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Population analysis, seed biology and restoration of *Hopea erosa* and *H. racophloea*, two critically endangered trees of Western Ghats

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Population analysis, seed biology and restoration of *Hopea erosa* and *H. racophloea*, two Critically Endangered trees of Western Ghats

(Final Report of project KFRI RP 661/2013)

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PROJECT PARTICULARS

1.	Project No.	:	KFRI RP 661/2013
2.	Title	:	Population analysis, seed biology and restoration of *Hopea erosa* and *H. racophloea*, two Critically Endangered trees of Western Ghats
3.	Special area of study	:	Population analysis, reproductive biology and storage physiology of seeds of the targetted species
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6.	Funding Agency	:	KFRI Plan Grant
7.	Duration of the Project	:	June 2013 – May 2016
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Contents

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Western Ghats is one of the biodiversity hotspots in the world due to high level of diversity and endemism, and also one of the richest centers of endemism in India. The southern Western Ghats lodges many Rare, Endangered and Threatened (RET) categories. The Kerala part of Western Ghats covers about 151 RET tree species. In India, the family, Dipterocarpaceae is diversified by 31 species with 16 endemics. The genus *Hopea* belonging to the Dipterocarpaceae family comprises 104 species. It is one of the significant groups of trees and some of them are red listed by the IUCN. Seven species of *Hopea* are reported from Kerala, and all of them are endemic to Western Ghats. Among them, *H. erosa* (Ccritically Eendangered) and *H. racophloea* (Eendangered) are under Threatened category. These two species are sparsely distributed in the Kerala part of Western Ghats. Detailed informations on population structure, reproductive biology and regeneration potential of the two species are lacking. Hence, a study was taken up to fill the above gaps with the objectives - population analysis, reproductive biology and storage physiology of seeds.

Reconnaissance survey was conducted for identifying populations of *H. erosa* and *H. racophloea*, based on the occurrence of the species documented in earlier studies. Quadrat method was used to determine distribution pattern of candidate species and their associates in a community**.** All the trees having Gbh ≥ 30 cm in the study plots were enumerated. Phytosociological analysis was carried out as per standard methods. Vegetative as well as reproductive dynamics were monitored. Floral characteristics were recorded and estimated percentage of fruit setting and fruit to seed setting. Pollen viability was estimated using standard methods. Seed handling techniques and seed storage conditions of the species were standardized.

Populations of *H. erosa* were located at Aralam (Kannur Dist.), Payyanikota and Vanaparvam (Kozhikode Dist.) and Karamanayar (Thiruvananthapuram Dist.). Detailed

population analysis done at Aralam, Payyanikotta and Karamanayar representing each District. *H. erosa* was the third layer species (10-20 m height) in the evergreen forests. It was at 31st position as regard to the dominance among 32 species with a density 1.53 trees ha^{-1} at Aralam, 25th position among 32 species with density 7.65 trees ha^{-1} at Payyankotta and 23rd position among 39 species with 6.12 trees ha^{-1} at Karamanayar. Regeneration of the species was very poor.

Populations of *H. racophloea* were identified at Nadukani (Malapuram Dist.), Rosemala (Kollam Dist.) and Thudimaram (Kannur Dist.). Only a few trees were presented in Thudimaram. Hence, population analysis was done at Nadukani and Rose mala. Vegetation profile of the tree populations showed that *H. racophloea* was the second layer species in the evergreen species. *H. racophloea* was in the 19th position in terms of dominance out of 46 species with 6.12 trees ha^{-1} at Nadukani and 27th position out of 48 species with 4.59 trees ha^{-1} at Rosemala. Regeneration was not bad promising at both the sites. Dominant category of regeneration was under 50-100 cm height class.

Flowering and fruit setting in *H. erosa* was reported only twice in the project period of three years. Flower bud initiation was reported from the end of January. About 22 to 26 days were required for blooming the flower buds. Honey bees play an important role in pollination along with beetles, ants and moths. Anthesis (*in situ*) was during night and it closed by 1.30 am. Pollen ovule ratio was 787:1. An average of 85-90% fertile pollens were present in each flower. Pollen size was 543.92 x 1080μ. Pollen grain shows 86% viability after 5 hr of collection and decreased to 71% after 17 hr. Pollen viability decreased with increased duration of anthesis. Pollen germination was highest at the time of anthesis and decreased towards closing. No gregarious flowering was observed in *H. racophloea* during the project period of three years. However, sporadic flowering was noticed in a few huge trees. Flower bud initiation was from January.

Fruit initiation in *H. erosa* commenced during March and took 4 months for fruit maturity. Young fruits were dark brown. One month after it became green and at maturity it turned to light yellowish with dark brown acrescent sepals. Fruit is a drupe. Average size of mature fruit was 24.12 x 14.3 mm. Mean weight of each seed was 2.176 g and it weighs a range of 350 to 380 seeds per kg. About 50 % of the naturally fallen seeds were infested by borer (*Phycitidae* sp.) and weevils (*Sitophilus vateriae*). A large number of flower buds were shed by fungal infestation (*Cladosporium* sp. & *Pencillium* sp.). Only 6-7% of the flower bud developed in to mature fruits. Fruit initiation in *H. racophloea* commenced from February and extended up to June. Young fruits are light green and turns yellowish green while maturity. Fruit is a nut with two wings having an average weight of 1.6 g (with wing) and 1.2 g (without wing). Seed weight was 940 seeds per kg. Fruit bears alliform sepals.

Mature seeds of *H. erosa* have high moisture content (51.8%) and commenced germination soon after shedding. Its critical moisture content was 47%, below which seeds became non-viable suggesting it into the recalcitrant group. Fresh seeds had 60% germination under laboratory conditions. Seed germination commenced in 6 days after sowing (DAS) and completed within 3 weeks. Naturally fallen seeds had very poor germination (10-15%) due to pest infestation. Fresh seeds of *H. racophloea* had about 35% moisture content and its critical moisture level was 30%, below which it became non-viable. Therefore, it comes under recalcitrant group. Cumulative germination of de-winged seeds was 77 per cent; however, seeds with wings have poor viability with 11% Germination. Germination commenced on 5 DAS and completed within 25 days. De-winging is a pretreatment to improve seed germination.

Seed viability of *H. erosa* maintained for 3½ months when seeds stored in earthen pot inside wet saw-dust (EP) at 20^{o}C. Germination decreased to 55.5% after 15 days of storage and further to 20% at 105 days and then to 3.3% at 120 days. Similarly, critical moisture content of seeds also maintained in EP at 20^{o}C. Seed viability of *H.*

racophloea maintained for 11 months when stored in EP at 20^{o}C. Seed viability decreased to 68.80% after 15 days of storage and further to 13.30% at 300 days and drastically decreased to 2.20% at 330 days. The study revealed that seeds in EP at 20^{o}C are the most effective storage condition for both the species of *Hopea*. The study indicated that storage condition plays an important role in conservation of recalcitrant seeds.

Survivability of regeneration under *in situ* condition was very poor (*H. erosa*: 0.5 - 0.8 %, *H. reacphloea*: 2.5 – 2.6 %) due to pest infestation (*Pythium/Fusarium* sp.). More or less same condition was in the restoration process also (*H. erosa* - 5 % & *H. racophloea* - 12 %). It is concluded that the study generated information on population structure of *H. erosa* and *H. racophloea*, and identified factors responsible for its rarity. Explored the causes for short viability of seeds and standardized optimum conditions for long-term seed storage. This study generated basis of essential database of the species towards their conservation efforts and supports further research. The study is benefitted in academic, forestry and commercial sectors. Being timber yielding trees, seedling production and augment planting *in situ* would ensure their sufficient supply of resources and being a sub-canopy species in the wet evergreen forest ecosystems, the augmentation of plants would enable the reconstitution of particular ecosystems and thereby improves productivity of the landscape. The data generated will be helpful for developing conservation protocols and management of existing populations through augmentation of the species.

1. INTRODUCTION

Forests of Western Ghats is one among the best representatives of non-equatorial tropical evergreen forests in the world and one of the richest centers of endemism in India; about 63% of India's arborescent evergreen taxa are endemic to the Western Ghats (Pascal, 1988 & 1991). Due to high level of biodiversity and endemism, it became one of the biodiversity hotspots of the world (Myers, 1988). MacKinnon and MacKinnon (1986) estimated 1500 endemic plant species in the Western Ghats. Latter, Nayar *et al*. (2014) reported 2,253 species are endemic to India and 1,273 of them are exclusively confined to the Western Ghats. The southern Western Ghats consists around 1,100 flowering species under Rare, Endangered and Threatened (RET) categories. Among them, 495 species are recorded from the Kerala part of Western Ghats and it covers 151 tree species (Sasidharan, 2003). The major causes of plant rarity are often associated with ecology and biology of the species which finally lead to the endangerment. Efforts like species recovery studies are therefore needed urgently to conserve and maintain genes, species and ecosystems along with sustainable use of biological resources (Jose & Pillai, 2014).

