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SEED BIOLOGY, GERMINATION BEHAVIOUR AND NURSERY MANAGEMENT OF GARCINIA GUMMI-GUTTA (L.) N. ROBSON

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ABSTRACT
Garcinia gummi-gutta (L.) N. Robson is an economically and medicinally important tree species distributed in the Western Ghats of India. The present study investigated the taxonomy, seed morphology, physiology, phytochemistry, storage behaviour, and germination characteristics of the species collected from different locations in Western Ghats. Morphological studies revealed that the species produces yellow segmented berries containing 5–9 elongated ovoid seeds with high moisture content (39.20–39.22%). Histological observations indicated a multilayered hydrophobic seed coat and lipid-rich seed tissues. Seed viability assessed through tetrazolium testing showed high viability (94.67%). Water uptake studies demonstrated gradual imbibition reaching 27.13% after 96 h.

Fresh seeds exhibited better germination than stored seeds in all sowing media. Among different sowing media, SM1 showed superior germination performance with higher germination percentage, germination rate, and germination value along with lower mean germination time. Pretreatment significantly influenced germination characteristics, and seeds with seed coat removal (T2) exhibited the highest germination percentage (93.25%) and germination value (0.426). Root trainers performed better than pots by reducing mean germination time and improving germination rate. Seed storage significantly reduced germination performance, confirming the recalcitrant nature of the seeds. Statistical analysis through two-way ANOVA revealed significant effects of sowing medium, pretreatment, container type, and storage period on germination parameters.

The study demonstrates that successful propagation of Garcinia gummi-gutta can be achieved using freshly extracted seeds, seed coat removal pretreatment, suitable sowing media, and root trainer containers. The findings provide valuable baseline information for conservation, nursery production, and sustainable utilization of this important indigenous species.

KEYWORDS: Garcinia gummi-gutta, seed biology, germination, recalcitrant seeds, nursery techniques, seed pretreatment, sowing medium

INTRODUCTION
Garcinia gummi-gutta (L.) N. Robson, belonging to the family Clusiaceae, is an important evergreen tree species native to the tropical forests of the Western Ghats of India (Scaria, et. al., 2025). The fruit rind is traditionally used as a souring agent in South Indian cuisine, especially in fish curry preparations, and also possesses considerable medicinal and nutraceutical importance. Various parts of the plant are traditionally used in indigenous medicine for treating digestive disorders, rheumatism, respiratory ailments, ulcers, and obesity-related conditions (Kaprakkaden & Ali, 2025). In recent years, increasing demand for the fruit and its phytochemical constituents has enhanced the commercial importance of the species. Despite its economic significance, natural regeneration of Garcinia gummi-gutta is often poor due to low seed storage potential and delayed germination (Vishnu, et. al., 2025; Bohra, Waman, & Karthika Devi, 2021; Bohra, Waman, & Karthika Devi, 2025). Propagation through seeds remains the most common method; however, successful seedling establishment is influenced by seed physiology, seed coat characteristics, sowing medium, storage conditions, and nursery practices.

Seeds of many tropical tree species exhibit recalcitrant storage behaviour characterized by high moisture content and sensitivity to desiccation (Lah, et. al., 2023). Such seeds lose viability rapidly during storage and require immediate sowing after collection. Understanding seed biology and germination ecology is therefore essential for developing suitable propagation and conservation strategies. Earlier studies on Garcinia species have primarily focused on phytochemistry and medicinal applications, while detailed investigations on seed anatomy, physiology, germination behaviour, and nursery techniques remain limited.

The present investigation was undertaken to study the taxonomy, fruit and seed morphology, seed physiology, phytochemical composition, viability, storage behaviour, and germination characteristics of Garcinia gummi-gutta. The study also aimed to evaluate the influence of sowing media, seed pretreatments, storage duration, and container type on germination performance to develop suitable nursery practices for large-scale propagation and conservation of the species.

2. MATERIALS AND METHODS
2.1 Study Area and Plant Material Collection
Mature trees of Garcinia gummi-gutta were identified from Cherai, Koratty, and Thuravoor regions of Western Ghats, India during the period from 2018 to 2025. Tree height, girth, and geographical coordinates were recorded. Fruits were

collected during the fruiting season from June to August. Ripened fruits were harvested directly from trees using poles fitted with collection nets, and freshly fallen fruits were also collected using tarpaulin sheets spread beneath the canopy.

2.2 Fruit and Seed Processing
Collected fruits were washed thoroughly under running water. The rind was removed manually, and seeds were separated carefully from the pulp using a blunt knife to avoid damage. Seeds were washed thoroughly to remove the pulpy aril and shade dried for three days before further analysis and storage experiments.

2.3 Morphological and Anatomical Studies
Fruit and seed morphology were studied using fresh samples collected from Koratty (L1) and Cherai (L2). Parameters such as fruit length, width, weight, seed number per fruit, seed length, width, thickness, and seed weight were recorded. For anatomical studies, transverse and longitudinal sections of fruits and seeds were prepared manually and observed under a light microscope. Histological features of the epicarp, mesocarp, testa, embryo, and storage tissues were examined.

2.4 Seed Hydration and Moisture Content
Due to the recalcitrant nature of seeds, its hydration studies were conducted using soaking method. Imbibition potential at regular intervals were recorded. Seed moisture content was determined using fresh seeds from L1 and L2. Wet biomass and dry biomass of seed lots were recorded and moisture content percentage were calculated using standard procedures.

2.5 Phytochemical and Histochemical Analysis
Qualitative phytochemical analysis of seed extracts was carried out using chloroform, methanol, and aqueous extracts following standard phytochemical screening methods for flavonoids, phenols, tannins, saponins, steroids, proteins, carbohydrates, glycosides, coumarins, and alkaloids.
Total phenol content was estimated quantitatively using spectrophotometric methods. Histochemical localization of seed metabolites was performed on hand sections using potassium iodide for starch, Sudan Black B for lipids, ferric chloride for phenols and tannins, picric acid for proteins, sulphuric acid for saponins, and Wagner's reagent for alkaloids.

