



## **Assessment of Seed Germination Behaviour of Endangered Medicinal tree *Aegle marmelos* (bael) through Hydropriming technique: A Conservational approach**

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### **ABSTRACT**

*Hydropriming is known as a seed pre-treatment which increases antioxidant enzymes such as glutathione and ascorbate. These enzymes decrease the activities of lipid peroxidation in the stage of germination. As a result, increases the percentage of germination and improves seed performance under environmental as well as laboratory conditions. Seeds of *Aegle marmelos* (bael) were treated as following: (i) unsoaked seed (control); (ii) hydropriming for 48 h; (iii) hydropriming for 96 h, and (iv) hydropriming for 144 h as T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub>, respectively. The seeds were soaked in distilled water at 26 °C except control. The hydro primed seeds for 96 h at 26 °C showed best germination (90.00%). ANOVA and DMRT showed significant results for all applied hydro primed pre-treatments in measured parameters than unprimed seeds. The evaluated healthy normal seedlings at germinator were transplanted to the environmental condition for seedling acclimatization, establishment and growth. Therefore, the objective of this study was to find out the effective hydropriming treatments for enhance germination, uniform seedling emergence along with short germination period and accelerating the regeneration potential of the species. It is a simple, inexpensive and convenient method of improving and obtaining uniform germination and seedling establishment for raising nursery conserve its diversity in the nature.*

**Key words:** RET species, hydropriming pre-treatment, seed germination, seedling vigour index, seedling acclimatization, seedling survival.

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### **INTRODUCTION**

Seed hydropriming (partially hydrates the seed, then seeds are dried) is known as seed pre-treatment along with maintenance of seed hydration level and essential metabolic activities needed for initiation of germination, but radicle emergence is avoided. Hydropriming improves seed performance under environmental conditions. It is safe, simple and inexpensive method to enhance early seed germination, faster emergence, better stands, increased plant vigour and drought tolerance, earlier flowering and harvest and higher yields. Seed priming is commonly used to reduce the time between seed sowing and seedling emergence, successful in improving germination and stand establishment [1, 2]. It is an invigoration technique to enhance seed germination, increasing final germination, accelerating the synchronized seedling, vigorous seedling establishment, stimulating vegetative growth and yield and seed tolerance to adverse environmental condition [3]. Notably, hydropriming studies have been observed in cereals, vegetable and flower species [4-10, 11] but limited on medicinal tree species.

*Aegle marmelos* (L.) Corr. is a popular medicinal tree belonging to the family Rutaceae commonly known as Bael tree, it is a moderate sized, slender, aromatic tree, 6.0 -7.5 m in height, and 90 to 120 cm in girth, with a somewhat fluted bole of 3.0-4.5 meter growing wild throughout the deciduous forests of India, ascending to an altitude of 1200 m in the western Himalayas and also occurring in Andaman island [12]. Its various parts are used as herbal medicine to treat a variety of ailments [13] roots are useful for treating diarrhea, dysentery and dyspepsia, aqueous extracts of the stem and root bark are used to treat malaria, fever, jaundice and skin diseases such as ulcers, urticaria and eczema [14] antiamebic and hypoglycaemic activities [15] antiulcer activity [16] antifungal activity [17].

However, the Foundation for Revitalization of Local Health Traditions (FRLHT), Bangalore, India assessed threat status of the tree as rare, endangered and threatened (RET) species, especially endangered species not only in India but also in the world and going to be extinct in the nature and normally grown with seeds in the nature [18]. Hence, its natural regeneration is very low and erratic in the nature due to its hard hydrophobic fruit cover (shell); in this way, seeds having remained close inside the fruits cavity and seeds are unable to emerge out for germination, ultimately seeds are deteriorate due to fungal infection during the rainy season. Notably, a few research documentation available on germination performance using hydro-primed technique in *A. marmelos* (bael) seeds.

Ultimately, an argent need for rapid uniform seed germination along with vigorous and synchronized seedlings establishment using the following objectives: i) to evaluate the effects of hydropriming treatments on seed germination behaviour under laboratory conditions, ii) to standardize the seed germination behaviour for raising nursery, and iii) to identify morphological traits for the assessment of quality seedlings.

