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VARIABILITY STUDIES FOR SEED AND SEEDLING TRAITS IN *CALOPHYLLUM INOPHYLLUM* (L.)AT SOUTH INDIA

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ABSTRACT

The present investigation was carried out at Forest College and Research Institute, Mettupalayam, Tamil nadu to identify the best half sibs of *Calophyllum inophyllum* across its natural distribution of South India for further collection of seeds for afforestation or breeding purpose. The seeds were collected from different climatic zones of South India. Seeds were measured for its length, width, Pod length, Width and then sown in nursery to study the variation in germination and initial growth parameters *viz.,* germination percent, germination value, peak value, mean daily germination of seedlings. Seeds collected from Western Ghats of Karnataka were superior compared to seeds from other parts in all the traits considered for the study. These seed sources can be further screened for tree improvement traits considering their immense value in yielding bio diesel.

Keywords: *Calophyllum inophyllum,* seed source, seed parameters, variability, Germination percent, Germination value.

INTRODUCTION

Calophyllum inophyllum (Poon tree) is a multipurpose tree belonging to the family Clusiaceae, commonly known as mangosteen family. This plant has multiple origins including East Africa, India, South East Asia, Australia and the South Pacific. *Calophyllum inophyllum* is known by various names around the world. This tree is widely available in India, South East Asia and Australia. It grows in areas with an annual rain of 1000–5000 mm at altitudes from 0 to 200 m. *Calophyllum inophyllum* is allow-branching and slow-growing tree with two distinct flowering periods of late spring and late autumn. But sometimes its flowering may occur throughout the year.

Calophyllum inophyllum grows best in sandy, well drained soils. However it tolerates clays, calcareous, and rocky soils. The tree supports a dense canopy of glossy, elliptical, shiny and tough leaves, fragrant white flowers, and large round nuts. Its size typically ranges between 8 and 20 m (25 – 65 ft) tall at maturity, sometimes reaching upto 35 m (115 ft). The growth rate of the tree is 1m (3.3 ft) in height per year on good sites. Its leaves are heavy and glossy, 10 – 20 cm (4 – 8 inch) long and 6 – 9 cm (2.4 – 3.6 inch) wide, light green when young and dark green when older. Fruits are spherical drupes and arranged in clusters. The fruit is reported to be pinkish-green at first. However, it turns later to be bright green and when ripe, it turns dark grey- brown and wrinkled. The tree yield is 100 – 200 fruits/kg. In each fruit, one large brown seed 2 – 4 cm (0.8 – 1.6 inch) in diameter is found. The trees yield 3000 – 10,000 seeds/tree/season. The seed is surrounded by a shell and a thin layer of pulp of 3 – 5 mm. *Calophyllum inophyllum* oil is non-edible and dark green. Traditionally, its oil has been used as a medicine, soap, lamp oil, hair grease and cosmetic in different parts of the world. Recently, *Calophyllum inophyllum* has been proposed as a source of biodiesel.

MATERIAL AND METHODS

The present study was under taken during the year 2014-15 at Forest College and Research Institute, Mettupalayam, situated at 11°19'N latitude and 77°56'E longitude and an altitude of 350 m above MSL. The average annual rainfall is 945 mm, most of which is received

between June to September. The temperature varies from 15 to 34.9^{0}C. The extensive survey was under taken across three different states and one union territory of India. A distance of at least 200 m was maintained between two trees and at least 25 kms between two seed sources. The individual tree was identified based on their phenotypical characteristics and the individual tree identity was also maintained. Seeds were extracted from 100 pods after sun drying for ten days for assessment of seed characteristics. Further same seeds were used for assessing germination and seedling characters. Seed parameters such as seed length, seed width, seed thickness, seed volume and 100 seed weight were recorded for each seed source.

The experiment was laid out in completely randomized design with five replications of 100 seeds each. The seeds were sown in the standard nursery bed and regular watering was done. Observations on daily germination were recorded up to 31 days from date of sowing. Germination percentage, peak value, mean daily germination, germination rate and germination value were recorded for each seed source. Data collected was analysed statistically using Mstatc program.

$$\text{Germination per cent} = \frac{\text{Number of seeds germinated}}{\text{Number of seeds sown}} \times 100$$

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

Where, PV- Peak Value of germination.
MDG- Mean Daily Germination

$$\text{Peak Value} = \frac{\text{Total germination per cent}}{\text{Total number of days}}$$

$$\text{Mean Daily Germination} = \frac{\text{Final germination per cent}}{\text{The number of days that took to reach Peak Germination}}$$