2.6 Seed Viability Test
Seed viability was assessed using the tetrazolium chloride test. Three replicates of 50 seeds each were incubated in tetrazolium solution, and viable seeds showing characteristic red staining were counted. Viability percentage was calculated accordingly.

2.7 Nursery Experiments
Fresh seeds collected from L1 and L2 were used for germination experiments. Seeds were sown in different sowing media (SM1, SM2, SM3, SM4, and SM5) under nursery conditions. Different pretreatments designated as T1 (control), T2, T3, T4, and T5 were applied before sowing. Seeds were also sown in root trainers (RT) and pots (PT) to study the effect of container type on germination.
For storage experiments, seeds were stored for one month (SS1), two months (SS2), and three months (SS3) in cool and dry conditions before sowing in different media.

2.8 Germination Parameters
Germination parameters such as germination percentage (%), mean germination time (MGT), germination rate per day, coefficient of velocity of germination (CVt) and germination value (GV) were calculated.

2.9 Statistical Analysis
Experimental data were analyzed using two-way analysis of variance (ANOVA) to determine the effects of sowing medium, location, pretreatment, container type, and storage period on germination characteristics. Mean comparisons were performed using Duncan's Multiple Range Test (DMRT) at $p \leq 0.05$. Data were represented as mean ± standard deviation.

3. RESULTS
Garcinia gummi-gutta belongs to the family Clusiaceae. It is commonly known as Malabar tamarind and locally known as Kodampuli, Pinaru in Malayalam, and Kodakapuli in Tamil

3.1. Species description
Garcinia gummi-gutta is a medium-sized evergreen tree 14-18 m tall and 40-53 inch girth with smooth pale grey bark and produces yellow latex (Table1) (Figure 4.6.1.a and b). Leaves dark green, oblanceolate, acute or obtuse, glabrous, entire; 10-12 cm long and 4-6 cm broad; closely arranged nerves. Flowers polygamous, and sessile; the male flower is 5-10 clustered in terminal cyme, yellowish green, 4 unequal sepals and 4 concave petals, stamens numerous, and monadelphous with rudimentary ovary surrounding it; the female flower is solitary, larger than the male flower, with numerous staminodes. Ovary grooved with 6-12 locules. Fruit a berry grooved to the middle. Flowering starts from March to April and about 3-4 months are required for the maturation of fruits from the time of flowering. Fruits collected from 2018 to 2022 from Koratty and Cherai from June to August.

Table 1. Tree measurements and location of Garcinia gummi-gutta

Sl.No.	Girth (inch)	Height (m)	Location	Coordinates
1	45.4	15	Cherai	10° 8' 14.13" N 76° 11' 25.16" E
2	52.8	18	Cherai	10° 8' 14.05" N 76°11' 30.96" E
3	48.2	16.5	Koratty	10° 15' 59.60" N 76° 20' 46.87" E
4	40.2	14	Koratty	10° 15' 43.48" N76° 20' 53.24"E
5	48.6	15.7	Thuravoor	10° 12' 18.72" N 76° 25' 47.82" E

Herbarium specimens were deposited in the herbarium of Western Ghats Forest Research Institute, Peechi with accession number 15712 and at Botanical Survey of India (BSI), Coimbatore and incorporated in the Madras herbarium with account number 202802.

3.2. Data on fruits and seeds

Fruits were washed under running water immediately after collection and fruit rinds were removed. The pulp was removed carefully with the help of a blunt knife without damaging the seed and thoroughly washed under running water.

3.2.1 Fruit and seed morphology

The fruit is a berry and the rind of the fruit is divided into 6-12 segments and 5-9 seeds per fruit. The fruit when matures turns pale yellow. Large-sized fruits weighed 5 numbers per kilogram and small-sized fruit weighed 10-11 numbers per kilogram. 6-8 elongated ovoid brown-colored seeds were found in each fruit. Seeds are covered with white, slimy aril which is sweet to sour. Fruits and seeds collected from L1 and L2 were measured for length, width, and weight (Table 2.). Fruits collected from L1 counted 5 to 8 (6.80 ± 0.94) seeds per fruit and 6 to 9 (7.13 ± 0.99) seeds per fruit from L2. Fruits collected from L1 recorded lengths ranging from 4.9 to 7.5 cm, width of 5.3 to 7.1 cm, and weight of 82 to 257g whereas fruits collected from L2 showed lengths ranging from 5 to 7.3 cm, width of 5.1 to 7.1 cm and weight 80 to 195 g. Dimension of seeds collected from L1 noted length range from 2.2 to 2.9 cm, width 0.9 to 1.2 cm, thickness 0.3 to 0.7 cm and weight 0.64 to 1.03 g and from L2 recorded length 2.1 to 2.6 cm, width 0.8 to 1.2 cm, thickness 0.3 to 0.7 cm and weight 0.62 to 0.98 g.

