

Effect of Seed Source on Germination and Early Growth in *Madhuca latifolia* in Odisha

SASWAT NAYAK¹ AND UTTAM KUMAR SAHOO^{2*}

Department of Forestry, School of Earth Sciences and Natural Resource Management, Mizoram University, Aizawl 796 004, Mizoram, India

¹Present address: College of Forestry, Orissa University of Agriculture and Technology, Bhubaneswar 751 001, India

E-mail: ¹S_nayak3@rediffmail.com; ²uttams64@gmail.com

*Author for correspondence

ABSTRACT

We evaluated the effect of seed source on seed germination and early seedling characters of *Madhuca latifolia* in Odisha. The results displayed significant variations ($P < 0.05$) in all germination and growth characters among provenances. Germination per cent ranged from 53 (P10) to 93.8 (P8) with variation in phenotype ($PCV = 22.7$) to be more than variation in genotype ($GCV = 12.48$). Environment effect was more pronounced than genotype in all germination character except mean daily germination. Collar diameter ($h^2 = 50\%$) was found to be more heritable than seedling length and biomass. The best provenance was found to be Nuapada (P8) for germination and growth characters.

Key Words: Mahua; Seed Traits; Phenotypic Variation; Genotypic Variation; Heritability

INTRODUCTION

Madhuca latifolia J.F. Macb. (syn. *M. indica* J.F. Gmel.; *Bassica latifolia* Roxb.) of family Sapotaceae, commonly known as 'Mahua', is native to India and found in central and north India, and subtropical region of Indo-Pak subcontinent (Ramadan et al. 2016). This species is a major source of natural fat (mahua butter) extracted from its seeds (Kulkarni et al. 2013, Arora and Kumar 2015). The fruits and flowers of this species are of multipurpose use (Nayak and Sahoo 2020a). The tree wins fame due to liquor distilled from the flowers, which are used to make vinegar. The predominance of mono unsaturated fatty acids in the seed oil help in reducing blood cholesterol and reducing incidence of heart diseases (Lawson 1995), besides the seed oil is used for soap, lubricating grease, fatty alcohols and candles making and has gained the importance as bio-diesel and is emerging as a viable alternative to fossil fuel (Lakshmikanth et al. 2013). The species shows enormous genetic diversity (Hedge et al. 2018). Therefore understanding the geographical distribution of morphometric relevant genetic variations and environmental factors driving adaptive divergence

within the species will help appropriate sourcing material for not only industrial use but also for tree improvement (Thangjam et al. 2020). Scanning of provenances/seed source capable of providing best-adapted trees is desirable for naturally available genetic variation to utilize the best material for maximum productivity and for further breeding program (Shiv Kumar and Banerjee 1986). Seed traits are crucial fitness-related traits that are expected to underpin survival and reproductive success of plants in different environments. Seed size is important physical indicator of seed quality that affects vegetation growth and field performance. Seed mass of species represent a complex adaptive compromises (Harper et al. 1970) and plays a vital role in the establishment of the juvenile phase of a plant's growth curve.

Genetic diversity of *Madhuca indica* is well reported (Singh and Singh 2005, Wani and Wani 2017) and the genetic variance within-population population can be even to an extent of 85% in *M. indica* and 71% in *M. hainanensis* (Nimbalkar et al. 2018). Similarly, the pattern of field/environmental variations in 20 genotypes of this species in and around Allahabad district of Uttar Pradesh was studied by Wani and Ahmad (2013).

Besides, relationship between seed traits and initial progeny growth performance of 23 genotypes of this species in Jharkhand state, India was studied by Divakar (2014). The species thrives well (Nayak and Sahoo 2020b) and shows wide variations in morphological traits and oil contents across different agroclimatic zones in Odisha (Nayak et al. 2020), however, how the seeds drawn from different sources influence the early growth performance of its seedlings is mostly lacking in Odisha. The objective of this paper was to evaluate the effect of seed source on germination and early growth characters of Mahua seedlings in Odisha.