Dipterocarpaceae is one of the main timber families in Southeast Asia that forms high proportion of the emergent and main canopy strata (Manokaran, 1996). Members of Dipterocarps have vital role in timber market as potential timber species and also a source of non-timber products for the livelihood of forest dwellers (Poore, 1989; Panayotou & Ashton, 1992). The family, Dipterocarpaceae has 17 genera with more than 500 species. Of which, 10 genera and 99 species are distributed in South Asia (FAO, 1985; Maury-Lechon & Curtet, 1998). In India, the family is diversified by 31 species with 16 endemics (peninsular India - 14, North East – 1, and Andaman Islands - 1) from 5 genera (Tewary & Sarkar, 1987).

The genus *Hopea* belongs to the family of Dipterocarpaceae comprises 104 species, naturally distributed in Sri Lanka, southern India to southern China, and southward throughout Malaysia to New Guinea. Most of them are canopy trees found in wet evergreen forests (100-1000 m asl). Eighteen species are reported from South Asia and 10 species from India (KFRI, 1978; Appanah & Turnbull, 1998). They are the source of

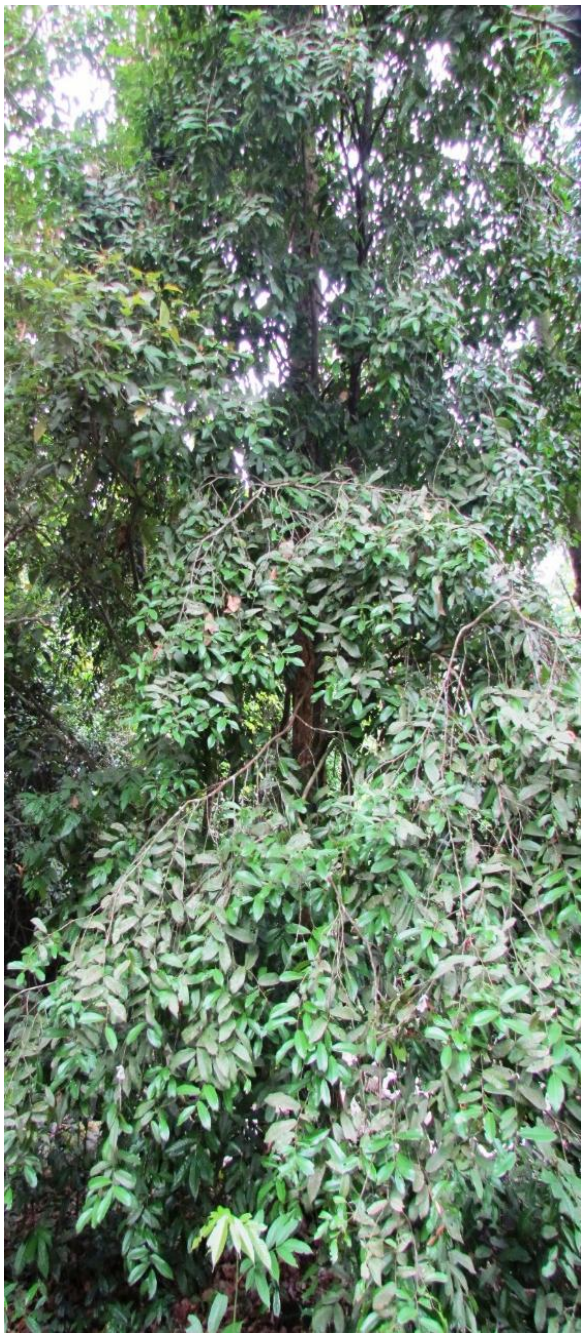
damar resin used in varnishes. Wood is durable and used for making boats, bridges and house construction (Ramesh & Pascal, 1997). *Hopea* is one of the significant groups of trees and some of them are red listed by the IUCN. Seven species of *Hopea* are reported from Kerala, and all of them are endemic to Western Ghats (Sasidharan, 2003). Among them, *H. erosa* (Bedd.) van Sloot. and *H. Jacobi* C.E.C. Fisch. are critically endangered. *H. glabra* Wight & Arn., *H. racophloea* Dyer and *H. utilis* (Bedd.) Bole are endangered, whereas *H. parviflora* Bedd. and *H. ponga* (Dennst.) Mabb. are vulnerable (Nayar, 1996; IUCN, 2012). Information on population structure, reproductive biology and regeneration potential of most of the species are meagre.

Hopea erosa and *H. racophloea* are Threatened species, which sparsely distributed in the Kerala part of Western Ghats. Population of *H. erosa* is reported from the evergreen forests of northern part and *H. racophloea* from the evergreen forests of southern part. *Hopea erosa* is a sub-canopy tree of about 18 m height mostly distributed on banks of streamlets. *Hopea racophloea* is a tall tree up to 35 m height. Extensive studies have not been undertaken on these two species except for a few review works (KFRI, 1978; Appanah & Turnbull, 1998). A preliminary work shows that wood and leaf extracts of *H. erosa* shows antioxidant activity (Vidya *et al*., 2013). Population structure, reproductive biology and regeneration potential of these two species are lacking. A brief overview of *H. erosa* and *H. racophloea* are presented in Table 1.

The present study was envisaged to understand population structure, reproductive biology and seed biology including seed storage and biochemical variation during shelf life. Study on phenological event is useful to understand natural regeneration potential. Timing of recurring biological events provide background for collection and synthesizing detailed quantitative information on rhythms of plant community that helps to understand regeneration process of the species. Information on reproductive biology is important for developing strategies to conserve and restoration. Investigation on biochemical changes during shelf life of seeds helps to assess possible reason for poor viability. Towards this direction, the study was focussed on *H. erosa* and *H. racophloea*, the threatened species in the Kerala part of Western Ghats (Figure 1). Following are the objectives of the study - population analysis, reproductive biology and storage physiology of seeds.

Table 1. General features of *H. erosa* and *H. racophloea*

Particulars	**Species**	
Scientific name	*Hopea erosa* (Bedd.) van Sloot.	*Hopea racophloea* Dyer in Hook. f.
Synonym	*Balanocarpus erosa* Bedd.	*Hopea malabarica* Bedd.
Local names	Eeyakam, Karakong	Naduvalippongu, Naikambagam Thondupoliyan-pongu
Global distribution	Southern western Ghats Endemic to the Western Ghats with disjunct distribution (Agasthyamalai Hills and western Anamalai in South Sahyadri and Ghats Wayanad in Central Sahyadri) (Sasidharan, 2004)	Peninsular India Endemic to the Western Ghats with disjunct distribution (Agasthyamalai Hills in South Sahyadri and Nilambur Ghats and Brahmagiris in Central Sahyadri (Sasidharan, 2004)
Distribution in Kerala	Kannur, Wayanad, Palakkad, Thrissur & Kollam Districts	Malappuram, Palakkad, Kollam & Thiruvananthapuram Districts
Type of forest of occurrence	Evergreen forests Sub-canopy trees in low elevation wet evergreen forests often common near streams, up to 700 m.	Evergreen forests Canopy trees in wet evergreen forests between 300 and 700 m.
Status	Critically Endangered (A1d+2d, B1+2e, C1, D ver 2.3)	Endangered (A1cd+2cd, B1+2c ver 2.3)
General description	Maximum height up to 18 m; bark pale brown, smooth. Leaves simple, alternate, oblong or oblong-lanceolate, apex acute or acuminate, base unequally cordate, margin entire, glabrous, coriaceous; lateral nerves 10-17 pairs, pinnate, arched, prominent, intercostae reticulate, prominent; petiole 17-50 mm, slender, glabrous, swollen tipped; stipule minute, lateral, deciduous. Flowers bisexual, greyish-yellow, subsessile, in unilateral, axillary racemed panicles.	Maximum height up to 35 m; bark dark brown, peeling off in strips, with the lower ends of each strip detached from stem and the upper end still attached to it and curved upwards; Leaves simple, alternate; stipules small, lateral, deciduous; petiole 5-10 mm, slender, glabrous; lateral nerves 4-6 pairs, pinnate, prominent, intercostae scalariform, prominent, domatia present. Flowers bisexual, pinkish-yellow, 2-4 together, in axillary unilateral racemose panicles;



Hopea erosa

Hopea racophloea

Fig. 1. Habit

2. MATERIALS AND METHODS

2.1. Study area

Reconnaissance survey was conducted for locating populations of *Hopea erosa* and *H. racophloea*, based on the occurrence of the species documented in earlier studies (district floras, regional floras, herbaria and other publications) and identified the study areas (Table 2 & Figure 2).