## MATERIALS AND METHODS

This study was carried out at the Seed Testing Laboratory, Department of Seed Science and Technology, HNB Garhwal University (A Central University), Srinagar Garhwal, Uttarakhand (India).

### Seed source and collection

The freshly matured and healthy fruits (yellowish in colour) of *A. marmelos* were harvested from healthy well growing plants from Chauras campus (about 650 m asl) of HNBU University, Srinagar Garhwal (Uttarakhand) India in the month of may-June, 2013. Seeds were extracted by breaking the fruits by hitting with a wooden mallet followed by washed in water for 30 minutes to remove the mucilage pulp. The seeds were then air-dried on newspaper under fan and stored for six months of storage in a cotton bag at room temperature. After completion of six months the quality seeds were taken from seed lot for conducting the experiment trial. Seed viability was observed with TZ test (a quick viability test); both immediately after collection (97%) and at the germination tests setup (90%) using 1% solution of tetrazolium chloride salt [19].

### Experimental design

Well dried quality seeds were used for germination experiment. The seeds were observed manually to eliminate stained, discoloured, wrinkled and damaged seeds then dipped in 0.5% (w/v) mercuric chloride (HgCl<sub>2</sub>) solution for 5-6 minutes followed by five times washing thoroughly double distilled water under aseptic conditions for complete sterilization. The washed seeds were fully immersed (1:2 w/v) in the distilled water under aseptic conditions and kept for different time intervals according to the experiment setup. We used four treatments, i.e., T<sub>1</sub>= unsoaked seed (control); T<sub>2</sub>= water soaking for 48 h (2 days); T<sub>3</sub>= water soaking for 96 h (4 days); and T<sub>4</sub>= water soaking for 144 h (6 days) at 25 ± 1 oC temperature in incubator. After water soaking, seeds were redried at the shade place for 24 hours. The germination trial was carried out according to the experimental design by sowing the seeds in top of the paper (TP) using Petri-dishes contained water soaked (Distilled water) Whatman No.1.filter paper and between the papers (BP) for analyzing the seedling growth (root and shoot) measurement in germinator at 26 ± 2 oC temperature with 80% humidity with dark condition, respectively. 400 seeds (100 seeds/replicate) were used to perform the germination experiment both in TP and BP, respectively [20]. The experiment was continuously observed within 24 hours interval and continued until no further germination occurred till. Radicle protrusion of 2 mm was scored as germination [11]. The healthy and normal evaluated seedlings were transplanted from seed germinator to the field (environmental condition) into plastic trays contained FYM and top soil (medium; 2:1 ratio) where the average maximum and minimum temperature was 30 ± 2 oC and 25 ± 2 oC, respectively along with average maximum and minimum relative humidity was 80 ± 2% and 60 ± 2%, respectively. for seedling acclimatization, establishment, growth and planting value assessment. A completely randomized design (CRD) with four replicates was used to analyze the experiment statistically.

### Seed germination parameters

The number of seeds germinated in each treatment was recorded regularly. At the end of germination test different seed germination parameters were calculated using the following formulas:

a) Mean daily germination (MDG) is an indicator of daily germination rate calculated as,

$$[21] \quad MDG = \frac{FG\%}{D}$$

b) Vigour index-I = Seedling length (cm) × Final germination (%) [22]

- c) Vigour index-II = Final germination (%) × Seedling dry weight (gm) [22]
- d) **Final germination (%)** =  $\frac{\text{Number of germinated seeds}}{\text{Total number of seeds}} \times 100$  [23]
- e) Allometric trait = the length of radical / the length of plumule [24]
- f) Germination value = MDG × peak value of germination [25]
- g) Germination index (GI) = Total germination % / Time (hr) taken for 50 % germination [26]
- h) **Seedling survival %** =  $\frac{\text{Number of seedling survive}}{\text{Total number of seedling}} \times 100$
- i) Speed of germination =  $\frac{X_1}{Y_1} + \frac{X_2 - X_1}{Y_2} + \dots + \frac{X_n - X_{n-1}}{Y_n}$  [27]