Estimation of oil content using Soxhlet method

For estimating oil, the seeds were depulped the kernels dried at 50° C for 16 hrs and allowed to cool in a desiccator. Five grams of seeds were pulverized to a fine powder in a porcelain mortar. Ground samples were placed in a filter paper and fastened in such a way to prevent escape of the meal and then carefully transferred to an extraction thimble. The thimble was then placed in a Soxhlet extractor to which sufficient quantity of solvent petroleum ether (40 - 60° C) was added and heated until eleven siphonings were completed. The oil content was recorded by evaporating the petroleum ether at 60° C. The entire extraction process was carried out in Soxhlet extractor according to AOAC (1970). The percentage of oil content was then calculated by using the formula.

$$\text{Oil per cent} = \frac{\text{Oil weight (g)}}{\text{Sample weight (g)}} \times 100$$

Table.1 *Calophyllum inophyllum* seed source collection from different places in South India

Location	Name of the progenies	State	Latitude	Longitude	Altitude (ft)
Vedaranyam	FCRICI 1	Tamilnadu	10°22'N	79°51'E	14
Nagapattinam	FCRICI 2	Tamilnadu	10°45'N	79°49'E	5
Velankanni	FCRICI 3	Tamilnadu	10°41'N	79°50'E	21
Thiruvarur	FCRICI 4	Tamilnadu	10°45'N	79°37'E	38
Pudhucherry	FCRICI 5	Tamilnadu	11°54'N	79°47'E	25
Tindivanam	FCRICI 6	Tamilnadu	12°13'N	79°39'E	140
Cuddalore	FCRICI 7	Tamilnadu	11°44'N	79°42'E	35
Nagercoil	FCRICI 8	Tamilnadu	08°09'N	77°22'E	148
Pechiparai	FCRICI 9	Tamilnadu	08°26'N	77°18'E	326
Mettupalayam	FCRICI10	Tamilnadu	11°19'N	76°58'E	1036
Thiruchencode	FCRICI 11	Tamilnadu	11°22'N	77°53'E	1405
Coimbatore I	FCRICI 12	Tamilnadu	10°59'N	76°54'E	1307
Coimbatore II	FCRICI 13	Tamilnadu	10°57'N	76°55'E	1373
Honnavara	FCRICI 14	Karnataka	14°15'N	74°26'E	43
Hubli	FCRICI 15	Karnataka	15°22'N	75°04'E	2127
Sirsi	FCRICI 16	Karnataka	14°39'N	74°52'E	2049
Bhatkal	FCRICI 17	Karnataka	13°59'N	74°31'E	51
Kumta	FCRICI 18	Karnataka	14°26'N	74°23'E	114
Udupi	FCRICI 19	Karnataka	13°20'N	74°43'E	49
Mangalore	FCRICI 20	Karnataka	12°54'N	74°51'E	79
Talugoppa	FCRICI 21	Karnataka	14°12'N	74°54'E	1961
Shimoga	FCRICI 22	Karnataka	13°53'N	75°33'E	1922
Sagar	FCRICI 23	Karnataka	14°09'N	75°00'E	1982
Tumkur	FCRICI 24	Karnataka	13°20'N	76°09'E	2974
Mandiya	FCRICI 25	Karnataka	12°29'N	76°54'E	2210
Mysore	FCRICI 26	Karnataka	12°22'N	76°40'E	2299
Tiruvandrum	FCRICI 27	Kerala	8°29'N	76°59'E	81
Thirissur	FCRICI 28	Kerala	10°29'N	76°17'E	161
Kottayam	FCRICI 29	Kerala	9°33'N	76°32'E	21
Upala	FCRICI 30	Kerala	12°41'N	79°54'E	35

RESULTS AND DISCUSSION

Data from the Table. 2 revealed that seed traits for all seed sources showed significant differences. The seeds collected from Honnavara region were longest, thickest and had higher mass as well as seed volume compared to all other seed sources. Seed length amongst various seed sources varied from 1.77 cm to 2.71 cm, seed width from 1.24 cm to 2.35 cm.