Table. 2. Study sites of *H. erosa* and *H. racophloea*

Location	District	*H. erosa*	*H. racophloea*	Latitude, Longitude & Elevation
Aralam WLS	Kannur	*		11.9740 N; 75.8421 E: Elevation: 169 m
Thudimaram			*	12.0508 N; 75.7841 E: Elevation: 601 m
Vanaparvam	Kozhikode	*		11.4988 N; 75.9720 E: Elevation: 089 m
Payyanikkota		*		11.5870 N; 75.8730 E: Elevation: 198 m
Nadugani	Malapuram		*	11.4337 N; 76.3853 E: Elevation: 565 m
Rosemala	Kollam		*	08.9296 N; 77.1757 E: Elevation: 545 m
Sankili		*		08.7858 N; 77.1190 E: Elevation: 305 m
Ponmudi	TVM		*	08.7554 N; 77.1074 E: Elevation: 691 m
Karamanayar		*		08.6525 N; 77.1914 E: Elevation: 603 m

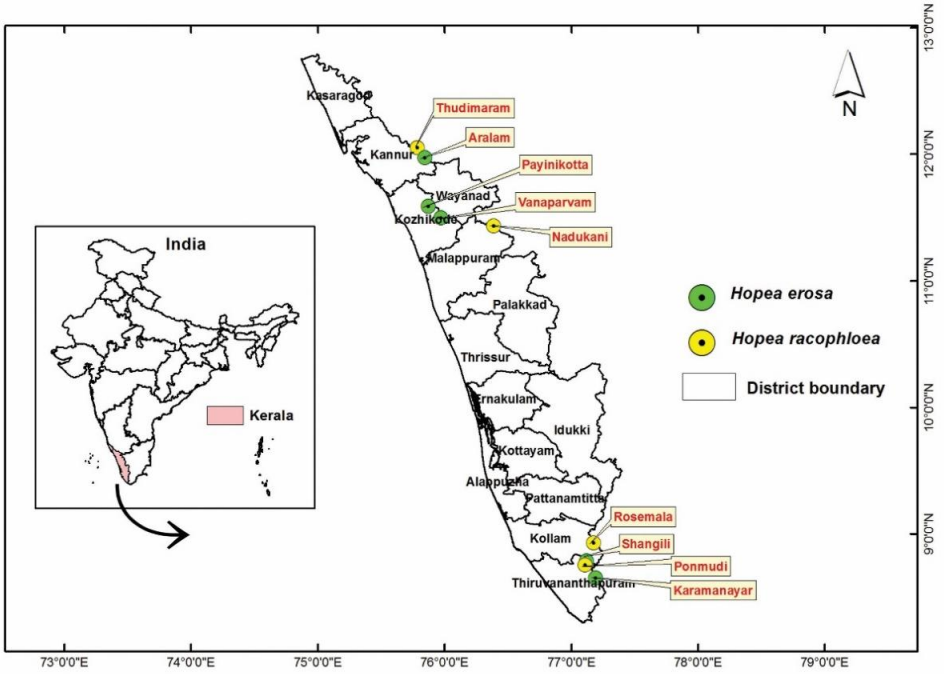


Fig. 2. Distribution of *H. erosa* and *H. racophloea* in the Western Ghats of Kerala

2.2. Population analysis

Monitoring plant population structure is an easy way for understanding environmental conditions of an area (Niemi & McDonald, 2004). Spatial distribution and population structure of a plant species may help in generation of quantitative database (Rama Chandra, 2011). It also helps in assessing loss of ecological services rendered by the species.

Quadrat method was used to determine population structure of tree species and distribution pattern of candidate species in the community**.** Populations of the two species were studied in randomly laid out six sample plots (33 x 33m size in each plot) at each study site. All the trees having girth at breast height (GBH) ≥ 30 cm in the study plots were enumerated and recorded height and GBH (Swarupanandan *et al*., 2013). Floristic diversity in terms of relative frequency, relative density, relative dominance and IVI were calculated as follows (Misra, 1968; Muller-Dombois & Ellenberg, 1974; Sivaram *et al*., 2006).

2.2.1. Density: Number of individuals of a species per unit area gives its density (D). This is computed as trees per ha.

2.2.2. Frequency: The chance of finding a species in a particular area in a particular trial sample is called its Frequency (F) and is expressed as the number of quadrates in which a species is found per total number of quadrates studied.

2.2.3. Basal Area: Area occupied by the stem is known as Basal Area (BA).

Basal area= $\pi r2$, r= GBH/2π.

where,

r = radius of stem at breast height level

GBH = girth of stem at breast height

2.2.4. Importance Value Index (IVI): It is defined as the sum of relative density (RD), relative frequency (RF) and relative basal area (RBA). It expresses as relative importance of the species in the community.

Thus, IVI = RD + RF + RBA

where,

RD = (Density of the species) / (Density of the stand)

RF = (Frequency of the species) / Σ (frequency of all the species)

RBA = (Basal Area of the species) / (Basal Area of all species)

Trees at each study site was classified into three strata based on tree height as: first layer (31 to 40 m), second layer (21 to 30 m) and third layer (10-20 m). Populations of the target species were categorized depending on reproductive phase. Crown projections were measured by four perpendicular radii of the tallest individuals of the species in the quadrate (Pascal, 1988; Parthasarathy & Sethi, 1997).

2.3. Population dynamics

Population dynamics covers the vegetative and reproductive stages of the species. Leaf initiation, maturity and senescence; flowering, fruiting and seed dispersal; regeneration, pest incidence, etc., were monitored and recorded for the study of vegetative as well as reproductive dynamics as follows (Murali & Sukumar, 1994; Daniel & Jayanthi, 1996; Vivek Menon, 2003; Jose *et al*., 2000; Jose & Pandurangan, 2002; Jose *et al*., 2004). Regeneration of the target species having ≤10 cm GBH were counted in all the quadrats. Regenerations were enumerated as un-established seedlings having ≤1 m height and established seedlings having ≥1 m height (Ramachandran *et al*., 2014).

2.4. Reproductive biology

Trees were selected with prominent branch having profuse flowering. Floral characteristics such as color, shape, type of inflorescence, flowering pattern, time of anthesis and closing, mode of pollination, etc., were recorded. Periodical observations were made for evaluating data on number of flowers/fruits/seeds per inflorescence. Estimated percentage of fruit setting and fruit to seed setting using the formula:

$$\text{Fruit setting percentage} = \frac{\text{No. of fruits formed}}{\text{No. of flowers per inflorescence}} \times 100$$

$$\text{Seed setting percentage} = \frac{\text{No. of seeds formed}}{\text{No. of flowers per inflorescence}} \times 100$$

$$\text{Fruit to set percentage} = \frac{\text{No. of seeds in fruit}}{\text{No. of flowers per inflorescence}} \times 100$$

Selected buds were emasculated (15 buds) and recorded periodical events to identify mode of pollination.

2.4.1. Pollen viability

Sampled flowers were dissected and released pollens to a clean slide. Viability of pollens estimated by colorimetric method (Stanley & Linskens, 1974). Pollen viability was scored according to staining level (pollen with bold red colour - viable, colorless - nonviable). Estimated pollen viability as ratio of the number of viable grains to the total number.

2.4.2. Pollen germination

Pollens were germinated in a drop of germination media on a cover glass. Pollen grains were sown on the drops with a clean brush, covered by a cover glass and rested on the cavity slide. Pollens were incubated in different concentrations of sucrose solutions. Germination was determined after 3-6 hours of incubation. Germination was recorded periodically for each incubation period. Pollen grains were counted as germinated if they were equal to minimum of twice the diameter of pre-germinated grains (Khan & Perveen, 2008). Assessed pollen viability in terms of germination percentage. Burst pollen grains were not counted as germinated pollen. Germination percentage was calculated by dividing the number of germinated pollen grains per field of view by total number of pollen per field of view. Recorded measurements of pollen tube length (μm) directly by Leica image Analyzer. Mean length was calculated from average of 20 pollen tubes measured from each drop).

2.4.3. Pollen-ovule ratio

Estimated pollen-ovule ratio by haemocytometer readings. Five samples of anther were evaluated with respect to ovule. The mean value correlate it as pollen ovule ratio (Wyatt*et al*., 2000).

2.5. Seed handling

Naturally fallen seeds were collected from ground as well as mature seeds from trees and used for the study on seed handling.

2.5.1. Seed Moisture content

Moisture content (MC%) of fresh seeds was estimated and open desiccated (air-dry) to the lowest safe moisture content (critical moisture content). MC% of seeds was determined by oven-dry method at 103 ± 2°C for 17 hr (ISTA, 1999).

$$\text{Moisture Content (MC) \%} = \frac{\text{Fresh weight of seed} - \text{Oven dry weight of seed}}{\text{Fresh weight of seed}} \times 100$$

2.5.2. Seed viability test

Germination trials were done for *H. erosa* and *H. racophloea* (n = 30; 4 replicates). Germination pattern of de-winged seeds of *H. racophloea* compared to the seeds with wings. Vermiculite was used as germination medium in all the cases. Observation was done according to the germination test rules (AOSA, 1981). Data were tabulated and computed germination parameters such as germination percentage, germination energy and germination index as follows (Paul, 1972; AOSA, 1983; Panwar & Bhardwaj, 2005).