Where,  $X_1, X_2$  and  $X_n$  are numbers of seeds germinated on first, second and nth day, respectively whereas,  $Y_1, Y_2$  and  $Y_n$  are number of days from sowing to first, second and nth count, respectively.

j) **Seedling growth rate (SGR)** =  $\frac{SL_1}{F_1} + \frac{SL_2 - SL_1}{F_2} + \dots + \frac{[SL_n - SL_{(n-1)}]}{F_n}$  [28]

Where,  $SL_1$  = the mean seedling length at first count;  $SL_2$  = the mean seedling length at second count;  $SL_1 - SL_2$  = the mean increase in length in second count;  $F_1$  = the days to first count;  $F_n$  = the days to final count and  $n$  = the number of germinated seeds

**Seedling length (shoot and root) (cm):** Ten normal seedlings were selected randomly and measured the shoot and root length of them. The total length of seedling was obtained by adding root and shoot length for each treatment.

**Seedling Growth measurement (mg):** Plants growth was estimated by measuring accumulation of root and shoot weight (after drying the plants material at 70 °C for 48-72 h.) Relative water content (RWC) was also measured and expressed as a percentage according to the following equation:

$$\text{RWC (\%)} = \frac{\text{FW} - \text{DW}}{\text{FW}} \times 100$$

Germination period (GP), the germination percentage (G %) and mean daily germination (MDG) were obtained using the following equations [29] modified from [27] and [21], respectively. Germination index is defined as cumulative daily total of germination over specific days [30]. Speed of germination, totality and their interaction is also described as germination index [31]. Germination rate is defined as number of seeds of particular variety likely to germinate over a specific period of time. Germination velocity index is defined as daily germination counting for estimation of seedling vigour [32]. Seed vigour is cumulative properties of seed which determine the quick and uniform emergence potential of seed and followed by potential seedling development under diverse field conditions [23].

#### Statistical analysis

Analysis of variance (ANOVA) was carried out at 1% and 5% level of significance using the SPSS software (version-20) to explore possible variation in the effect of hydropriming. Whereas, means were compared using Duncan's multiple range test (DMRT) [33] at 5% level of probability.

## RESULTS

The findings of the present study are being presented in this chapter under the following observational headings:

### Seed morphology

The fresh seeds of *A. marmelos* (bael) were rough and creamy white in coloured which covered with light yellow transparent mucilage pulp in the fruit cavities. The seeds were spherical flat round and their average diameter was  $0.5 \pm 0.09$  cm and thickness  $0.2 \pm 0.1$  cm.

### Assessment of hydropriming effects on the seedling planting value

According to the ANOVA one way results, the hydropriming showed significant effect on germination starting date (GSD), germination closing date (GCD), germination period (GP) and mean daily germination (MDG) at  $p < 0.01$  level of significance and the final germination percentage had significant effect at  $p < 0.05$  level (see Tab. 1). The 144 h soaked seeds with distilled water ( $T_4$ ) were able to begin the germination on the 5th day after seed sowing and followed by 6th, 6th, and 10th days in 96 h soaked seeds ( $T_3$ ), 48 h soaked seeds ( $T_2$ ) and unsoaked seeds ( $T_1$ ) and finally they reached 87.50, 90.00, 78.25 and 74.00% final germination at the end of the experiment, respectively. The DMRT (Duncan Multiple Range Test) analysis showed no significant effect in final germination percentage in  $T_4$  but significant than  $T_1$  (control) (Tab.1). The  $T_2$  and  $T_3$  treatment showed minimum days for seed germination (GSD) than unsoaked control but DMRT showed no significant effects ( $p > 0.05$  level) among the  $T_2, T_3$  and  $T_4$  whereas significant than control. The minimum GCD found in  $T_4$  which was significant (at  $p < 0.05$  level) than other three hydropriming treatments. The minimum GP recorded in  $T_4$ , it was also significant at DMRT 0.05 level than  $T_2$  and  $T_3$  along with control (unsoaked seeds). The maximum MDG (5.28) was recorded in  $T_4$ , it

showed significant at  $p < 0.05$  level in DMRT followed by 4.67, 4.07 and 2.92 in T<sub>3</sub>, T<sub>2</sub> and T<sub>1</sub>, respectively (see Tab.1).