Table 2. Data on fruits and seeds

Location	Dimensions of	Mean length (cm)	Mean width (cm)	Mean thickness (cm)	Mean weight (g)
L1	Fruit	6.27 ± 0.95	6.19 ± 0.59	-	141.1 ± 50.27
	Seed	2.44 ± 0.18	1.10 ± 0.09	0.487 ± 0.119	0.818 ± 0.12
L2	Fruit	6.19 ± 0.71	5.93 ± 0.58	-	132.2 ± 32.46
	Seed	2.39 ± 0.18	1.07 ± 0.12	0.447 ± 0.119	0.761 ± 0.108

Data were represented as mean ± SD

3.2.2. Fruit and seed anatomy

The anatomy of the fruit rind exhibited a single layer of epicarp. The mesocarp was composed of thin-walled parenchyma cells. A detailed study of the seed coat of Garcinia gummi-gutta revealed three layers with an outer dark brown fibrous layer, a middle brown layer, and an inner yellow-brown layer. Three layers were appressed and appeared as a single layer. The testa of seeds was about 0.2-0.3 mm thick, veiny, and made of thick-walled cells. The seed coat consists of a hydrophobic suberin layer that provides a mechanical barrier for both water uptake and gaseous exchange and may hinder imbibition. Embryo fills the space in the seed and cotyledons were rudimentary. The longitudinal section of the seeds showed an elliptical procambial ring consisting of two to three layers. Seeds were deposited with large amounts of lipids.

3.2.3. Seed Physiology

Seed hydration

Uptake of water at 12-hour intervals was recorded from intact seeds of Garcinia gummi-gutta soaked in water. An uptake of 10.58±0.64 ml water per 100 seeds and an uptake percentage of 13.10 ± 0.31 % was noted after 12 h of soaking and it increased to 21.89 ± 0.74 ml per 100 seeds and 27.13 ± 0.32 % of water uptake after 96 h of soaking. Further, no significant water uptake was noted in soaked seeds (Table 3).

Table 3. Data on mean water uptake rate of the Garcinia

Time	Water uptake (ml/100 seeds)	Water uptake (%)
12 h	10.58 ± 0.64	13.10 ± 0.31
24 h	16.62 ± 0.93	20.59 ± 0.39
36 h	19.28 ± 0.91	23.88 ± 0.28
48 h	20.52 ± 0.90	25.44 ± 0.22
60 h	21.25 ± 0.88	26.34 ± 0.13
72 h	21.65 ± 0.76	26.83 ± 0.17
84 h	21.84 ± 0.75	27.07 ± 0.28
96 h	21.89 ± 0.74	27.13 ± 0.32

Data were represented as mean ± SD, n=4

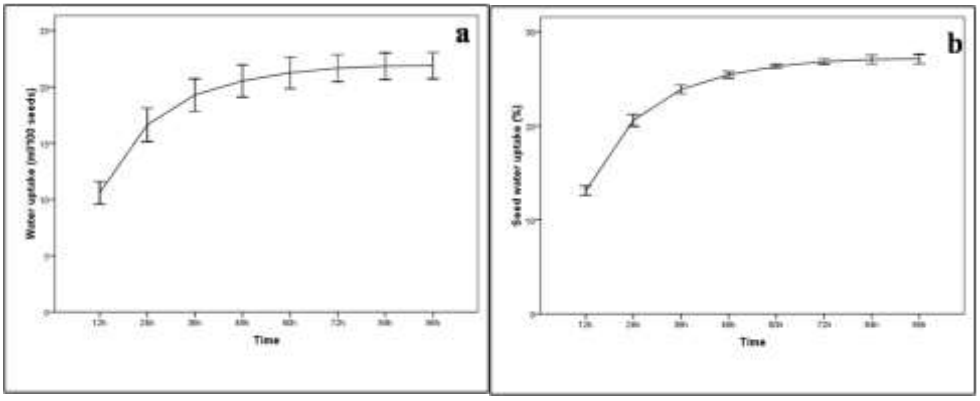

Figure 1 a & b. (a) Mean imbibition of water per 100 seeds and (b) percentage of water absorbed at regular intervals

Moisture content

The percentage of moisture content of 100 seeds each collected from L1 and L2 was calculated based on wet biomass and dry biomass. The mean moisture content percentage of 39.20 ± 1.02 % was recorded in seeds collected from L1 and 39.22 ± 1.07 % in seeds collected from L2 (Table 4).

Table 4.6.4 Mean seed moisture content percentage of Garcinia gummi-gutta

Location	Wet biomass/100 seeds (g)	Dry biomass/100 seeds (g)	Moisture content (%)
L1	81.20 ± 2.26	49.38 ± 1.79	39.20 ± 1.02
L2	77.65 ± 3.41	47.18 ± 1.66	39.22 ± 1.07

Data were represented as mean ± SD, n=4

3.2.4. Phytochemistry – Qualitative test

Seed extracts with chloroform, methanol, and water were subjected to qualitative phytochemical analysis and exhibited the presence of flavonoids, phenol, tannin, saponin, steroid, coumarin, carbohydrates, glycosides, and alkaloids. Methanol extracts of seed showed higher amounts of phenol and steroids. Tannin was not observed in chloroform and water extracts of seeds whereas carbohydrates were not found in methanol and water extract. Proteins were absent in all three extracts of *Garcinia gummi-gutta* seeds (Table 5).

Table 5. Qualitative phytochemical analysis of seed extracts of Hydnocarpus pentandrus

Sl.No	Components	Chloroform	Methanol	Water
1	Flavonoid	+	+	+
2	Phenol	+	+++	+
3	Tannin	-	+	-
4	Saponin	+	+	+

5	Steroids	+	+++	+
6	Protein	-	-	-
7	Coumarin	+	+	+
8	Carbohydrate	+	-	-
9	Glycosides	+	+	+
10	Alkaloids	+	+	+

Quantitative analysis- Total phenol content

Total phenol content was higher in the methanol extract of *Garcinia gummi-gutta* seeds and least in the chloroform extract (Table 6).

Table 6. Quantitative analysis of total phenol content of seeds

Total phenol	Methanol	Chloroform	Water
Seed	2.276 ± 0.008	1.935 ± 0.045	2.063 ± 0.024

Histochemistry:

Seed hand sections of *Garcinia gummi-gutta* were treated with potassium iodide for starch test and no staining was detected. In Sudan black B test lipid droplets were stained blue-black. The ferric chloride test for phenol and tannin test showed positive results. The picric acid test for protein gave no color showing the absence of protein. Seed sections treated with sulphuric acid for saponin test, yellow to orange color were observed. Wagner's test for alkaloids in seed sections stained golden yellow.