Table 2. Seed characteristics as influenced by various place in South India

Name of the progenies	Pod length (cm)	Pod width (cm)	Seed length (cm)	Seed width (cm)	100 Seed weight (g)
FCRICI 1	2.91	2.23	1.95	1.87	485.52
FCRICI 2	3.43*	2.51	2.13	2.06**	499.51
FCRICI 3	3.06	2.69	1.91	1.75	442.44
FCRICI 4	2.49	2.56	2.13	2.03**	480.40
FCRICI 5	3.02	2.40	2.19	1.96*	485.46
FCRICI 6	2.98	2.56	2.09	1.47	490.19
FCRICI 7	2.97	2.45	2.13	1.51	482.31
FCRICI 8	2.93	2.50	2.08	1.38	434.29
FCRICI 9	3.37*	2.33	2.04	1.79	439.44
FCRICI10	2.95	2.20	2.15	1.49	504.46*
FCRICI 11	2.43	2.43	2.00	1.45	476.58
FCRICI 12	2.89	2.38	2.03	1.32	485.31
FCRICI 13	2.75	2.59	2.00	1.42	472.51
FCRICI 14	3.95**	3.42**	2.71**	2.35**	536.26*
FCRICI 15	2.50	2.56	1.87	1.33	474.26
FCRICI 16	2.65	2.40	1.89	1.43	500.32
FCRICI 17	2.86	2.40	1.77	1.24	470.27
FCRICI 18	3.07	2.52	2.19	1.34	495.48
FCRICI 19	2.83	2.31	2.17	1.50	506.27*
FCRICI 20	2.62	2.91*	2.14	1.28	480.34
FCRICI 21	2.76	2.50	2.00	1.28	487.52
FCRICI 22	2.84	2.40	2.09	1.41	444.19
FCRICI 23	3.46*	2.30	2.13	1.28	501.18*
FCRICI 24	3.33*	2.83*	2.22	2.14**	495.62
FCRICI 25	2.99	2.68	2.52*	2.19**	450.38
FCRICI 26	2.77	2.70	2.31	1.50	510.35**
FCRICI 27	2.69	2.56	2.16	1.44	469.57
FCRICI 28	3.41*	2.46	2.07	1.48	468.37
FCRICI 29	2.90	2.57	1.88	1.42	522.21**
FCRICI 30	2.53	2.58	2.21	1.55	523.42**
Mean	**2.94**	**2.53**	**2.11**	**1.59**	**483.81**
SEd	0.19	0.15	0.15	0.16	8.67
CD (0.05)	0.39	0.30	0.31	0.32	17.35
CD (0.01)	0.52	0.40	0.41	0.43	23.08

Pod length and pod width varied from 2.50 cm to 3.95 cm and 2.20 cm to 3.42 cm respectively. The 100 seed weight ranged between 434.29 g to 536.26 g. These variations may be due to the fact that, this species grows over a wide range of climatic conditions as well as soil types and altitudes. Similar findings were revealed by Sudhir Kumar (2003) in *Jatropha curcas* and Vasanth Reddy *et al.* (2007) in *Pongamia pinnata*.

Analysis of variance (ANOVA) revealed that the results were statistically significant for all the germination attributes (Table. 3). Overall germination per cent was on/or above the 50 per cent. Maximum germination per cent was found in Honnavara region (78.00 %), owing to higher germination value (5.11) and peak value of germination (3.06). It was followed by Nagercoil region (63.33 %) and Nagapattinam region (63.00 %) seed sources. Whereas, minimum germination per cent was recorded on MAngalore region (41.67 %). The size and shape of seeds is variable depending on the structure and form of the ovary and environmental conditions under which plant is growing. It is evident from the result that seeds from Honnavara region was found to be superior with respect to germination percentage. This is in line with study made by Dwivedi (1993) in Azadirachta indica and Devagiri et al. (1998) in Dalbergia sissoo.

Table 3. Seed germination attributes in different place in South India

Name of the progenies	Seed germination attributes				
	Germination percent	Germination value	Peak value	Mean daily germination	Oil %
FCRICI 1	58.00	4.41	2.30	1.92	44.90
FCRICI 2	63.00	4.71*	2.85*	1.69	52.33**
FCRICI 3	56.67	3.68	1.92	1.93	47.77
FCRICI 4	51.00	3.87	2.18	1.79	42.60
FCRICI 5	53.00	3.32	2.09	1.59	49.63**
FCRICI 6	53.00	3.32	1.98	1.67	41.00
FCRICI 7	54.33	3.07	1.93	1.59	38.23
FCRICI 8	63.33	3.80	2.17	1.74	46.37
FCRICI 9	59.00	4.45	2.55	1.56	51.43**
FCRICI10	54.67	3.10	1.97	1.57	45.83
FCRICI 11	49.00	3.64	2.06	1.77	44.40
FCRICI 12	59.67	3.50	2.04	1.70	43.67
FCRICI 13	53.33	3.48	2.20	1.57	50.57**
FCRICI 14	73.00**	5.11**	3.06**	1.79	55.60**
FCRICI 15	51.00	3.64	2.31	1.56	39.30
FCRICI 16	48.00	3.63	1.99	1.83	44.77
FCRICI 17	49.67	3.25	1.92	1.68	51.43**
FCRICI 18	54.00	3.48	2.02	1.73	45.50
FCRICI 19	50.67	2.88	1.98	1.43	43.53
FCRICI 20	41.67	2.54	1.77	1.44	43.00
FCRICI 21	49.67	2.53	1.68	1.51	43.63
FCRICI 22	47.33	3.26	1.89	1.72	46.33
FCRICI 23	51.00	3.62	1.98	1.82	48.17
FCRICI 24	51.00	2.63	1.68	1.73	37.86
FCRICI 25	47.00	2.96	1.87	1.66	48.33
FCRICI 26	50.67	2.90	2.09	1.40	35.67
FCRICI 27	52.33	2.24	1.70	1.33	47.70
FCRICI 28	49.67	2.72	1.98	1.52	44.45
FCRICI 29	52.33	2.63	1.74	1.53	44.70
FCRICI 30	49.33	3.29	2.06	1.58	46.30
Mean	**53.21**	**3.39**	**2.06**	**1.64**	**45.50**
	6.54	0.60	0.31	0.40	1.46
	13.0	1.20	0.62	NS	2.93
	17.4	1.59	0.82	NS	3.89