ANOVA and DMRT showed no significant effect on the some parameters of evaluated seedling i.e., root length, shoot fresh weight, root fresh weight, seedling fresh weight, relative water content (see Tab.2). The maximum shoot length was recorded in 48 h soaked seeds (9.25 cm) showed significant effect than other hydropriming treatments followed by 96 h soaked seeds (8.27 cm), unsoaked seeds (8.11 cm). There was no significant effects between T<sub>1</sub> and T<sub>3</sub> but showed significant effect than T<sub>4</sub>, the analysis of variance showed significant effects among the treatments ( $p < 0.05$  level) (Tab.2 & Fig. 1 D-F). The maximum root length was recorded in T<sub>1</sub> (6.04 cm) but it was not significantly difference than other treatments. 48 h soaked seeds showed maximum seedling length but mean value was not significant (DMRT, at  $p < 0.05$  level) than unsoaked seeds but significant than T<sub>3</sub> and T<sub>4</sub>, ANOVA showed significant effects among the treatments (see Tab. 2). The maximum shoot dry weight was recorded in T<sub>3</sub> (0.20 gm) which showed significant effect at DMRT 0.05 level but ANOVA showed no difference among them. Root dry weight was same for T<sub>3</sub> and T<sub>4</sub> as (0.10 gm) which was significant than T<sub>1</sub> and T<sub>2</sub>, the ANOVA showed significant effects ( $p < 0.01$  level) among the treatments. The seedling dry weight was found maximum in T<sub>4</sub> but showed no significant effect than T<sub>3</sub>, significant than T<sub>1</sub> and T<sub>2</sub>, analysis of variance showed significant effects ( $p < 0.01$  level) among the treatments (see Tab. 2). The maximum vigour index I was recorded in T<sub>3</sub> (1236.94) but showed on significant effect than T<sub>1</sub> and T<sub>2</sub> (DMRT, at  $p < 0.05$  level), it was significant for T<sub>4</sub>, ANOVA showed no significant effects among the treatments. The vigour index II was maximum in T<sub>4</sub> followed by T<sub>3</sub> as 27.52 and 29.99, respectively, both showed significant effect than T<sub>1</sub> and T<sub>2</sub>, the analysis of variance showed significant effect among the treatments ( $p < 0.01$  level). The speed of germination was maximum in T<sub>4</sub> ( $p < 0.05$  level DMRT) followed by T<sub>3</sub>, T<sub>2</sub> and T<sub>1</sub>. ANOVA showed significant effect ( $p < 0.01$  level) for speed of germination, germination velocity and germination index among the treatments. DMRT showed significant effects ( $p < 0.05$  level) for germination velocity and germination index in T<sub>3</sub> and T<sub>4</sub> (see Tab. 3).

#### Effects of hydropriming on seedling parameters at environmental condition

Perusal the data in Tab. 3, that all the hydropriming pre-treatments showed no significant effects on the seedling survival percentage at environmental condition at DMRT ( $p > 0.05$  level) and ANOVA ( $p > 0.05$  and  $p > 0.01$  level of significance). The maximum seedling survival percentage was recorded in T<sub>3</sub> and T<sub>4</sub> but DMRT showed no significant effects ( $p > 0.05$  level) among the mean value of all the treatments. Seedling growth rate was found maximum (0.96) in 48 h soaked seeds followed by 0.87, 0.85 and 0.78 in T<sub>3</sub>, T<sub>1</sub> and T<sub>4</sub>, respectively (see Tab. 4 & Fig. 1 C-F). DMRT showed significant effects ( $p < 0.05$  level) for T<sub>2</sub> than other treatments whereas, ANOVA showed significant effects ( $p < 0.05$  level) among the treatments (see Tab. 4). The maximum seedling (root and shoot) length and leaves per seedling was recorded in unsoaked seeds (T<sub>1</sub>) but DMRT and ANOVA showed no significant effects ( $p > 0.05$  level) however, ANOVA showed significant effects ( $p < 0.05$  level) among the seedling root length.