Seed viability test

Fresh seeds of *Garcinia gummi-gutta* were subjected to a tetrazolium test to study viability percentage. Three lots of 50 seeds each were tested and a mean viability percentage of 94.67 ± 1.15 % was noted.

Seed processing and storage

Seeds were taken out from the fruit rind, pulp scooped out from the seed, washed thoroughly under running water to remove the pulp, and rubbed with a cloth. Seeds quickly after washing were spread out and dried under shade for 3 days. Mature, healthy seeds were stored in containers in a dry and cool place.

3.3. Nursery practices

Fresh seeds of *Garcinia gummi-gutta* collected from L1 and L2 were sown in SM1, SM2, SM3, SM4, and SM5. Seeds without pretreatment (T1), and pretreated seeds T2, T3, T4, and T5 were sown in different sowing mediums in root trainers. Seeds were sown in different sowing mediums in pots. Stored seeds for SS1, SS2, and SS3 were sown in different sowing mediums in root trainers.

3.3.1. Germination characteristics of seeds sown without pretreaments

Fresh seeds of *Garcinia gummi-gutta* collected from L1 and L2 without pretreatment were sown in different sowing mediums and germination studies were conducted. The highest germination percentage of 77 % was recorded in seeds collected from L1 and sown in SM1 and the lowest germination percentage of 50.25% in seeds collected from L2 and sown in SM5. Minimum mean germination time (MGT) and maximum germination rate per day were noted in seeds collected from both L1 and L2 and sown in SM1. Maximum MGT and minimum germination rate per day were observed in seeds collected from L1 and L2 and sown in SM5. A lesser CV_t value of 9.84% was found in seeds collected from L2 and sown in SM5 and a higher CV_t value of 13.77% in seeds collected from L1 and sown in SM4. Seeds collected from L1 and sown in SM1 recorded a greater germination value (GV) of 0.150 and a lesser of 0.043 in seeds collected from L2 and sown in SM5 (Table 4.6.7).

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Table 7. Germination characteristics of fresh seeds sown without pretreatment

Sowing Medium	Location	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV

SM1	L1	77.00	160.44	0.0062	11.87	0.150
	L2	73.50	162.82	0.0061	11.99	0.128
SM2	L1	69.50	168.94	0.0059	13.53	0.107
	L2	66.75	172.95	0.0058	10.86	0.099
SM3	L1	62.75	177.41	0.0056	12.87	0.081
	L2	64.00	182.41	0.0055	10.84	0.087
SM4	L1	72.00	164.65	0.0060	13.77	0.116
	L2	71.50	165.51	0.0060	11.86	0.118
SM5	L1	54.75	201.44	0.0050	11.02	0.049
	L2	50.25	199.57	0.0050	9.84	0.043

3.3.2. Germination characteristics of pretreated fresh seeds in different sowing medium

Seeds subjected to different pre-treatments T1 (control), T2, T3, T4, and T5 were sown in different sowing mediums. Seeds with seed coat removed and sown in SM1 showed a higher germination percentage of 93.25 % while T1 sown in SM3 exhibited lower germination of 62.75 % in root trainer. MGT was lower in seeds T2 sown in SM1 (101.65 days) and higher MGT of 170 days in seeds T1 sown in SM3. Seeds T2 sown SM1 recorded a greater germination rate of 0.0099 per day and CV_t of 18.97%. A maximum germination value of 0.426 was obtained in seeds with seed coat removed and sown in SM1 while a minimum of 0.082 in seeds without pretreatment and sown in SM3 (Table 8).

Table 8 Germination characteristics of pretreated seeds of Garcinia gummi-gutta

Sowing Medium	Pretreatment	Germination %	MGT($\bar{t}$) (day)	Germination rate(day^{-1})	CV_t (%)	GV
SM1	T1	77.00	160.44	0.0062	11.87	0.151
SM2		69.50	168.94	0.0059	13.53	0.107
SM3		62.75	177.41	0.0056	12.87	0.082
SM4		72.00	164.65	0.0060	13.77	0.116
SM1	T2	93.25	101.65	0.0099	18.97	0.426
SM2		86.25	112.23	0.0089	17.38	0.324
SM3		81.25	120.69	0.0083	17.79	0.251
SM4		90.75	105.19	0.0095	17.81	0.397
SM1	T3	87.00	139.41	0.0072	13.80	0.243
SM2		78.25	150.80	0.0067	11.84	0.176
SM3		71.00	162.01	0.0062	11.09	0.133
SM4		82.25	138.55	0.0072	12.76	0.219
SM1	T4	75.50	120.31	0.0083	14.52	0.232
SM2		68.50	128.30	0.0078	14.24	0.172
SM3		63.25	134.74	0.0074	13.43	0.135
SM4		70.75	123.12	0.0081	14.28	0.202
SM1	T5	90.00	108.14	0.0093	17.83	0.370
SM2		83.50	119.22	0.0084	15.43	0.285
SM3		80.50	125.18	0.0080	15.01	0.249
SM4		86.00	114.21	0.0086	17.02	0.314

3.3.3 Germination characteristics of fresh seeds in different sowing mediums and container

Seeds with the seed coat removed were sown in different sowing mediums in RT and PT. Germination percentage was higher in seeds sown in coir pith in root trainer. Seeds sown in perlite in pots recorded a greater MGT of 139.48 days and the least in seeds sown in coir pith in root trainer. Germination rate and CV_t was higher in all sowing medium in RT

compared to that in PT. Greater germination values were recorded in seeds sown in SM1 in RT and the lowest values in seeds sown in SM3 in PT (Table 9).