They found that the variation observed in the seed characters may be attributed to adverse environment and differences in their distribution range this in turn affect the germination of seeds. Seedlings of Honnavara region higher oil content (55.60 %). It was followed by Nagapattinam (52.33 %) and Pechiparai region (51.43 %).

Correlation studies

Genotypic and Phenotypic correlation

Pod length (1.805), Pod width (0.498), Seed length (0.562), seed width (1.399)Seed weight (-0.394), Germination percent (3.099), Germination value (2.693), Peak value (2.712) and Mean daily germination (1.853) showed positive significant correlation with oil content (Tables 4).

Table 4. Genotypic (G) and phenotypic (P) correlation of seed and seedling attributes of *Calophyllum inophyllum* in South India

Characters		Pod length	Pod width	Seed length	Seed width	100 Seed weight	Germination percent	Germination value	Peak value	Mean daily germination	Oil %
Pod length	**(G)**	1.000	0.396*	0.566**	0.547**	0.148	1.181**	0.663**	0.803**	0.533**	1.805**
	(P)	1.000	0.230	0.343*	0.406*	0.130	0.300	0.198	0.236	0.074**	0.253*
Pod width	**(G)**		1.000	0.822**	0.525**	0.323*	0.630**	0.196	0.450**	0.067	0.498*
	(P)		1.000	0.389*	0.326**	0.186	0.065	0.065	0.165	-0.053	0.062
Seed length	**(G)**			1.000	0.753**	0.363*	0.669**	0.283	0.576**	-0.124	0.562*
	(P)			1.000	0.438**	0.248	0.085	0.005	0.137	-0.090	0.099
Seed width	**(G)**				1.000	0.110	0.778**	0.557**	0.704**	0.498**	1.399**
	(P)				1.000	0.084	0.232	0.375*	0.342*	0.229	0.098
100 Seed weight	**(G)**					1.000	0.205	0.061	0.246	-0.147	-0.394
	(P)					1.000	0.070	-0.015	0.104	-0.117	-0.059
Germination percent	**(G)**						1.000	1.127**	1.301**	0.238	3.099**
	(P)						1.000	0.542**	0.523**	0.353*	0.143
Germination value	**(G)**							1.000	0.991**	0.607**	2.693**
	(P)							1.000	0.848**	0.638**	0.200*
Peak value	**(G)**								1.000	0.346*	2.712 **
	(P)								1.000	0.232	0.208*
Mean daily germination	**(G)**									1.000	1.853**
	(P)									1.000	0.037
Oil %	**(G)**										1.000
	(P)										1.000

(** Significant at 1% level * Significant at 5% level)

Pod length (0.253), Germination value (0.200), Peak value (0.208) showed positive but significant correlation with oil content. Pod width (0.062), seed length (0.099), seed width (0.098), Seed weight (-0.059), Germination percent (0.143), Mean daily germination (0.037), showed positive but non-significant correlation with oil content (Tables 5 and 6). A highly significant and positive correlation existed between Pod length (0.253), Germination value (0.200),Peak value (0.208). Significant correlation among various seed germination and seedling traits suggests that test weight may prove to be important criteria in selection of geographic seed sources for raising stock for bulk commercial plantations.

This study identifies two best sources for *Calophyllum inophyllum*L based upon seed and seedling traits for those places of Honnavara and Nagergoil which were sampled. On a short term basis, breeding zones may be set up in these environmentally homogeneous areas. However, this may be preliminary as only seedling traits have been considered. Hence, seed source screening provides a great opportunity to the tree breeder to screen and capture natural variation for success of afforestation, besides providing information on the raw material for breeding and evolving improved planting stock within a seed source.

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