**Table: 1.** Summary of hydropriming effects on specific expressions of seed germination of *Aegle marmelos* at laboratory condition.

T	GSD (d)	GCD (d)	GP (d)	FG (%)	MDG
T1	10.00±0.82b	26.25±1.26c	16.25±1.26b	74.00±3.65a	2.92±0.16a
T2	6.00±0.82a	19.25±0.96b	13.25±0.96a	78.25±2.36ab	4.07±0.25b
T3	6.00±0.82a	19.25±0.96b	13.25±0.96a	90.00±10.80b	4.67±0.74bc
T4	5.00±0.82a	16.75±1.50a	11.75±0.96a	87.50±9.57b	5.28±0.96c
F-value	29.500**	47.235**	13.154**	4.026*	10.459**
Sig.	0.000	0.000	0.000	0.034	0.001

**Where:** T= treatment; T<sub>1</sub>= unsoaked seeds (control); T<sub>2</sub>= hydropriming with distilled water for 48 h; T<sub>3</sub>= hydropriming with distilled water for 96 h; T<sub>4</sub>= hydropriming with distilled water for 144 h; GSD= germination starting date; GCD= germination closing date; GP= germination period; FG= final germination percentage; MDG= mean daily germination.

**Table 2.** Summary of hydropriming effects on the seedling parameters measurement in *Aegle marmelos* (Bael) in seed germinator after 25th days from date of sowing (at  $26 \pm 2$  °C temperature with 80% humidity with dark condition).

T	Shoot L (cm)	Root L (cm)	Seedling L (cm)	Shoot FW (gm)	Root FW (gm)	Seedling FW (gm)	Shoot DW (gm)	Root DW (gm)	Seedling DW (gm)	RWC (%)
T1	8.11±0.43ab	6.04±0.63a	14.15±0.78b	1.83±0.36a	0.75±0.08a	2.59±0.36a	0.19±0.03a	0.09±0.01a	0.28±0.03ab	89.10±2.21a
T2	9.25±0.68b	5.88±0.33a	15.13±0.55b	1.83±0.20a	0.70±0.07a	2.52±0.16a	0.19±0.01a	0.08±0.01a	0.27±0.01a	89.22±0.79a
T3	8.27±1.10ab	5.63±0.93a	13.90±1.97ab	1.85±0.18a	0.79±0.01a	2.63±0.19a	0.20±0.01ab	0.10±0.00b	0.30±0.01bc	88.61±0.52a
T4	7.04±0.80a	4.97±0.71a	12.01±1.49a	1.97±0.23a	0.80±0.07a	2.77±0.30a	0.22±0.02b	0.10±0.00b	0.32±0.01c	88.48±1.48a
F-value	5.186*	1.910ns	3.879*	0.270ns	2.134ns	0.596ns	2.929ns	6.759**	5.618*	0.261ns
Sig.	0.016	0.182	0.038	0.846	0.149	0.629	0.077	0.006	0.012	0.852

**Where,** T= treatment; T<sub>1</sub>= unsoaked seeds (control); T<sub>2</sub>= hydropriming with distilled water for 48 h; T<sub>3</sub>= hydropriming with distilled water for 96 h; T<sub>4</sub>= hydropriming with distilled water for 144 h; RWC% = relative water content percent.

**Note:** \* and \*\* mean significant at  $p < 0.05$  and  $p < 0.01$  levels of significance, respectively; ns: not significant.