Germination percentage: (Number of germinated seeds / Total number of seeds) × 100

Germination Energy (GE): The germination energy, defined as the germination percentage when the mean daily germination reached its peak. It is assumed as a measure of the vigour of seedlings.

Germination index (GI): which expressed as speed of germination calculated as described in the Association of Official Seed Analyst (AOSA, 1983).

Speed of germination = n1/d1+n2/d2+n3/d3+----------

where, n = number of germinated seeds

d = number of days

2.5.3. Seed storage

Seeds of both the species were open desiccated under shade to the desired moisture content. To arrive at the desired critical moisture content (lowest safe moisture content) periodic seed viability test (in terms of germination) was adopted as described by Kumar *et al*. (1997). The open desiccated seeds were treated by dusting with systemic fungicide (50% WP Carbendazim - Bavistin) to avoid pathogenic infestation. The seeds were stored under following conditions to standardize optimum storage condition for extending seed longevity.

1) Seeds in open tray at room temperature – about 32^{o} C (OT)
2) Seeds in earthen pot inside wet saw dust at room temperature (EP)
3) Seeds in earthen pot inside wet saw dust at 16^{o}C (16^{o}C)
4) Seeds in earthen pot inside wet saw dust at 20^{o}C (20^{o}C)

Storage container and storage medium is depicted in Figure 3.

Periodical viability tests (germination) of the stored seeds were carried out with 15 days interval in order to evaluate seed longevity and to standardize the best storage condition.

Fig. 3. Seed storage container and medium

2.6. Biochemical evaluation

Biochemical study was conducted to evaluate influence of antioxidant on seed deterioration during storage. Estimated reduced Glutathione (GSH) and Glutathione peroxidases (GPx) by standard methods in stored seeds and compared with fresh seeds. The GSH and GPx assays were carried out using fresh seeds. The seeds were washed thoroughly in running tap water followed by phosphate buffer in order to remove any dirt or soil particles adhered and blotted gently to remove any water droplets. Reduced glutathione was assayed according to the method of Moron *et al*. (1979) and glutathione peroxidase using Rotruck *et al*. (1973).

2.7. Seedling survivability under *in situ* condition

Survivability of seedlings under *in situ* condition was evaluated by monitoring regeneration in a point center quadrats (30 m diameter around mother trees) for two years.

2.8. Statistical analysis

Statistical analysis was carried out using statistical package, SPSS (version 22.0.0.0). One-way multiple analysis of variance was done to test the null hypothesis that there is no statistically significant difference in seed germination, germination energy and germination index. Values were tested for any significance using Independent t-test.

3. RESULTS AND DISCUSSION

3.1. Population structure

Population structure of *H. erosa*, *H. racophloea* and their associate tree species are presented in this section.

3.1.1. *Hopea erosa*

Populations of *H. erosa* were located at Aralam in Kannur Dist., Payyanikota and Vanaparvam in Kozhikode Dist. and Karamanayar in Thiruvananthapuram Dist. Detailed population analysis was done at Aralam, Payyanikotta and Karamanayar representing each District (Tables 3, 5 & 7).

Vegetation profile of the study site at Aralam revealed that *Artocarpus hirsutus* Lam, *Dipterocarpus bourdillonii* Brandis, etc., as the first layer species. *Alstonia scholaris* (L.) R. Br., *Bombax ceiba* L., *Calophyllum calaba* L., *Diospyros candolleana* Wight, *Drypetes venusta* (Wight) Pax & Hoffm, *Euonymus indicus* B. Heyne ex Wall, *Grewia nervosa* (Lour) Panighrahi, *Hopea parviflora* Bedd., *Hydnocarpus pentandra* (Buch.-Ham.) Oken, *Kingiodendron pinnatum* (Roxb. ex DC) Harms, *Lagerstroemia microcarpa* Wight, *Lagerstroemia speciosa* (Linn.) Pers, *Lepisanthes tetraphylla* (Vahl) Radlk, *Mangifera indica* L., *Myristica beddomei* King, *Olea dioica* Roxb, *Persea macrantha* (Nees) Kosterm, *Polyalthia fragrans* (Dalz.) Bedd, *Pterygota alata* (Roxb.) R. Brown, *Sterculia villosa* Roxb, *Solenocapus indicus* Wt. & Arn, *Stereospermum colais* (Buch.-Ham. ex Dillwyn) Mabb, *Syzygium densiflorum* Wall. ex Wt. & Arn., *Vateria indica* L., *Vitex altissima* L.f., etc. as the second layer strata. *Baccaurea courtallensis* (Wt.) M.-A., *Bischofia javanica* Blume, *Canarium strictum* Roxb., *Cinnamomum malabatrum* (Burm. f.) Blume, *Dysoxylum malabaricum* Bedd. ex Hiern, *Ficus hispida* L.f., *Garcinia morella* (Gaertn.) Desr, *Holigarna arnottiana* Hook. f., *Hopea erosa* (Bedd.) van Sloot., *Knema attenuata* (Hook. F. & Thomson) Warb, *Melicope lunu-ankenda* (Gaertn.) Hartley, *Pongamia pinnata* (L.) Pierre, *Reinwardiodendron anamalense* (Bedd.) Mabb., etc. as the third layer species. Dominant species of the vegetation in terms of IVI has shown

that *V. indica* attained with highest Index value (IVI - 47.550) and *H. erosa* (IVI - 1.570) showed 31st position among 32 species with a density 1.53 trees ha^{-1} (Table 3). Population Structure of *H. erosa* in the Aralam site is presented in Table 4. Out of 10 candidate trees, six were in reproductive phase with a height of 10-14 m and remaining were in pre-reproductive phase having 9 to 11 m height.

Table. 3. Pattern of vegetation at Aralam

Position	Species	Family	D	RD	RF	RBA	IVI
1	*Vateria indica*	Dipterocarpaceae	67.34	17.121	6.593	23.836	47.550
2	*Myristica beddomei*	Myristicaceae	45.91	11.673	6.593	8.085	26.352
3	*Hydnocarpus pentandra*	Flacourtiaceae	26.02	6.615	5.495	9.214	21.323
4	*Grewia nervosa*	Tiliaceae	22.96	5.837	5.495	6.675	18.006
5	*Diospyros candolleana*	Ebenaceae	21.43	5.448	5.495	5.033	15.975
6	*Dipterocarpus bourdillonii*	Dipterocarpaceae	18.37	4.669	5.495	5.213	15.376
7	*Drypetes venusta*	Euphorbiaceae	26.02	6.615	5.495	2.302	14.411
8	*Myristica beddomei*	Myristicaceae	13.77	3.502	6.593	1.954	12.049
9	*Hopea parviflora*	Dipterocarpaceae	7.65	1.946	3.297	6.105	11.347
10	*Buccuria courtallensis*	Euphorbiaceae	18.37	4.669	5.495	0.840	11.004
11	*Stereospermum colais*	Bignoniaceae	7.65	1.946	3.297	4.439	9.682
12	*Knema atteunata*	Myristicaceae	9.18	2.335	4.396	2.457	9.187
13	*Holigarna arnottiana*	Anacardiaceae	10.71	2.724	2.198	4.137	9.059
14	*Vitex altissima*	Verbenaceae	7.65	1.946	2.198	3.941	8.084
15	*Cinnamoum malabaratum*	Lauraceae	13.77	3.502	3.297	1.196	7.994
16	*Polyalthia fragrans*	Annonaceae	7.65	1.946	3.297	1.538	6.781
17	*Artocarpus hirsutus*	Moraceae	6.12	1.556	3.297	1.884	6.737
18	*Reinwardiodendron anamalense*	Meliaceae	9.18	2.335	2.198	1.909	6.441
19	*Calophyllum calaba*	Clusiaceae	6.12	1.556	2.198	1.655	5.409
20	*Pongamia pinnata*	Fabaceae	7.65	1.946	2.198	0.939	5.082
21	*Lagestromia speciosa*	Lytheraceae	3.06	0.778	1.099	2.310	4.187
22	*Dysoxylum malabaricum*	Meliaceae	4.59	1.167	2.198	0.496	3.861
23	*Kingiodendron pinnatum*	Fabaceae	3.06	0.778	2.198	0.549	3.525
24	*Olea dioica*	Oleaceae	6.12	1.556	1.099	0.604	3.259
25	*Ficus hispida*	Moraceae	6.12	1.556	1.099	0.349	3.005
26	*Schleichera oleosa*	Sapindaceae	3.06	0.778	1.099	0.950	2.827
27	*Mangifera indica*	Anacardiaceae	3.06	0.778	1.099	0.523	2.401
28	*Lepisanthes teraphylla*	Sapindaceae	3.06	0.778	1.099	0.274	2.151
29	*Alstonia scholaris*	Apocynaceae	3.06	0.778	1.099	0.168	2.045
30	*Humboldtia sp.*	Fabaceae	1.53	0.389	1.099	0.267	1.755
31	*Hopea erosa*	Dipterocarpaceae	1.53	0.389	1.099	0.082	1.570
32	*Canarium strictum*	Burseraceae	1.53	0.389	1.099	0.077	1.565