MATERIALS AND METHODS

Sampling Design

Seeds were collected from ten provenances (Figure 1) whose geo-coordinates, altitude and climatic attributes are given in Table 1. The details of the physico-chemical properties of the seed collection sites are given in Table 2. From each provenance, ten phenotypically superior candidates plus tree (CPT) aged (25-30 years) having good history of seed production and without insect or

pest attack weremarked for collection of fruits. Sampling was carried out in large stands which are shown to exhibit high rates of outcrossing. Within a provenance matured fruits were collected from trees spread at least 100 m apart, to avoid sampling from close relatives. From each tree, 3 random samples of fruits (each containing 100 fresh, ripened) were collected during the month of May-June. These fruits were dried in shade for 10 days before seed extraction using secateurs. Fruits collected from different maternal trees were kept separate, to enable population trait variation within provenances for estimation of heritability. The seeds and kernels were measured for their morphological variations (size, weight) using a seed calliper (for size) and a sensitive balance (for weight).

The seeds from 10 CPT of each provenance were sown in black polythene bag (size 20x15 cm) filled with a mixture of silt and soil in 2:1 ratio kept in the experimental nursery of College of Forestry, Orissa University of Agriculture and Technology, Bhubaneswar. Each sample tree was considered as a treatment within each provenance for which three replications were laid out in randomized block design, totaling 300 polybags by sowing one seed per polybag. Irrigation was applied to keep the soil moist and germination was counted daily. Germination percent (GP) mean daily

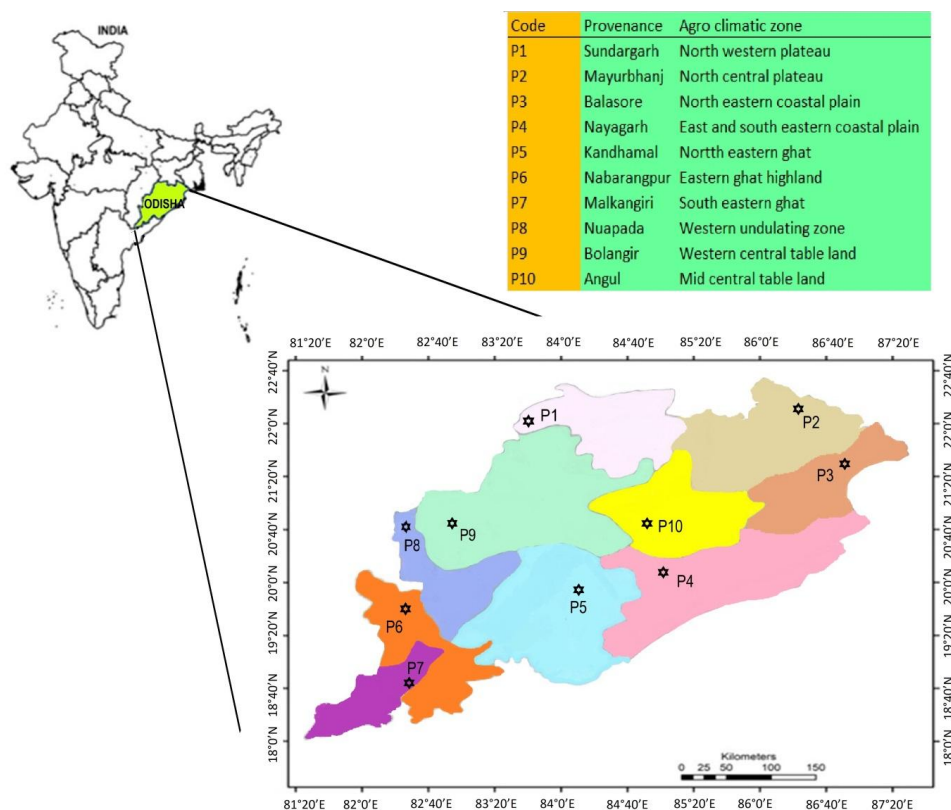


Figure 1. Map of the study site showing 10 seed sources of *M. latifolia* in Odisha, India