**Table 3.** Assessment of seedling physiological measures in *A. marmelos* (Bael).

T	Vigour Index I	Vigour index II	AT	SG	GV	GI
T1	1080.90±49.43ab	21.16±2.50a	0.75±0.09a	4.59±0.13a	12.95±0.87a	0.17±0.00a
T2	1182.67±20.20ab	21.25±1.14a	0.64±0.086a	9.66±0.30b	39.33±2.75b	0.41±0.01b
T3	1236.94±84.30b	26.99±3.63b	0.68±0.05a	10.19±1.49b	48.55±13.45bc	0.44±0.07bc
T4	1051.23±175.09a	27.52±2.09b	0.70±0.04a	12.21±1.94c	65.76±23.37c	0.53±0.11c
F-value	2.964ns	7.800**	1.834ns	27.668**	10.573**	21.749**
Sig.	0.075	0.004	0.195	0.000	0.001	0.000

**Where,** T= treatment; T<sub>1</sub>= unsoaked seeds (control); T<sub>2</sub>= water soaking for 48 h (2 days); T<sub>3</sub>= water soaking for 96 h (4 days); T<sub>4</sub>= water soaking for 144 h (6 days); AT= allometric traits; SG= speed of germination; GV= germination velocity; GI= germination index.

**Note:** \* and \*\* mean significant at  $p < 0.05$  and  $p < 0.01$  levels of significance, respectively; ns: not significant.

**Table 4.** Summary of hydropriming treatment effects on specific expressions of seedling parameters of *A. marmelos* (bael) at environmental condition.

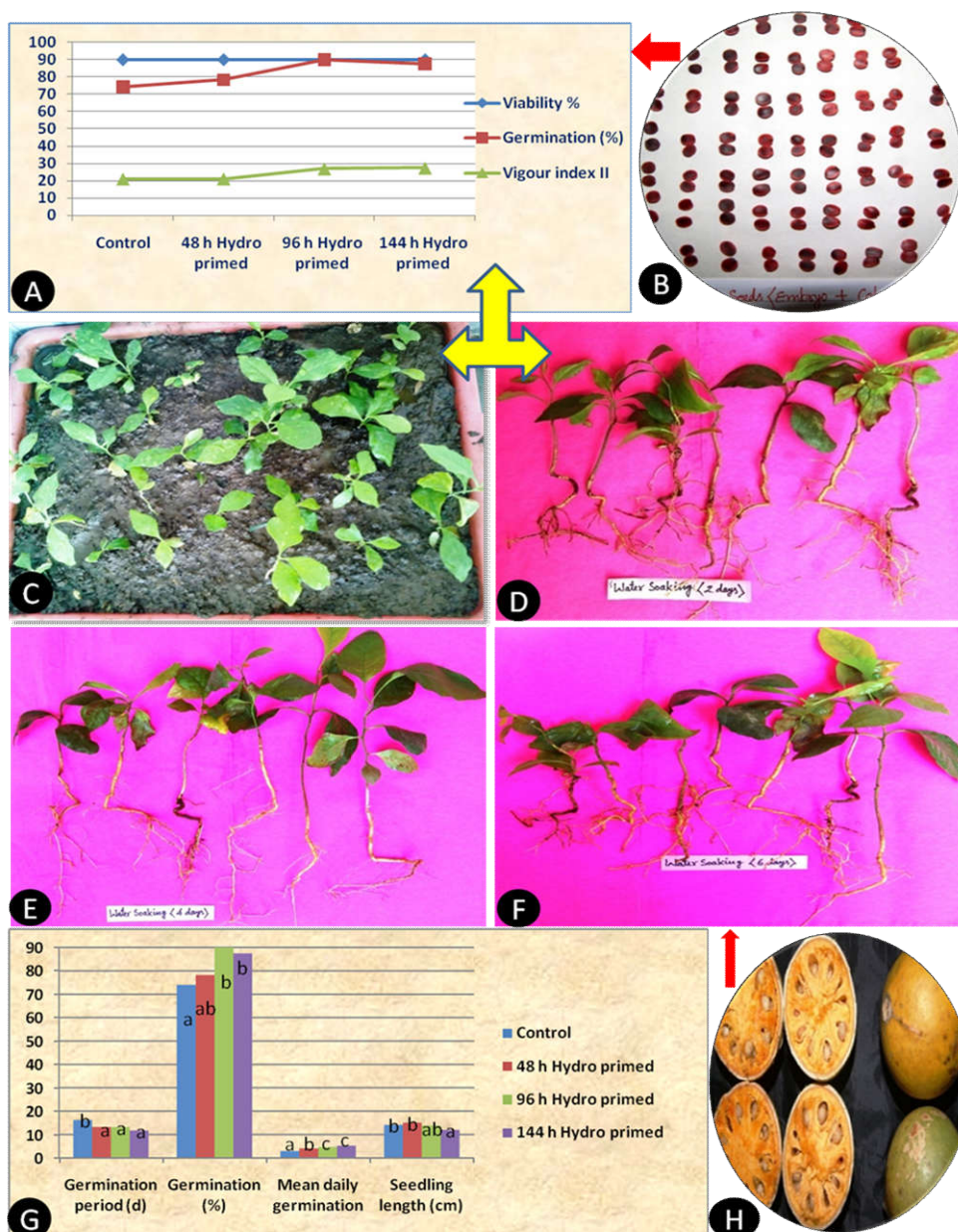
T	SS%	SGR	Root L (cm)	Shoot L (cm)	Seedling L (cm)	Leaves/Seedling
T1	95.25±2.50a	0.85±0.01b	11.00±0.91b	13.98±0.80a	24.98±1.71b	7.25±0.96ab
T2	96.5±1.29a	0.96±0.01c	10.83±1.02b	13.78±1.00a	24.60±0.41ab	6.25±0.96a
T3	97.25±0.96a	0.87±0.02b	10.75±1.04b	13.08±0.76a	23.83±1.76ab	7.00±0.82ab
T4	97.00±1.41a	0.76±0.01a	8.83±0.33a	13.53±1.58a	22.35±1.36a	8.00±0.82b
F-value	1.169ns	136.031**	5.447**	0.511ns	2.685ns	2.632ns
Sig.	0.362	0.000	0.013	0.682	0.094	0.098