***Note**: D = Density; RD = Relative Density; RF = Relative Frequency; RBA = Relative Basal Area; IVI = Importance Value Index*

Table 4. Population Structure of *Hopea erosa* at Aralam (Gbh ≥ 30 cm)

Sl. No.	GBH (cm)	Radius (cm)	Basal area (cm^2)	Height at 1st branching (m)	Total tree height (m)
1	65	10.35	336.21	5	13
2	58	09.23	267.70	6	10
3	62	09.87	305.90	7	12
4	50	07.96	198.94	4	9
5	68	10.82	367.97	6	14
6	59	09.39	277.01	5	11
7	67	10.66	357.22	7	13
8	49	07.80	191.07	4	9
9	63	10.03	315.84	7	12
10	56	08.91	249.55	7	10

Species such as *B. courtallensis*, *Diospyros buxifolia* (Blume) Hiern, *Haldina cordifolia* (Roxb.) Ridsdale, *K. pinnatum*, *Myristica malabarica* Lam**.,** *Palaquium ellipticum* (Dalz.) Bail, *P. fragrans*, *V. indica*, *V. altissima*, etc. were noted as the major second layer associates in the Payyanikota site. First layer strata was not observed in the site. *Actinodaphne malabarica* Balakr, *Butea monosperma* (Lam) Taub, *C. calaba*, *Cynometra travancorica* Bedd, *Dillenia pentagyna* Roxb, *D. candolleana*, *Garcinia gummi-gutta* L**.,** *H. arnottiana*, *H. erosa*, *H. pentandra***,** *K. attenuata*, *L. microcarpa*, *Macaranga peltata* (Roxb.) Muell.-Arg., *M. beddomei*, *Oroxylum indicum* (Linn.) Vent, *R. anamalaiense* and *Spondias pinnata* (L. f.) Kurz were recorded as the third layer species. *Diospyros candolleana* (IVI - 20.294) was the dominant species and *H. erosa* (IVI - 4.061) was 25th position among 32 species having density 7.65 trees ha^{-1} (Table 5). Table 6 represents the population structure of *H. erosa* at Payyanikota site. The age profile showed that out of 9 candidate trees, six were in reproductive stage.

Table 5. Pattern of vegetation at Payyanikotta

Sl. No	Species	Family	D	RD	RF	RBA	IVI
1	*Diospyros candolleana*	Ebenaceae	41.32	7.714	5.455	7.125	20.294
2	*Vateria indica*	Dipterocarpaceae	29.08	5.429	2.727	11.369	19.525
3	*Palaquium ellipticum*	Sapotaceae	21.43	4.000	4.546	7.874	16.419
4	*Myristica malabarica*	Myristicaceae	35.20	6.571	4.546	4.873	15.990
5	*Diospyros buxifolia*	Ebenaceae	19.90	3.714	4.546	6.944	15.204
6	*Vitex altisima*	Verbenaceae	22.96	4.286	3.636	6.566	14.488
7	*Kingiodendron pinnatum*	Fabaceae	16.84	3.143	3.636	6.974	13.754
8	*Hydnocarpus pentandra*	Flacourtiaceae	19.90	3.714	5.455	4.206	13.375
9	*Cinnamomum verum*	Lauraceae	21.43	4.000	3.636	5.723	13.359
10	*Polyalthia fragrans*	Annonaceae	29.08	5.429	3.636	4.102	13.167
11	*Oroxylum indicum*	Bignoniaceae	32.14	6.000	5.455	1.698	13.152
12	*Baccaurea courtallensis*	Euphorbiaceae	27.55	5.143	6.364	1.184	12.691
13	*Haldina cordifolia*	Rubiaceae	18.37	3.429	3.636	4.597	11.662
14	*Holigarna arnottiana*	Anacardiaceae	22.96	4.286	4.546	2.750	11.581
15	*Butea monosperma*	Fabaceae	22.96	4.286	4.546	2.516	11.347
16	*Knema attenuata*	Myristicaceae	24.49	4.571	3.636	2.770	10.977
17	*Actinodaphne malabarica*	Lauraceae	19.90	3.714	3.636	1.941	9.291
18	*Dillenia pentagyna*	Dilleniaceae	12.24	2.286	3.636	3.196	9.118
19	*Garcinia gummi-gutta*	Clusiaceae	13.77	2.571	3.636	2.542	8.750
20	*spondias pinnata*	Anacardiaceae	16.84	3.143	2.727	1.585	7.455
21	*Reinwardtiodendron anamalaiense*	Meliaceae	10.71	2.000	1.818	1.654	5.472
22	*Macaranga peltata*	Euphorbiaceae	9.18	1.714	2.727	1.001	5.442
23	*Schleichera oleosa*	Sapindaceae	9.18	1.714	1.818	1.472	5.004
24	*Cynometra travancorica*	Fabaceae	6.12	1.143	1.818	1.495	4.456
25	*Hopea erosa*	Dipterocarpaceae	7.65	1.429	1.818	0.814	4.061
26	*Myristica beddomei*	Myristicaceae	6.12	1.143	0.909	0.979	3.031
27	*Calophyllum calaba*	Clusiaceae	4.59	0.857	0.909	0.596	2.362
28	*Xanthophyllum arnotianum*	Xanthophyllaceae	4.59	0.857	0.909	0.257	2.023
29	*Dimocarpus longan*	Sapindaceae	3.06	0.571	0.909	0.449	1.930
30	*Archidendron bigeminum*	Fabaceae	3.06	0.571	0.909	0.126	1.606
31	*Cynometra beddomeii*	Fabaceae	1.53	0.286	0.909	0.369	1.564
32	*Lagestroemia microcarpa*	Lytharaceae	1.53	0.286	0.909	0.258	1.453

***Note**: D = Density; RD = Relative Density; RF = Relative Frequency; RBA = Relative Basal Area; IVI = Importance Value Index*

Table 6. Population Structure of *Hopea erosa* at Payyanikota (Gbh ≥ 30 cm)

Sl. No.	GBH (cm)	Radius (cm)	Basal area (cm^2)	1st branching at (m)	Tree height (m)
1	55	08.75	240.72	6	10
2	66	10.50	346.64	9	12
3	71	11.30	401.15	7	13
4	63	10.03	315.84	6	12
5	50	07.96	198.94	5	9
6	57	09.07	258.55	5	9
7	58	09.23	267.70	6	10
8	61	09.71	296.11	7	12
9	64	10.19	325.95	7	13

Similar to Payyanikota, no first layer strata was observed from the study sites at Karamanayar. *Actinodaphine bourdillonii* Gamble, *Antiaris toxicaria* (Pers.) Lesch., *Bridelia retusa* (L.) Spreng, *Cassia fistula* L., *D. buxifolia*, *Dipterocarpus indicus* Bedd and *H. parviflora* were the second layer species. *Albizia chinensis* (Osb.) Merr, *B. courtallensis*, *Bridelia spinosa* (Roxb.) Willd., *C. calaba*, *C. strictum*, *Careya arborea* Roxb**,** *Caryota urens* L., *C. malabatrum*, *Cynometra beddomei* Prain., *Dipterocarpus indicus* Bedd, *D. pentagyna*, *Dimocarpus longan* Lour, *D. candolleana*, *D. venusta*, *Elaeocarpus serratus* L**,** *Flacourtia montana* Graham, *G. gummi-gutta*, *Grewia tiliifolia* Vahl, *Holarrhena pubescens* (Buch.-Ham.) Wall. ex G. Don, *H. erosa*, *H. parviflora*, *K. attenuata***,** *M. peltata*, *Mimusops elengi* L, *M. beddomei*, *M. malabarica*, *P. pinnata*, *Terminalia bellerica* (Gaertn.) Roxb., *Syzygium cumini* (L.) Skeels and *V. indica* were under the third layer strata. Age phase of selected candidate species shows that only two trees was in set of future. *Hopea parviflora* (IVI - 24.139) was the dominant species and *H. erosa* (IVI - 4.953) was 23rd position among 39 species with 6.12 trees ha^{-1} (Table 7). Populations of the candidate species, *H. erosa* was recorded only in scattered distribution along the banks of streamlets in all the study areas. Population structure of the species in Karamanayar study area is given in the Table 8. Only two small trees (9-12 m ht) of the species was recorded from the study site. Most of the regeneration of the species were <100 cm height.