Table 9. Germination characteristics of seeds sown in different containers

Sowing Medium	Container	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
SM1	RT	93.25	101.65	0.0099	18.97	0.426
	PT	92.50	120.93	0.0083	15.69	0.320
SM2	RT	86.25	112.23	0.0089	17.38	0.324
	PT	87.25	130.18	0.0077	14.52	0.257
SM3	RT	81.25	120.69	0.0083	17.79	0.251
	PT	84.00	139.48	0.0072	13.77	0.226
SM4	RT	90.75	105.19	0.0095	17.81	0.397
	PT	91.00	129.79	0.0077	15.36	0.292

3.3.4. Germination characteristics of stored seeds in different sowing medium

Seeds stored for 1 month, 2 months, and 3 months were sown in different sowing mediums in root trainers. A higher Germination percentage of 54% was noted in SS1 sown in SM1 and was highly reduced in SS3 sown in all mediums. MGT was found lower and the germination rate was higher in SS1 compared to SS2 and SS3 in all sowing mediums. CV_t reduced from SS1 to SS3. A maximum GV of 0.077 was recorded in SS1 sown in SM1 and a minimum of 0.001 in SS3 sown in SM3 (Table 10).

Table 10 Germination characteristics of stored seeds

Sowing Medium	Seed storage	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
SM1	SS1	54.00	166.67	0.0060	8.15	0.077
SM2		49.00	173.76	0.0058	7.73	0.059
SM3		47.25	177.81	0.0056	7.67	0.053
SM4		51.00	168.58	0.0060	8.11	0.068
SM1	SS2	29.50	180.70	0.0055	8.56	0.019
SM2		25.75	194.46	0.0052	7.42	0.013
SM3		22.75	198.87	0.0050	7.17	0.010
SM4		28.50	185.61	0.0054	7.92	0.018
SM1	SS3	12.00	190.49	0.0053	6.64	0.003
SM2		9.75	200.32	0.0050	6.52	0.002
SM3		7.75	201.65	0.0050	5.90	0.001
SM4		11.00	193.80	0.0052	6.33	0.003

3.3.Statistical analysis of germination characteristics

3.3.1. Analysis of Germination characteristics of fresh seeds in different sowing mediums and locations

A two-way ANOVA was conducted to compare the effect of the location of collection, medium, and their interaction in germination characteristics of fresh seeds without pretreatment. In all germination characteristics, the level of significance was 1% among the sowing medium. But germination characteristics showed no significance among location and interaction between medium and location except in CV_t where significance was at 1% level among location and 5% level in their interaction (Table 11). A greater mean germination percentage of 75.25 % was recorded in seeds sown in SM1 and less in SM5. Seeds sown in SM5 showed a maximum mean germination time of 200.5 days. A significantly higher mean germination rate was obtained both in seeds sown in SM1 and SM4. A maximum mean CV_t of 12.82 % was recorded from seeds sown in SM4 and 12.61 % in seeds collected from L1. Mean GV was significantly higher in seeds sown in SM1 (0.139) and lower (0.046) in seeds sown in SM5. All germination characteristics except the CV_t of seeds collected from different locations were homogenous (Table 12) (Table 13).

Table 11. A two-way ANOVA examining the effect of sowing medium and location of collection of seeds and their interactions on seed germination characteristics

Germination characteristics	Source of Variation	Sum of Squares	Df	Mean Square	F-value	P-value
Germination (%)	Medium	2496.650	4	624.162	46.812	<0.001**
	Location	40.000	1	40.000	3.000	0.094ns
	Medium * Location	43.750	4	10.938	0.820	0.522ns
	Error	400.000	30	13.333		
	Total	178278.000	40			
MGT($\bar{t}$) (day)	Medium	7730.767	4	1932.692	75.135	<0.001**
	Location	43.139	1	43.139	1.677	0.205ns
	Medium * Location	58.896	4	14.724	0.572	0.685ns
	Error	771.691	30	25.723		
	Total	1242194.500	40			
Germination rate(day-1)	Medium	7.277E-6	4	1.819E-6	80.256	<0.001**
	Location	3.600E-8	1	3.600E-8	1.588	0.217ns
	Medium * Location	5.150E-8	4	1.287E-8	0.568	0.688ns
	Error	6.800E-7	30	2.267-8		
	Total	0.001	40			
CVt (%)	Medium	24.547	4	6.164	8.970	<0.001**
	Location	23.547	1	23.547	34.270	<0.001**
	Medium * Location	9.107	4	2.277	3.313	0.023*
	Error	20.613	30	0.687		
	Total	5689.845	40			
Germination value	Medium	0.040	4	0.010	79.549	<0.001**
	Location	<0.001	1	<0.001	2.364	0.135ns
	Medium * Location	0.001	4	<0.001	1.873	0.141ns
	Error	0.004	30	<0.001		
	Total	0.428	40			

**Significant at 1% level; * Significant at 5% level; ns-non significant at 5% level

Table 12. Comparison of germination characteristics of seeds in different mediums

Sowing Medium	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
SM1	75.25a	161.63d	0.0062a	11.93b	0.139a
SM2	68.13b	170.94c	0.0058b	12.20ab	0.103c
SM3	63.38c	179.91b	0.0056c	11.85b	0.084d
SM4	71.75ab	165.08d	0.0061a	12.82a	0.117b
SM5	52.50d	200.50a	0.0050d	10.43c	0.046e

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$

Table 13. Comparison of germination characteristics of seeds from different locations

Location	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
L1	67.20a	174.57a	0.0058a	12.61a	0.101a
L2	65.20a	176.65a	0.0057a	11.08b	0.095a