**Where,** T= treatment; T<sub>1</sub>= unsoaked seeds (control); T<sub>2</sub>= water soaking for 48 h (2 days); T<sub>3</sub>= water soaking for 96 h (4 days); T<sub>4</sub>= water soaking for 144 h (6 days); SS%= seedling survival percentage and SGR= seedling growth rate.

**Note:** \* and \*\* mean significant at  $p < 0.05$  and  $p < 0.01$  levels of significance, respectively; ns: not significant.

## DISCUSSION

Seed germination and seedling growth are critical phase of regeneration at the initial life stages and often subjected to high mortality rates. The three early phases of germination are: (i) imbibition, (ii) lag phase and (iii) protrusion of the radical through the testa (seed coat). Priming may be helpful in reducing the risk of poor stand establishment under laboratory as well as in nursery conditions. It is a procedure that partially hydrates seed, followed by drying of seed for a time period, therefore germination processes begin, but radicle emergence does not occur. There are reports that hydration of seed up to, but not exceeding, the lag phase with priming permits early DNA replication [34] increased RNA and protein synthesis [35, 36] greater ATP availability [37] repair of deteriorated seed parts [38, 39]. Hydration helps radicle protrusion through the seed coat and shortens the time to seed germination. It also improved seed performance might be attributable in part to the decreased lipid peroxidation and increased antioxidative activities during seed imbibitions.



**Fig. 1. Propagation behaviour along with seedling growth measurement of *A. marmelos* (bael).**

**A.** Graph showed seed viability along with germination and vigour; **B.** Assessment of seeds viability; **C.** Emerged seedling (under environmental condition) in plastic tray; **D.** Evaluated seedlings under water soaking pre-treatment for 48 h (2 days); **E.** Evaluated seedlings under water soaking pre-treatment for 96 h (4 days); **G.** Graph showed germination along with GP, MDG and seedling length; **F.** Evaluated seedlings under water soaking pre-treatment for 144 h (6 days), and **H.** Matured seeds in fruit cavity.