Table 1. Characteristics of provenance

Provenance	Forest division	Village/ Site of seed collection	Latitude (decimal)	Longitude (decimal)	Altitude (m)	MAR (mm)	RH (%)	MAT (°C)
P1	Sundargarh	Surulata	21.973°	83.61°	282.32	1183.9	51.2	27.4
P2	Mayurbhanj	Chandbil, Kitabeda	22.135°	86.448°	74.61	1358.9	61.6	26.8
P3	Balasore	Tinkosia Kuladhia	21.489°	86.672°	83.65	1600.1	64.3	28
P4	Nayagarh	Neliguda, Saliagocha	20.363°	84.78°	146.92	1077.8	66.2	28
P5	Kandhamal	Srirampur	20.145°	84.207°	592.35	1232.4	53.1	27.8
P6	Nabarangpur	Kelia	19.534°	82.467°	612.62	1480.7	61.6	26.8
P7	Malkangiri	Boipariguda, Sugriguda	18.779°	82.538°	575.22	1514.3	58.3	25.3
P8	Nuapada	Tanwat, Gobara	20.74°	82.545°	387.63	894.7	57.2	24.3
P9	Bolangir	Kantabhanji Khaprakhol	20.705°	82.944°	297.00	1136.8	53.1	27.8
P10	Angul	Bhagabanpur, Raibahal	20.873°	84.46°	46.25	1172.3	61.8	27.4

MAT-mean annual temperature, MAR-mean annual rainfall, RH- relative humidity

Table 2. Soil characteristics of the seed collection sites

Provenance	Village/ Site of seed collection	Soil pH	EC (dS/m)	SOC (%)	N (kg ha ⁻¹)	P (kg ha ⁻¹)	K (kg ha ⁻¹)	S (kg ha ⁻¹)
P1	Surulata	5.58	0.024	0.82	71.88	7.48	173.04	17.25
P2	Chandbil, Kitabeda	5.92	0.031	0.66	75.00	2.49	187.26	25.25
P3	Tinkosia Kuladhia	6.73	0.083	0.48	100.00	8.25	154.85	12.40
P4	Neliguda, Saliagocha	5.69	0.061	0.93	93.75	4.34	185.01	28.00
P5	Srirampur	6.08	0.092	0.24	118.75	8.25	901.78	13.32
P6	Kelia	6.04	0.041	0.97	90.63	4.51	246.62	37.00
P7	Boipariguda, Sugriguda	5.51	0.063	1.04	96.88	5.58	463.34	26.25
P8	Tanwat, Gobara	5.70	0.04	0.93	125.00	3.7	185.10	33.25
P9	Kantabhanji Khaprakhol	5.85	0.049	0.96	140.63	3.98	289.56	48.00
P10	Bhagabanpur, Raibahal	6.97	0.118	0.45	131.25	7.05	135.4	10.36

EC- electrical conductivity, SOC-soil organic carbon, N-available nitrogen, P-available phosphorus, K-available potassium, S-available Sulphur. The values are mean value of the site. Key as in Table 1.

germination (MDG), peak value (PV) and germination value (GV) of the seeds the sample trees were evaluated following Czabator (1962). After 90 days from seed sowing, seedling length (SL), collar diameter (CD) were measured using a measuring tape. Seedling biomass (BM) was measured after drying the material in an oven at 85°C for 24 hours and shoot vigor index (SV) was calculated by multiplying seedling length with corresponding germination

Data Analysis

Tukey's post hoc test was also performed to determine the significance of pair wise separation of the provenances for germination and seedling growth characters of the seeds. Various genetic parameters like genotypic coefficient of variance (GCV), phenotypic coefficient of variance (PCV) and broad sense heritability (h^2) were estimated for all the seed germination and seedling growth traits. Generalized linear model were used for

variance component analysis and estimation of genotype (σ_g^2) and environmental error (σ_e^2) with the help of restricted maximum likelihood method. PCV, GCV and h^2 were determined following equations given by Loha et al. (2006). Percent data sets (germination percentage) were arcsine transformed before ANOVA to meet normality assumption. Besides, Pearson's correlation was carried out within and among the germination and seedling characters, ecological, edaphic and climatic factors of the different provenances.