Table 7. Pattern of vegetation at Karamanayar

Sl. No.	Species	Family	D	RD	RF	RBA	IVI
1	*Hopea parviflora*	Dipterocarpaceae	15.30	5.181	4.651	14.306	24.139
2	*Vateria indica*	Dipterocarpaceae	18.37	6.218	4.651	9.611	20.480
3	*Baccuria courtallensis*	Euphorbiaceae	24.49	8.290	5.814	1.849	15.953
4	*Vitex altisima*	Verbenaceae	12.24	4.145	3.488	7.431	15.065
5	*Cinnamomum malabatrum*	Lauraceae	15.30	5.181	4.651	4.396	14.228
6	*Grewia tilifolia*	Tiliaceae	10.71	3.627	4.651	5.420	13.698
7	*Artocarpus hirsutus*	Moraceae	7.65	2.591	2.326	8.470	13.386
8	*Myristica malabarica*	Myristicaceae	13.77	4.663	3.488	3.342	11.493
9	*Diospyrous buxifolia*	Ebenaceae	7.65	2.591	2.326	6.353	11.269
10	*Syzygium cumini*	Myrtaceae	12.24	4.145	4.651	2.315	11.111
11	*Diospyrous candolleana*	Ebenaceae	13.77	4.663	3.488	2.318	10.469
12	*Elaeocarpus serratus*	Elaeocarpaceae	7.65	2.591	4.651	2.720	9.961
13	*Bridelia retusa*	Euphorbiaceae	7.65	2.591	4.651	2.400	9.642
14	*Canarium strictum*	Burseraceae	9.18	3.109	3.488	2.868	9.465
15	*Dipterocarpus indicus*	Dipterocarpaceae	10.71	3.627	2.326	2.901	8.853
16	*Antiaris toxicaria*	Moraceae	4.59	1.554	2.326	4.415	8.295
17	*Myristica beddomei*	Myristicaceae	10.71	3.627	2.326	2.334	8.287
18	*Dillenia pentagyna*	Dilleniaceae	7.65	2.591	3.488	0.859	6.938
19	*Mimosoups elengi*	Sapotaceae	6.12	2.073	2.326	2.199	6.597
20	*Terminalia bellerica*	Combretaceae	4.59	1.554	1.163	2.987	5.704
21	*Caryota urens*	Arecaceae	6.12	2.073	2.326	1.289	5.687
22	*Dimocarpus longan*	Sapindaceae	7.65	2.591	2.326	0.752	5.668
23	*Hopea erosa*	Dipterocarpaceae	6.12	2.073	2.326	0.555	4.953
24	*Holarhnea pubescens*	Apocynaceae	6.12	2.073	2.326	0.257	4.655
25	*Pongamia pinnata*	Fabaceae	4.59	1.554	2.326	0.505	4.385
26	*Albizia chinensis*	Fabaceae	4.59	1.554	2.326	0.413	4.293
27	*Drypetes venusta*	Euphorbiaceae	4.59	1.554	1.163	1.084	3.801
28	*Knema attunata*	Myristicaceae	4.59	1.554	1.163	0.545	3.262
29	*Hydnocarpus pentandra*	Flacourtiaceae	3.06	1.036	1.163	0.933	3.132
30	*Calophyllum calaba*	Clusiaceae	3.06	1.036	1.163	0.919	3.118
31	*Carya arborea*	Lecythidaceae	4.59	1.554	1.163	0.210	2.927
32	*Garcinia gummigutta*	Clusiaceae	3.06	1.036	1.163	0.644	2.843
33	*Macaranga peltata*	Euphorbiaceae	3.06	1.036	1.163	0.628	2.827
34	*Pterocarpus marsupium*	Fabaceae	3.06	1.036	1.163	0.531	2.730
35	*Schlecheria oleosa*	Sapindaceae	3.06	1.036	1.163	0.341	2.540
36	*Flacourtia montana*	Flacourtiaceae	3.06	1.036	1.163	0.204	2.403
37	*Cynometra beddomei*	Fabaceae	1.53	0.518	1.163	0.592	2.273
38	*Cassia fistula*	Fabaceae	1.53	0.518	1.163	0.087	1.768
39	*Actinodaphine bourdilloni*	Lauraceae	1.53	0.518	1.163	0.018	1.699

***Note**: D = Density; RD = Relative Density; RF = Rerlative Frequency; RBA = Relative Basal Area; IVI = Importance Value Index*

Table 8. Population Structure of *Hopea erosa* at Karamanayar (Gbh ≥30 cm)

Sl. No.	GBH (cm)	Radius (cm)	Basal area (cm^2)	1st branching at (m)	Tree height (m)
1	60	9.55	286.48	7	12
2	39	6.21	121.04	4	9

3.1.2. *Hopea racophloea*

Populations of *H. racophloea* was identified at Nadukani in Malapuram District, Rose mala in Kollam District and Thudimaram in Kannur District. Only a few individual trees were encountered at Thudimaram. Hence, population analysis was done only at Nadukani and Rosemala (Tables 9 & 11). Vegetation profile of the population at Nadukani showed that *Cullenia exarillata* Robyns and *Artocarpus hirsutus* Lam were the species of first layer strata. *Alstonia scholaris* (L.) R. Br., *C. calaba, C. strictum, E. serratus, H. arnottiana, Hopea racophloea* Dyer, *Meusa ferrea* L., *M. malabarica, P. ellipticum, Schleichera oleosa* (Lour.) Oken, *T. bellerica* and *Toona ciliata* Roem, were second layer species. *Baccaurea courtallensis, C. urens, Cinnomomum verum* Presl, *D. pentagyna, D. candolleana, E. serratus, K. attenuata, K. pinnatum, M. peltata, M. malabarica, O. dioica, P. fragrans, R. anamalense* and *Solenocarpus indicus* Wight & Arn. were the third layer species. *Palaquium ellipticum* was the dominant species and *H. racophloea* was in 19th position out of 46 species with 6.12 trees ha^{-1} (Table 9). Table 10 represents the population structure of *H. racophloea*. Age profile revealed that all the ten candidate trees were in reproductive stage. Dominant category of its regeneration was 50-100 cm height class.

Vegetation profile of study sites at Rosemala showed that *Artocarpus gomezianus* Wall. ex Trecul ssp. *Zeylanicus* Jarrett, *B. ceiba, D. candolleana, D. bourdillonii, K. pinnatum, M. indica, S. pinnata, T. bellerica, Tetrameles nudiflora* R. Br., *V. indica* and *Xanthophyllum arnottianum* Wight, were the first layer species. *Artocarpus hirsutus, A. toxicaria, B. ceiba, D. candolleana, H. parviflora, H. racophloea, K. attenuata, K. pinnatum, M. peltata, P. ellipticum, P. fragrans, T. bellerica, S. oleosa,* were under second layer category. *Baccaurea courtallensis, H. pentandra, P. fragrans, S. oleosa* and *S. cumini* were the third layer category. *Vateria indica* was the dominant species in the study sites and the rank of *H. racophloea* was 27th position among 48 species with 4.59 trees ha^{-1} (Table 11). Population structure of *H. racophloea* is shown in Table 12.

Age profile of the species at Rosemala showed that all the ten trees were under reproductive phase. In general, population of *H. racophloea* in all the study sites was distributed in scattered patches on hill slopes.

Table 9. Pattern of vegetation at Nadukani

Sl. No.	Species	Family	D	RD	RF	RBA	IVI
1	*Palaquium ellipticum*	Sapotaceae	33.67	13.174	8.571	23.650	45.395
2	*Knema atteunata*	Myristicaceae	35.20	13.773	8.571	10.026	32.370
3	*Melicope lanu ankenda*	Rutaceae	22.96	8.982	8.571	7.523	25.077
4	*Meusa ferrea*	Clusiaceae	22.96	8.982	8.571	6.025	23.578
5	*Cullenia exarillata*	Bombacaceae	10.71	4.192	5.714	11.239	21.144
6	*Holigarna arnottiana*	Anacardiaceae	15.30	5.988	7.143	6.701	19.832
7	*Diospyrus candollena*	Ebenaceae	16.84	6.587	5.714	4.742	17.043
8	*Artocarpus hirsutus*	Moraceae	9.18	3.593	7.143	5.157	15.893
9	*Cinamoum malabatrum*	Lauraceae	12.24	4.790	4.286	3.002	12.078
10	*Myristica malabarica*	Myristicaceae	12.24	4.790	4.286	2.724	11.800
11	*Calophyllum calaba*	Clusiaceae	9.18	3.593	4.286	3.440	11.319
12	*Baccuaria courtallensis*	Euphorbiaceae	13.77	5.389	4.286	0.676	10.351
13	*Terminalia bellerica*	Combretaceae	6.12	2.395	2.857	4.419	9.672
14	*Spondias indica*	Anacardiaceae	6.12	2.395	4.286	2.453	9.134
15	*Dillenia pentagyna*	Dilleniaceae	6.12	2.395	2.857	2.846	8.099
16	*Hopea racophloea*	Dipterocarpaceae	6.12	2.395	2.857	1.078	6.330
17	*Alstonia scholaris*	Apocynaceae	4.59	1.796	2.857	1.459	6.113
18	*Kingiodendron pinnata*	Fabaceae	3.06	1.198	2.857	0.999	5.053
19	*Vateria indica*	Dipterocarpaceae	4.59	1.796	1.429	1.420	4.645
20	*olia dioica*	Oleaceae	3.06	1.198	1.429	0.080	2.706
21	*Caryota urens*	Arecaceae	1.53	0.599	1.429	0.341	2.369

