination rate (100%). An earlier study on *Macaranga peltata* revealed that combination of concentrated sulphuric acid and gibberellic acid resulted in improved germination rate [32].

5. Conclusions

The study gave update to seed characteristics and germination behaviour of *B. malabarica*. The investigation has documented maturity indices for determining optimum seed collection period of the species *B. malabarica*, and the data is very helpful for further seed biological studies and experiments on the species. The study suggested optimum period for collection of seed of *B. malabarica* is when yellowish-green colour of pods turned to blackish-green, dehydrated and hard. The study recommended that concentrated sulphuric acid treatment for 30 min is the best pretreatment for enhancing seed germination and reducing germination period.

Acknowledgements

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