RESULTS

Significant differences among provenances were displayed for all seed germination characters (Table 3). Highest variation among germination characters was found in germination value (GV) (CV = 42.23%) and minimum in mean daily germination (MDG) (CV = 25.57%) (Figure 2). Pair wise separation (Tukey test)

Table 3. Variance component analysis for seed germination and seedling growth characters

Source of variation	df	Variance components				Expected mean square
Germination traits		GP	MDG	PV	GV	
Provenance	9	97.48*	0.61*	0.18*	1.95*	$\sigma_e^2 + 10\sigma_g^2$
Error	90	322.53	0.20	0.70	7.07	σ_e^2
Growth traits		SDL	CD	BM		
Provenance	9	2.80*	0.03*	0.02*		$\sigma_e^2 + 10\sigma_g^2$
Error	90	9.63	0.03	0.11		σ_e^2

*Significant at P<0.05, GP-germination percent, MDG-mean daily germination, PV-peak value, GV-germination value, SDL-seedling length, CD-collar diameter, and BM-seedling biomass

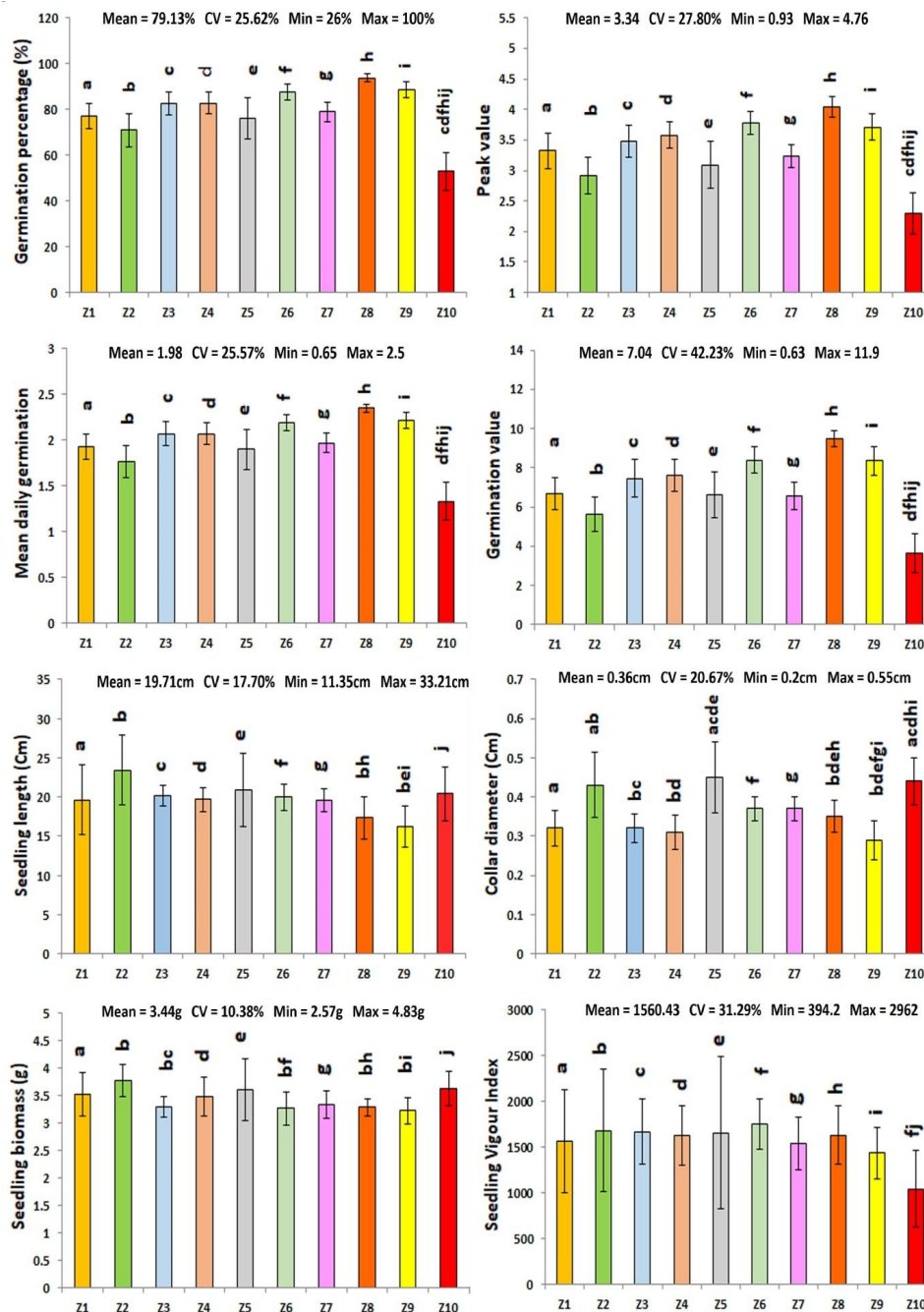


Figure 2. Variation in seed germination and seedling growth characters between provenances. The value with same letter between the provenances are significantly different at P<0.05.

for these characters (GP, PV, MDG and GV) found to differ significantly with other provenances. These characters were maximum at P8 and minimum at P10. Table 10 shows the effect of genotype and environment on the germination and seedling characters. Percentage of seeds germinated among all provenances (GP) ranged from 53% to 93.80% with variation in phenotype (PCV = 22.70) to be more than the variation in genotype (GCV = 12.48). Similarly, Peak value (PV) of germination ranged from 2.30 to 4.04 with PCV (25.07) > GCV (12.67), mean daily germination (MDG) ranged from 1.33 to 2.35 with PCV (22.64) < GCV (39.45) and germination value (GV) ranged from 3.65 to 9.47 with PCV (37.78) > GCV (19.84). Heritability estimate among all the germination traits found highest in mean daily germination ($h^2 = 73\%$) and lowest in germination peak value ($h^2 = 20.34$). Differences between provenances were also found significant ($P < 0.05$) for all the seedling growth characters (SDL, CD, SDB and SV). Variation among seedling growth characters was found highest in seedling vigour (SV)

(CV = 31.29%), followed by length of the seedling (SDL) (CV = 17.70%) and least in biomass content (BM) (CV = 10.38) (Figure 2). However, comparison of broad sense heritability (h^2) among these characters found collar diameter ($h^2 = 50\%$) to be more heritable than seedling length and biomass (Table 4).

Pearson correlation found significant increase in all the germination characters with increase in altitudinal gradient and decreasing longitude (Table 5). For the seedling characters SDL has positive relationship with RF, RH and LON, while BM has positive relation with LON and LAT. Strong positive correlation was observed among all the germination characters and also among all the seedling growth characters (SDL, CD, BM, SV) (Table 6). However, between seed germination and seedling growth characters only SV correlate positively with GP, MDG, PV and GV (Table 6). Influence of seed characters on the final germination traits were also seen in this study (Table 7). For seed characters SWT α SDL; KL α SDL; KT α CD; KW α GP, MDG, PV, GV; and KWT α SDL, CD, BM.