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$

3.3.2. Analysis Germination characteristics of pretreated fresh seeds in different sowing medium

The impact of pretreatment, sowing medium, and their interactions in germination characteristics was analyzed by two-way ANOVA. All germination characteristics were found significant ata 1% level between sowing medium and pretreatment except in CV_t where among medium significance level was 5%. Interaction between medium and pre-

treatment was significant at a 5 % level in germination rate and germination value and not significant in all other germination characteristics (Table 14). A maximum mean germination percentage of 84.55% was recorded in seeds sown in SM1 among sowing medium and 87.88% in T2 among different pre-treatments. A lower germination percentage was exhibited by T1 and sown in SM3. Minimum MGT was recorded in SM1 among sowing medium and T2 among different pretreatments. Germination rate and CV_t were higher in T2 sown in SM1. A maximum mean GV of 0.350 was recorded in T2 and 0.284 in seeds sown in SM1. The least GV was obtained in T1 and SM3 (Table 15) (Table 16).

Table 14. A two-way ANOVA examining the effect of sowing medium and pretreatments of seeds and their interactions on seed germination characteristics

Germination characteristics	Source of Variation	Sum of Squares	Df	Mean Square	F-value	P-value
Germination (%)	Medium	1745.438	3	581.813	31.742	<0.001**
	Treatment	4470.950	4	1117.738	60.981	<0.001**
	Medium * Treatment	65.750	12	5.479	0.299	0.987ns
	Error	1099.750	60	18.329		
	Total	499891.000	80			
MGT($\bar{t}$) (day)	Medium	3824.233	3	1274.744	84.059	<0.001**
	Treatment	36270.137	4	9067.534	597.931	<0.001**
	Medium * Treatment	235.834	12	19.653	1.296	0.245ns
	Error	909.891	60	15.165		
	Total	1472525.599	80			
Germination rate(day-1)	Medium	1.320E-5	3	4.400E-6	79.815	<0.001**
	Treatment	<0.001	4	2.736E-5	496.336	<0.001**
	Medium* Treatment	1.600E-6	12	1.333E-7	2.419	0.012*
	Error	3.307E-6	60	5.512E-8		
	Total	0.005	80			
CVt (%)	Medium	22.920	3	7.640	3.422	0.023*
	Treatment	351.521	4	87.880	39.366	<0.001**
	Medium * Treatment	32.102	12	2.675	1.198	0.305ns
	Error	133.942	60	2.232		
	Total	17972.340	80			
Germination value	Medium	0.145	3	0.048	69.750	<0.001**
	Treatment	0.589	4	0.147	212.568	<0.001**
	Medium * Treatment	0.019	12	0.002	2.305	0.017*
	Error	0.042	60	0.001		
	Total	4.996	80			

**Significant at 1% level; * Significant at 5% level; ns-non significant at 5% level

Table 15. Comparison of germination characteristics of seeds in different medium

Sowing Medium	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
SM1	84.55a	125.99d	0.0082a	15.40a	0.284a
SM2	77.20c	135.89b	0.0075c	14.48ab	0.213c
SM3	71.75d	144.00a	0.0071d	14.04b	0.170d
SM4	80.35b	129.14c	0.0079b	15.13a	0.250b

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$

Table 16. Comparison of germination characteristics of different pretreated seeds

Treatment	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
T1	70.31c	167.86a	0.0060e	13.01d	0.114d
T2	87.88a	109.94e	0.0092a	17.98a	0.350a
T3	79.63b	147.69b	0.0068d	12.38d	0.193c
T4	69.50c	126.62c	0.0079c	14.11c	0.185c
T5	85.00	116.69d	0.0086b	16.32b	0.305b

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$

3.3.3. Analysis of germination characteristics of fresh seeds in different sowing medium and container

The effect of container volume, sowing medium, and their interaction in germination characteristics of seeds was examined by two-way ANOVA.

Table 17. A two-way ANOVA examining the effect of sowing medium and volume of containers and their interactions on seed germination characteristics

Germination characteristics	Source of Variation	Sum of Squares	Df	Mean Square	F-value	P-value
Germination (%)	Medium	497.344	3	165.781	10.505	<0.001**
	Container	5.281	1	5.281	0.335	0.568ns
	Medium * Container	13.094	3	4.365	0.277	0.842ns
	Error	378.750	24	15.781		
	Total	250289.000	32			
MGT($\bar{t}$) (day)	Medium	1483.330	3	494.443	38.048	<0.001**
	Container	3251.002	1	3251.002	250.166	<0.001**
	Medium * Container	54.515	3	18.172	1.398	0.268ns
	Error	311.889	24	12.995		
	Total	466015.943	32			
Germination rate(day-1)	Medium	7.636E-6	3	2.545E-6	38.120	<0.001**
	Container	1.639E-5	1	1.639E-5	245.434	<0.001**
	Medium* Container	5.859E-7	3	1.953E-7	2.925	0.054ns
	Error	1.602E-6	24	6.677E-8		
	Total	0.002	32			
CVt (%)	Medium	11.905	3	3.968	2.206	0.113ns
	Container	79.475	1	79.475	44.178	<0.001**
	Medium * Container	2.725	3	0.908	0.505	0.683ns
	Error	43.176	24	1.799		
	Total	8752.858	32			
Germination value	Medium	0.085	3	0.028	34.725	<0.001**
	Container	0.046	1	0.046	56.117	<0.001**
	Medium * Container	0.009	3	0.003	3.577	0.029*
	Error	0.020	24	0.001		
	Total	3.270	32			

**Significant at 1% level; * Significant at 5% level; ns-non significant at 5% level

All germination characteristics appeared significant at 1% level between the sowing medium and between container volumes except in CV_t among medium and germination percentage among container volume where there was no significance. Interaction between container volume and sowing medium was found significant at a 5% level in germination value and no significant effect in all other germination characteristics (Table 17). Among the sowing medium higher mean germination percentage of 92.88% was recorded in seeds sown in SM1 and it was homogenous among containers. Mean MGT was found minimum and mean germination rate maximum in seeds sown in SM1 in root trainers. Greater mean CV_t was recorded in root trainers and homogenous among medium. A higher mean GV of 0.373 was noted in SM1 among different sowing mediums and of 0.350 in RT among containers (Table 18) (Table 19).