In the present study it was reported that, hydro primed seeds of *A. marmelos* showed significant increased in germination performance along with resultant effect on seedling planting value depends on the adopted method of hydropriming technique and duration of pre-sowing treatment. Similarly, previous study observed similar observation on the hydroprimed technique [40]. Notably, the effects of seed hydropriming (48 h, 96 h and 144 h water soaking) on *A. marmelos* (bael) showed faster emergence and recorded final germination as 78.25, 90.00 and 87.50%, respectively and improved other morphological traits significantly than unprimed seeds it was so that hydropriming allows the hydration of membranes and proteins, and the initiation of various metabolic systems along with germination potential to high level before germination period. The presoaking of seeds is arrested when the seeds are dried or moisture is withheld, but recommence when the seeds imbibe water for the second time [41]. Ashraf and Rauf [42] reported that final germination percentage, fresh and dry weight of corn seed increased by seed priming significantly. So due to all this information and present research has been clearly noticed that most effects of seed priming is due to seed hydration (hydropriming). So,

optimization of priming technique is very important to achieve the best time and concentration combination that leads higher and faster germination along with seedling uniformity and field stand. Total seedling emergence percentage showed significantly higher emergence in primed seeds than unprimed seeds in *A. marmelos*. Similarly, Ghassemi et al. [43] in lentil, Hosseini and Kasra [44] in basil and Ramesh [45] in Pungam seed reported improved germination rate, root weight compared to unprimed and chemo primed seed treatment. Seeds of *A. marmelos* with hydro primed for 48 h which recorded significantly higher germination percentage (90.00%) than control (unsoaked seeds) (74.00%). Venudevan and Srimathi [46] was found similar result 80.00% germination was found in the seeds primed with 9 h water soaked treatment. The present results revealed that the germination percentage increased when the seeds were put for maximum imbibition period, this was due to hydration of the seeds and at same time hydrolytic enzymes were activated in the endosperm converting complex stored food materials into metabolically useful chemicals that resulted in growth of the embryo. During the water imbibition period temperature (at  $25 \pm 1$  °C) also plays an important role for the pre-germination activities and radicle emergence consistently, similarly. Similarly, Daniel et al. [47] reported that respiration, radicle protrusion and cell division occurred sooner in primed seeds as compared to non primed seed when they were imbibed at 25 °C. Similarly, Fujikura et al. [48] has described hydropriming as a simple and inexpensive method of seed priming.

*A. marmelos* has tremendous medicinal as well as aesthetic properties and high dependency by the human being on the plants, this has led to a loss of biodiversity in bael-growing areas and consistently this medicinal tree has facing threads status on its natural regeneration, similarly, due to hard fruit covering and its hydrophobic nature. It was also observed during the experiment the fully hydrated seeds (high level of moisture content) were able to fast germinate and uniform seedling emergence than less imbibed and control that denotes the seeds may be have a partially recalcitrant nature that delay fast germination and required maximum time for field emergence. Similarly, the same results were seen by Venudevan and Srimathi [46] that *A. marmelos* seeds required maximum hydration level to fast germination. Therefore, optimal yield could be achieved by fast germination and uniform emergence at the nursery. This implies that hydro priming is the key factor to enhance germination, uniform seedling emergence and resistance to unfavourable environmental factors that inherit seed germination (light, temperature and water).

## CONCLUSION

Seeds of *A. marmelos* hydro primed for 96 h were recorded significantly higher germination percentage (90.00%) along with all seedling quality parameters within short germination period than control (unsoaked seeds). Furthermore, the results showed that hydropriming improves seed germination characteristics of the bael seeds in the laboratory and field conditions.

Therefore, this method of the hydropriming techniques can be used to promote sustainable cultivation of *A. marmelos* in the most of the areas of the world with raising uniform healthy seedlings at nurseries. Since this method of priming is a simple, inexpensive, and therefore does not require chemicals to the farmers offered to the emergence of this plant have a higher percentage and uniformity.

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