Table 4. Genotype and environment effect on seed germination and seedling growth characters

Characters	Overall mean	Coefficient of variation (%)		Heritability (%)
		Genotype	Environment	
Germination traits				
GP%	79.13	12.48	22.70	23.21
MDG	1.98	39.45	22.64	75.22
PV	3.34	12.67	25.07	20.34
GV	7.04	19.84	37.78	21.62
Growth traits				
SDL	19.71	8.49	15.75	22.54
CD	0.36	48.11	48.11	50
BM	3.44	4.50	9.42	18.60

GP – germination percent, MDG-mean daily germination, PV-peak value, GV-germination value, SDL-seedling length, CD-collar diameter, and BM-seedling biomass

Table 5. Correlation between seed germination and growth characters with geographic characters of provenances

	TEMP	RF	RH	ALT	LON	LAT
GP	-0.162	-0.058	-0.078	.248*	-.199*	-0.12
MDG	-0.162	-0.057	-0.078	.248*	-.199*	-0.12
PV	-0.153	-0.089	-0.047	.212*	-.199*	-0.096
GV	-0.158	-0.103	-0.07	.235*	-.206*	-0.105
SDL	0.118	.242*	.201*	-0.118	.380**	0.145
CD	-0.068	0.092	0.024	0.083	0.138	-0.058
BM	0.127	-0.049	0.028	-0.177	.309**	.223*
SV	-0.047	0.104	0.02	0.169	0.059	-0.016

GP – germination percent, MDG-mean daily germination, PV-peak value, GV-germination value, SDL-seedling length, CD-collar diameter, and BM-seedling biomass, Temp-temperature, RF-rainfall, RH-relative humidity, Alt-altitude, Lon-longitude, Lat-latitude, *, **significant at $P < 0.5$, 0.01 respectively.

Table 6. Correlation among and between seed germination and seedling growth characters

	GP	MDG	PV	GV	SDL	CD	BM	SV
GP	1							
MDG	0.899**	1						
PV	0.919**	0.920**	1					
GV	0.956**	0.956**	0.974**	1				
SDL	0.006	0.006	0.057	0.023	1			
CD	-0.099	-0.099	-0.101	-0.092	.642**	1		
BM	-0.032	-0.032	0.041	0.009	.780**	.533**	1	
SV	.813**	.813**	.770**	.785**	.567**	.275**	.419**	1

GP – germination percent, MDG-mean daily germination, PV-peak value, GV-germination value, SDL-seedling length, CD-collar diameter, and BM-seedling biomass, SV-seedling vigour, *, ** significant at P<0.5, 0.01 respectively.

Table 7. Correlation between germination and growth characters with the seed characters

	SL	SW	ST	HL	SWT	KL	KT	KW	KWT
GP	-0.038	-0.028	0.009	-0.03	0.064	-0.017	-0.03	.235*	0.054
MDG	-0.039	-0.029	0.008	-0.030	0.064	-0.017	-0.030	.234*	0.054
PV	-0.049	-0.006	0.075	-0.061	0.127	-0.062	0.036	.257**	0.092
GV	-0.063	0.006	0.064	-0.059	0.098	-0.076	0.042	.259**	0.088
SDL	.253*	0.112	0.138	0.171	.200*	.228*	0.167	-0.081	.206*
CD	.238*	0.037	0.150	0.075	0.119	0.158	.212*	-0.164	.236*
BM	.234*	0.136	0.122	0.167	0.162	0.115	0.114	-0.099	.203*
SV	0.131	0.058	0.093	0.096	0.166	0.125	0.063	0.140	0.158

SL-seed length, SW-seed width, ST-seed thickness, HL-hilum length, SWT-seed weight, KL-kernel length, KT-kernel thickness, KW-kernel width, KWT-kernel weight, GP-germination percent, MDG-mean daily germination, PV-peak value, GV-germination value, SDL-seedling length, CD-collar diameter, and BM-seedling biomass, SV-seedling vigour, *, ** significant at P<0.5, 0.01 respectively.