Table 18. Comparison of germination characteristics of seeds in different mediums

Sowing Medium	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
SM1	92.88a	111.29c	0.0091a	17.33a	0.373a
SM2	86.75b	121.20b	0.0083c	15.95ab	0.291b

SM3	82.63c	130.09a	0.0077d	15.78b	0.239c
SM4	90.88a	117.49b	0.0086b	16.58ab	0.345a

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$

Table 19. Comparison of germination characteristics of seeds from different container

Container	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
RT	87.88a	109.94b	0.0092a	17.98a	0.350a
PT	88.69a	130.09a	0.0077b	14.83b	0.274b

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$

3.3.4. Analysis of germination characteristics of stored seeds in different sowing medium

Germination characteristics of SS1, SS2, and SS3 in different sowing mediums were analyzed. The significance level among the sowing medium and stored period was 1% in all germination characteristics except in CV_t where it was 5% significant among medium. Interaction between medium and container volume showed a 1% level significance in germination value and no significance in all other germination characteristics (Table 20). A higher mean germination percentage of 50.31% was exhibited by SS1 and a lower mean germination percentage of 10.13 % in SS3.

Table 20. A two-way ANOVA examining the effect of sowing medium and storage period and their interactions on seed germination characteristics of stored seeds

Germination characteristics	Source of Variation	Sum of Squares	Df	Mean Square	F-value	P-value
Germination (%)	Medium	235.063	3	78.354	14.559	<0.001**
	Storage period	13058.042	2	6529.021	1213.134	<0.001**
	Medium * Storage period	16.125	6	2.688	0.499	0.805ns
	Error	193.750	36	5.382		
	Total	53929.000	48			
MGT($\bar{t}$)(day)	Medium	1373.789	3	457.930	33.786	<0.001**
	Storage period	5300.338	2	2650.169	195.532	<0.001**
	Medium * Storage period	88.047	6	14.674	1.083	0.391ns
	Error	487.931	36	13.554		
	Total	1668899.868	48			
Germination rate(day-1)	Medium	1.238E-6	3	4.128E-7	33.534	<0.001**
	Storage period	4.501E-6	2	2.251E-6	182.827	<0.001**
	Medium * Storage period	7.666E-8	6	1.278E-8	1.038	0.417ns
	Error	4.432E-7	36	1.231E-8		
	Total	0.001	48			
CVt (%)	Medium	4.882	3	1.627	3.114	0.038*
	Storage period	24.043	2	12.021	23.006	<0.001**
	Medium * Storage period	1.672	6	0.279	0.533	0.779ns
	Error	18.811	36	0.523		
	Total	2637.052	48			
Germination value	Medium	0.001	3	<0.001	13.477	<0.001**
	Storage period	0.034	2	0.017	778.617	<0.001**
	Medium * Storage period	0.001	6	9.443E-5	4.285	0.002**
	Error	0.001	36	2.204E-5		
	Total	0.072	48			

**Significant at 1% level; * Significant at 5% level; ns-non significant at 5% level

Table 21. Comparison of germination characteristics of stored seeds in different medium

Sowing Medium	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
SM1	31.83a	179.28d	0.0056a	7.78a	0.0328a
SM2	28.17b	189.51b	0.0053c	7.22ab	0.0247b
SM3	25.92c	192.78a	0.0052d	6.91b	0.0216b
SM4	30.17a	182.66c	0.0055b	7.45ab	0.0295a

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$

Table 22. Comparison of germination characteristics of stored seeds in different stored periods

Sowing Medium	Germination %	MGT($\bar{t}$) (day)	Germination rate(day-1)	CVt (%)	GV
SS1	50.31a	171.70c	0.0058a	7.91a	0.0642
SS2	26.63b	189.91b	0.0053b	7.77a	0.0149b
SS3	10.13c	196.56a	0.0051c	6.35b	0.0023c

Mean with the same superscript letter indicates no significant difference in Duncan's multiple rangetest at $p \leq 0.05$
Minimum mean MGT and maximum mean germination rate and CV_t were found in SS1. The lowest mean germination value of 0.0023 was recorded in SS3 (Table 21) (Table 22).

4. DISCUSSION

The present study provides detailed information on the taxonomy, seed biology, physiology, and germination behaviour of *Garcinia gummi-gutta* collected from different regions of Western Ghats. The species is an important indigenous multipurpose tree of the Western Ghats and exhibits considerable ecological and medicinal significance (Pramanik, et. al., 2021; Warrier, et. al., 2020; Karthik, Ramana, & Rathod, 2024; Deepthy, Vishnu, & Siril, 2026). The morphological characteristics observed in the present investigation were consistent with earlier descriptions reported for the species, including evergreen habit, yellow latex production, polygamous flowers, and segmented berry fruits containing multiple seeds. Minor variations in fruit size, seed number, and seed dimensions between locations may be attributed to environmental conditions, genetic variability, and local edaphic factors (Scaria et. al., 2025; Gurudev Singh, 2025; Shaju, & Shaji, 2025). The fruits produced 5–9 seeds per fruit with appreciable seed biomass and high moisture content. Seeds possessed a thick multilayered seed coat with hydrophobic properties and abundant lipid reserves (Kaprakkaden, & Ali, 2025). Histochemical analysis confirmed the predominance of lipids and phenolic compounds and the absence of starch and proteins. Such biochemical composition suggests that the seeds rely mainly on lipid metabolism during germination (Patil, Muhammed, & Anu-Appaiah, 2016; Anilkumar, et. al., 2023). The high seed moisture content (~39%) together with the rapid decline in viability during storage indicates that the seeds exhibit recalcitrant behaviour (Warrier, et. al., 2020). Recalcitrant seeds are generally sensitive to desiccation and cannot withstand prolonged storage (Pammenter, & Berjak, 2014), which explains the progressive reduction in germination percentage from SS1 to SS3.