***Note**: D = Density; RD = Relative Density; RF = Rerlative Frequency; RBA = Relative Basal Area; IVI = Importance Value Index*

Table 10. Population Structure of *H. racophloea* at Nadukani (Gbh ≥30 cm)

Sl. No.	GBH (cm)	Radius (cm)	Basal area (cm^2)	1st branching at (m)	Tree height (m)
1	198	31.51	3119.76	9	30
2	110	17.51	0962.89	7	19
3	138	21.96	1515.47	9	22
4	125	19.89	1243.40	8	20
5	97	15.44	0748.74	5	15
6	160	25.46	2037.18	8	20
7	170	27.06	2299.79	8	24
8	150	23.87	1790.49	7	22
9	90	14.32	0644.58	6	18
10	127	20.21	1283.51	11	27

Table11. Pattern of vegetation at Rosemala

Sl.no.	Species	Family	D	RD	RF	RBA	IVI
1	*Vateria indica*	Dipterocarpaceae	42.85	11.429	6.742	11.512	29.682
2	*Diospyros candolleana*	Ebenaceae	33.67	8.980	6.742	10.799	26.520
3	*Schleichera oleosa*	Sapindaceae	29.08	7.755	5.618	4.839	18.212
4	*Polyalthia fragrans*	Annonaceae	21.43	5.714	6.742	4.275	16.730
5	*Terminalia bellerica*	Combretaceae	12.24	3.265	3.371	9.968	16.604
6	*Artocarpus hirsutus*	Moraceae	15.30	4.082	5.618	5.761	15.460
7	*Baccuria courtallensis*	Euphorbiaceae	19.90	5.306	5.618	1.362	12.286
8	*Hopea parviflora*	Dipterocarpaceae	16.84	4.490	3.371	3.664	11.525
9	*Spondias pinnata*	Anacardiaceae	9.18	2.449	3.371	5.471	11.291
10	*Hydnocarpus pentandra*	Flacourtiaceae	12.24	3.265	4.494	3.388	11.147
11	*Antiaris toxicaria*	Moraceae	16.84	4.490	3.371	2.484	10.344
12	*Cinnamomum malabatrum*	Lauraceae	13.77	3.674	4.494	1.451	9.619
13	*Kingiodendron pinnatum*	Fabaceae	9.18	2.449	3.371	3.538	9.358
14	*Tetrameles nudiflora*	Datiscaceae	6.12	1.633	2.247	4.489	8.368
15	*Myristica Sp.*	Myristicaceae	13.77	3.674	1.124	2.218	7.015
16	*Mangifera indica*	Anacardiaceae	7.65	2.041	2.247	2.564	6.852
17	*Macaranga peltata*	Euphorbiaceae	6.12	1.633	3.371	1.791	6.794
18	*Hopea racophloea*	Dipterocarpaceae	12.24	3.265	1.124	2.280	6.669
19	*Syzygium cumini*	Myrtaceae	6.12	1.633	3.371	1.205	6.208
20	*Knema atteunata*	Myristicaceae	7.65	2.041	2.247	1.501	5.789
21	*Xanthophyllum arnottianum*	Xanthophyllaceae	7.65	2.041	2.247	1.480	5.768
22	*Bombax ceiba*	Bombacaceae	4.59	1.225	2.247	1.569	5.041
23	*Dipterocarpus bourdillonii*	Dipterocarpaceae	6.12	1.633	1.124	2.089	4.845
24	*Stereospermum colais*	Bignoniaceae	10.71	2.857	1.124	0.587	4.568
25	*Palaquium ellipticum*	Sapotaceae	4.59	1.225	1.124	1.932	4.280
26	*Drypetes elata*	Euphorbiaceae	7.65	2.041	1.124	0.935	4.099
27	*Gluta travancurica*	Anacardiaceae	1.53	0.408	1.124	2.257	3.788
28	*Vitex ultissima*	Verbenaceae	3.06	0.816	1.124	1.378	3.318
29	*Artocarpus gomezianus ssp. Zeylanicus*	Moraceae	1.53	0.408	1.124	1.003	2.535
30	*Aporosa cardiosperma*	Euphorbiaceae	1.53	0.408	1.124	0.602	2.134
31	*Lannea coromandellica*	Anacardiaceae	3.06	0.816	1.124	0.164	2.104
32	*grewia tilifolia*	Tiliaceae	1.53	0.408	1.124	0.491	2.023
33	*Croton klotzschianus*	Euphorbiaceae	3.06	0.816	1.124	0.075	2.015
34	*Syzygium lanceolatum*	Myrtaceae	1.53	0.408	1.124	0.361	1.893
35	*Vatica roxburghiana*	Dipterocarpaceae	1.53	0.408	1.124	0.303	1.835
36	*Terminalia paniculata*	Combretaceae	1.53	0.408	1.124	0.194	1.726
37	*Phyllanthus acidus*	Euphorbiaceae	1.53	0.408	1.124	0.023	1.554

Table 12. Population Structure of *H. racophloea* at Rosemala (Gbh ≥30 cm)

Sl. No.	GBH (cm)	Radius (cm)	Basal area (cm^2)	1st branching at (m)	Tree height (m)
1	128	20.37	1303.80	8	15
2	86	13.69	588.55	7	12
3	98	15.60	764.26	9	14
4	148	23.55	1743.06	6	25
5	94	14.96	703.15	6	16
6	138	21.96	1515.47	7	20
7	116	18.46	1070.79	10	18
8	118	18.78	1108.04	11	19
9	98	15.60	764.26	7	15
10	87	13.85	602.32	6	13

Hopea erosa is facing extremely high risk of extinction, its distribution is <100 km² due to severe habitat fragmentation and extreme fluctuation in extent of occurrence, hence placed under Critically Endangered (CR) as per IUCN criteria. Similarly, *H. racophloea* is an endangered species because its distribution is <5,000 km^2 due to severe habitat fragmentation and fluctuations in extent of occurrence and facing high risk of extinction (IUCN, 2012). In Kerala, major population of *H. erosa* is reported from the northern part and *H. racophloea* from northern and southern part.

3.2. Reproductive biology

3.2.1. *Flowering phenology*

3.2.1.1. *Hopea erosa*: Figure 4 is represented flower buds, immature flower, blooming, mature flower and fruits of *H. erosa*. Flowering and fruit setting was reported only twice (2014 & 2016) during the project period. It is monecious in nature. Inflorescence is axillary or terminal racemed panicles. Flower bud initiation reported from the end of January. About 22 to 26 days required for blooming the flower buds. Blooming was at around 18.30 hr – 19.30 hr (24 hr clock) and closing during 24.00 hr to 1.30 hr. Each node contains 2-4, commonly 3 inflorescence. Each panicle has about 8-32 buds. Flower buds are globular with swollen tip with pinkish-red. Flowers are grayish-yellow having

pleasant smell during opening. Flowers have five petals, twisted, spathulate and tip acute. Sepals five, imbricate, dark brownish in the two outer and light yellow with dark brownish margin in two inner, remaining half-dark brown and half-light yellow. Fifteen stamens are found in one bud, anthers short, ovate, exceeded by the apical awn. Ovary superior, 3 celled and 2 ovules in each cell. Fruit setting was noticed in emasculated buds; hence, pollination is enabled through cross pollination. Honey bees play an important role in pollination along with beetles, ants and moths. Anthesis dehiscence (*in situ*) was during night (19.15 – 23.30 hr) and closing by 1.30 hr. (Table 13).

Table 13. Phenomenon of anthesis in *H. erosa*

Location	Time of Anthesis	Duration	Flower closing time
Vanaparvam	19.30 - 23.30 hr	4 hrs	23.30 – 1.00 hr
Payyanikotta	19.15 - 23.15 hr	4 hrs	23.15 - 1.30 hr

flower bud initiation | flower buds | blooming | mature flowers | fruits

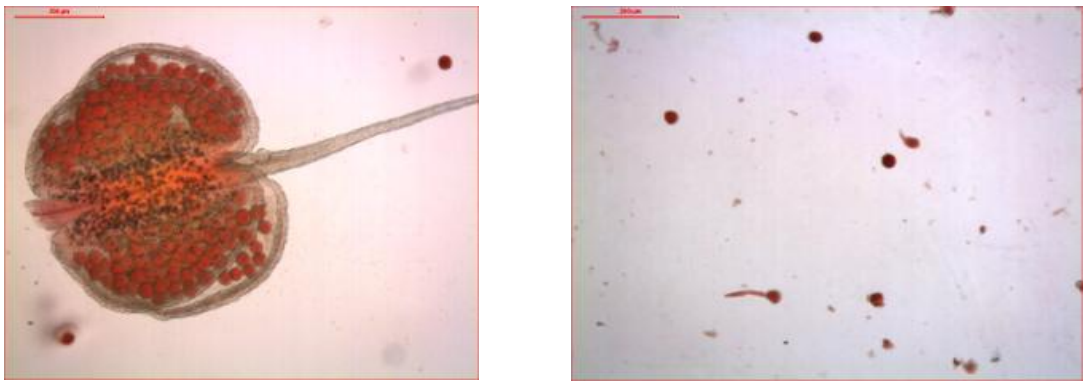
Fig. 4. Flowering and fruiting of *Hopea erosa*

Pollen ovule ratio: Three hundred and fifteen pollen grains were counted in an anther. Fifteen anthers were noticed in a flower. Hence, about 4,725 pollen grains were in a flower. A flower has six ovules and hence pollen ovule ratio was 787:1.