DISCUSSION

All provenances showed significant differences in seed germination and seedling characters. Many authors have advocated the importance of correlation between measured traits of genotypes at young age which may help prediction of growth at an advanced stage (Chaturvedi and Pandey 2005). Since the tree has a long gestation period, the study of its growth at juvenile stage is imperative to determine the relative performance of various genotypes. We found strong positive correlation between kernel width with germination characters and kernel weight with seedling growth characters. These findings were also inline as reported by Saverimuttu and Westoby (1996) that the seedling from a larger seeds may have sufficient reserves to continue growth for a much longer period. Many other authors (Owoh et al. 2011, Mtambalika et al. 2014) also reported that seed size and weight also affects the seedling biomass. However, effect on germinability and seedling growth by other maternal factors such as age of the tree, day length, light intensity and the amount of bio-chemical constituents in the seeds could not be ruled out for this species. Rosental

et al. (2014) suggested that metabolic activities involved within seed are mainly responsible for the germination and mobilization of seed reserves normally considered to be apost-germination process responsible for seedling development (Eastmond and Graham 2001, Pritchard et al. 2002). Heavier seeds have reported more oil and better seedling growth in the nursery (Aslan 1975). Kanmaz and Ova (2015) discussed that fat content has no significant correlation with the germination percentage of *Linum usitatissimum* seeds rather according to Ataide et al. (2013) the lipid concentration in seeds of *Dalbergia nigra* remained constant or even increased slightly during germination. However, among physico-chemical properties correlation was found significant for specific gravity, saponification value and iodine value. No effect of concentration of oil content on free fatty acid was found for this species. Similar result was reported by Munasinghe and Wansapala (2015) who studied variation in oil content and fatty acid profile of *M. latifolia* in different agroclimatic zones of Sri Lanka.

The variations in a trait may partly be due to genetic factors and in part due to environmental effect. The degree to which the genetic and/or environment factors

influence the phenotypic traits of *M. latifolia* vary widely. Wani and Ahamad (2013) reported significant influence of environment on seed germination and early growth traits of *M. indica* while Divakar (2014) found that most phenotypic traits of this species was influenced by genetic control. In the present study we encountered intra-provenance variations in germination capacity, growth and seedling vigor in most seed sources. The intra-provenance variation variability associated with the mother effects indicate plasticity within the species and the potential to adopt to wider ecological amplitude. This study also found significant variations among provenances for all the germination and seedling growth traits. The GCV values of the traits evaluated in the present study were lower than the PCV values thus indicating lesser effect of genotypic over phenotypic variations. However, heritability estimate of these traits found huge influence of environmental factors except for the mean daily germination. In a progeny trial of *M. indica* Wani and Ahmad (2013) reported very high to high heritability for various traits such as leaf area (99.39%), germination speed (84.56%), germination per cent (77.9%), mean daily germination (76.7%) and seedling height (73.89%). Divakar (2014) on the other hand reported low to moderate heritability in progeny growth character of this species. Heritability in broad sense provides useful indication about the relative value of selection in the traits in hand. In our study maximum heritability was found for mean daily germination (75.22%) followed by collar diameter (50%). Similar studies on the variability parameter estimates considering genetic parameters were carried out in *Azadirachta indica* (Dhillon et al. 2003), *Pongamia pinnata* (Kumaran 1991), *Dalbergia sissoo* (Dogra et al. 2005), *Jatropha curcus* (Ginwal et al. 2004), *Balanites aegyptiaca* (Elfeel and Warrang 2006, Elfeel 2010, Freigoun et al. 2017) and *Celtis australis* (Singh et al. 2006). Heritability and variations are the key to start a tree improvement programme. Therefore, results from this study could be a key to screen the best traits among a pool of traits that could be improve with successive breeding technique.

CONCLUSION

The results of the present study suggest that Nuapada (P8) is the best provenance for obtaining quality seedling. The extent of variability observed in CPTs across various provenances further offer strategies for tree breeders. GCV values of all traits were found comparatively lower than PCV. The estimate of broad sense heritability indicate that a considerable portion

of variance in the species is additive, which offer good scope for genetic improvement and for potential oil yield for industrial use.

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Author Contributions: SN & UKS designed the study; SN conducted the field survey and data collection; SN & UKS analyzed the data; UKS supervised the entire work; SN wrote the first draft; UKS contributed in final write up. Both authors read and approved the final manuscript.

Conflict of interest: We declare that we have no conflict of interest.

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