Water uptake studies demonstrated gradual imbibition up to 96 h, after which no significant increase was observed. The relatively slow imbibition pattern may be associated with the compact testa and suberin-rich seed coat, which acts as a barrier to water penetration and gaseous exchange. This structural characteristic likely contributes to delayed germination and prolonged mean germination time observed in untreated seeds. Tetrazolium testing revealed high viability (94.67%), indicating that low germination in some treatments was not due to embryo mortality but rather dormancy-associated barriers imposed by the seed coat.

Fresh seeds showed better germination performance than stored seeds across all sowing media, emphasizing the importance of immediate sowing after extraction (Jaganathan, et. al., 2024). Germination percentages of untreated fresh seeds ranged from 50.25% to 77%, with significant variation among sowing media. Among the media tested, SM1 consistently recorded the highest germination percentage, germination rate, and germination value with reduced mean germination time. The superior performance of SM1 may be related to better aeration, moisture retention, and root penetration properties that favour embryo growth and seedling establishment. In contrast, SM5 produced poor germination performance and prolonged germination duration, suggesting that it was less suitable for maintaining the moisture and aeration balance required for successful germination.

Pretreatment significantly improved germination characteristics. Removal of the seed coat (T2) produced the highest germination percentage (93.25%), shortest mean germination time, highest germination rate, and greatest germination value. These findings clearly indicate that the seed coat acts as a major mechanical barrier restricting germination. Removal of the testa probably facilitated rapid imbibition and enhanced oxygen diffusion to the embryo, thereby accelerating metabolic activation and radicle emergence. Treatments T5 and T3 also improved germination compared to untreated seeds, although their effects were less pronounced than complete seed coat removal. The significant improvement obtained through pretreatment demonstrates that dormancy in *Garcinia gummi-gutta* is primarily associated

with seed coat-imposed constraints rather than physiological embryo dormancy (Bohra, Waman, & Devi, 2021). Container type also influenced germination behaviour. Root trainers generally exhibited lower mean germination time, higher germination rate, and higher germination value compared to pots. The improved performance in root trainers may be due to enhanced drainage, better root zone aeration, and reduced root disturbance. Although germination percentages between containers were statistically homogeneous, seedling vigour and speed of germination were superior in root trainers, indicating their suitability for nursery production of the species.

Storage studies revealed a sharp decline in germination percentage and germination value with increasing storage duration. Seeds stored for one month retained moderate viability, whereas seeds stored for three months showed extremely poor germination. Increased mean germination time and reduced coefficient of velocity of germination in stored seeds indicate deterioration in physiological activity during storage. The sensitivity of the seeds to storage further supports their recalcitrant nature. Since recalcitrant seeds are unable to tolerate dehydration and low-temperature storage, rapid loss of viability is expected under conventional storage conditions.

Statistical analyses through two-way ANOVA demonstrated that sowing medium and pretreatment significantly influenced all germination parameters. Interaction effects between treatments and media were significant for germination rate and germination value, suggesting that pretreatment efficiency varied depending on the medium used. In contrast, location of seed collection had no significant effect on most germination characteristics, indicating that the species maintains relatively stable germination behaviour across the studied regions. Significant differences in coefficient of velocity of germination among locations may be due to microenvironmental influences affecting seed maturation.

Overall, the present study demonstrates that successful propagation of *Garcinia gummi-gutta* depends greatly on seed freshness, removal of the seed coat, appropriate sowing medium, and suitable nursery containers (Nair, Mohanan, & Mathew, 2005). Preliminary observations on seed and plantation technology for Garcinia gummi-gutta (Guttiferae): an indigenous and economic tree crop of the Indian Peninsula. *Forests, Trees and Livelihoods*, *15*(1), 75-87.. The findings contribute valuable information for large-scale propagation, conservation, and domestication of this economically and medicinally important species.

5. CONCLUSION

The present investigation provides comprehensive information on the taxonomy, seed biology, physiology, and germination behaviour of *Garcinia gummi-gutta*. The species produces recalcitrant seeds characterized by high moisture content, lipid-rich tissues, and rapid loss of viability during storage. Fresh seeds exhibited high viability and superior germination compared to stored seeds, confirming the necessity for immediate sowing after seed extraction.

Among the sowing media tested, SM1 proved to be the most effective for germination and seedling establishment, producing higher germination percentage, germination rate, and germination value with reduced mean germination time. Pretreatment by removal of the seed coat significantly enhanced germination performance, indicating that the testa acts as a mechanical barrier to water uptake and embryo emergence. Root trainers were more suitable than pots for raising seedlings due to improved germination dynamics and seedling vigour.

Storage adversely affected all germination parameters, with substantial decline in germination after three months of storage. Therefore, long-term storage under conventional conditions is unsuitable for the species. The study highlights that successful nursery production of *Garcinia gummi-gutta* can be achieved by using freshly extracted seeds, removing the seed coat before sowing, and utilizing suitable sowing media and root trainer containers.

The findings of the present study provide important baseline information for propagation protocols, nursery management, conservation programmes, and sustainable utilization of *Garcinia gummi-gutta*. Further studies on biochemical changes during storage, cryopreservation techniques, and field establishment of seedlings may support long-term conservation and commercial cultivation of this valuable species.

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