Pollen viability: Acetocarmine test resulted that an average of 85-90 per cent fertile pollens were produced in each flower.

Pollen germination: Figure 5 shows the anther and germinating pollens of *H. erosa*. Average size of pollen was 543.92μ diameter and 1080μ length. Viability of pollens varied with period after collection. Pollen grain shows 86 per cent viability after 5 hours of collection and decreased to 71 per cent after 17 hours. Pollen viability decreased with

increased duration after anthesis. Pollen grains commenced germination in 10 per cent sucrose solution for different incubation period. Germination was highest at the time of anthesis and decreased as to the time of closing. Length of pollen tube varied with the time of incubation period. Average size of pollen grain was 33.07μ diameter under pre-germination period and reduced to 31.01μ after germination commenced.



anther pollen germination

Fig. 5. Anther and pollen germination of *Hopea erosa*

3.2.1.2. *Hopea racophloea*: No gregarious flowering was observed during the project period. However, sporadic flowering was noticed in a few huge trees (>35 m height). Hence, couldn't conduct detailed study on its floral biology. Figure 6 is the flower buds, mature flower and fruits of *H. racophloea*. It is also monecious in nature. Inflorescence is axillary or terminal raceme panicles. Flower bud initiation was from January. Flower is yellowish pink. Inflorescence with 20-30 buds. Sepals five, equal, spathulate, obtuse, glabrous. Petals five, glabrous, hairs present on outside with a twisted flattened appendage at apex. Each flower bears 15 stamens and three-celled superior ovary with two ovules in each cell.

Intermittent flowering with 3-8 years interval is common in most of the Dipterocarps (Wycherley, 1973). Earlier study reported that *H. parviflora* produces flowers and fruits once in two years (Sundarapandian *et al*., 2005). Information on flowering phenology is important for the study of plant-animal interactions and are useful indicators to assess the impact of environmental perturbations on trees (Singh & Singh, 1992; Kushwaha & Singh, 2005). Timing of recurring biological events provide background for seed collection and helps to understand regeneration process of the species.

flower buds flowers fruits

Fig. 6. Flowering and fruiting of *Hopea racophloea*

3.2.2. Fruit development

3.2.2.1. *Hopea erosa*: Fruit initiation was commenced during March. About four months were required for fruit maturity. Young fruits were dark brown. One month after it became green and while maturity it was light yellowish with dark brown acrescent sepals. Fruit is a drupe. Average size of mature fruit was 24.12 x 14.3 mm and ovoid or oblong in shape with acrescent sepals having a size of 2-2.5 cm long. Mean weight of each seed was 2.176 g and it weighs a range of 350 to 380 seeds per kg. About 50 per cent of the naturally fallen seeds were infested by borer (*Phycitidae*sp.) and weevils (*Sitophilusvateriae*). A large number of flower buds were shed by fungal infestation (*Cladosporium* sp. and *Pencillium*sp.). Quantitative observation on flowering and fruiting is presented in Table 14. Only about 6-7 per cent of the flower buds were developed in to mature fruits.

Table 14. Quantitative observation on floral behaviour of *Hopea erosa*

Characters	Study site	
	Vanaparvam	Payyanikotta
No. of flower buds in secondary branches	1139	1307
No. of secondary branches	21	18
No. of flowers in secondary branches	367	403
Fruit setting	82	87

3.2.2.2. *Hopea racophloea*: Fruit formation started in February and extended up to June. Young fruits are light green and turns yellowish green while maturity. Fruit is a nut with two wings having an average weight of 1.6 g (with wing) and 1.2 g (without wing). Seed weight was about 940 seeds per kg. Fruit bears alliform sepals having a size range 7.5 x 2.3 cm, and a single seed possess 10.1 x 8.05 cm.

3.2.3. Seed attributes and seed viability

3.2.3.1. *Hopea erosa*: Figure 7 represents mature fruits and pest infected fruits of *H. erosa* and pests (beetle and weevil) that infested the fruits. Mature seeds had high moisture content (51.8%) and commenced germination soon after shedding from mother trees as in other Dipterocarps reported by Rajeswari and Kaveriappa (2000). Critical moisture content of seeds was 47 per cent, below which the seeds became non-viable. Seeds were short lived and incapable of withstanding desiccation below the critical moisture level. Hence, it is recalcitrant group as stated by Tompsett (1987). Generally, recalcitrant seeds maintain high moisture content at maturity (often >30 to 70%) and are sensitive to desiccation below 12-30 per cent, depending on species (Chacko *et al*., 2002). Earlier studies reported the critical moisture level of recalcitrant seeds as *H. helferi* (47%), *H. nervosa* (43-50%), *H. hainanensis* (35-38%), *Syzigium cumini* (10%), etc. (Tamari, 1976; Sasaki, 1980; Song *et al*., 1984; Anandalakshmi *et al*., 2005).

Seed attributes such as weight, size and moisture content and germination parameters are presented in Table 15. Fresh seeds had 60 per cent germination under laboratory condition. Respective germination energy (GE) and germination index (GI) were 23.30 and 0.28. It showed an epigeal type of germination. Seed germination commenced in 6 days after sowing (DAS) and completed in 3 weeks. Though, it showed better germination, seedling survivability was very poor due to fungal infestation. However, under natural condition (*in situ*), *i.e*., naturally fallen seeds had very poor germination (10-15%), which might be due to pest infestation as mentioned above. Germinating seeds and seedling are shown as Figure 8.

fruits | pest infested fruits | beetle | weevil

Fig. 7. Mature fruits and pest infected fruits of *Hopea erosa*

Table 15: Seeds attributes of *Hopea eroa*

Number of seeds/kg	Length (mm)	Breadth (mm)	MC (%)	G%	GE	GI
353 (350 – 380)	24.12 (22 – 26)	14.3 (13 – 17)	51.8	60	23.30	0.28

Note: *MC = Moisture content; G = germination; GE = germination energy; GI = germination index*

seed germination

seedling establishment

Fig. 8. *Hopea erosa*

3.2.3.2. *Hopea racophloea*: Seed characteristics are presented in Table 16. Fresh seeds had about 35 per cent moisture content with 77 per cent germination (de-winged seeds); however, seeds with wings have poor viability with 11 per cent germination. Germination energy (GE) and germination index (GI) were 47.78 & 1.19 for de-winged seeds and 8.68 & 0.16 for seeds with wings. Germination was epigeal type and germination commenced on 5 DAS and completed with 25 days. Critical moisture level was 30 per cent, below which seeds were non-viable; therefore, it comes under recalcitrant group. The study revealed that de-winged seeds had better germination than seeds with wings; hence, de-winging is a pretreatment to improve seed germination. Figures 9 and 10 are the seed processing, de-winged seeds and seedlings of *H. racophloea*.

de-winging of fruits

de-winged fruits

Fig. 9. *Hopea racophloea*

Table 16. Seed characteristics of *H. racophloea*

Number of seeds/kg		Length (mm)	Breadth (mm)	MC (%)	G%		GE		GI	
WS	DWS	10.1	8.5	34.59	WS	DWS	WS	DWS	WS	DWS
573	936				10.68 ± 1.81	77.36 ± 8.98	8.68	47.78	0.16	1.19

Note: *WS = seeds with wings; DWS = de-winged seeds; MC = Moisture content; G = germination; GE = germination energy; GI = germination index*

Fig. 10. *Hopea racophloea* - seedlings

Since the mean value of germination per cent, GE and GI is higher for de-winged seeds. Levene's test for equality if variances are used to test the null hypothesis that variances among the study groups are significantly equal revealed that germination per cent and GI are not significantly equal. Independent sample t-test is used to test the null hypothesis that no statistically significant difference in mean values. It showed statistically significant difference in germination per cent.

3.2.4. Seed storage

3.2.4.1. *Hopea erosa*: Seed longevity under different storage condition is presented in Table 17. Study resulted that viability of seeds maintained for 3½ months when seeds stored in earthen pot inside wet saw-dust (EP) at 20^{o}C. Germination decreased from 60

per cent (fresh seeds) to 55.5 per cent after 15 days of storage and further to 20 per cent at 105 days and then to 3.3 per cent at 120 days. Viability of seeds maintained for 75 days when stored in EP at room temperature (23.3%) and decreased to 2.2 per cent at 105 days after storage. Seeds stored in open tray at room temperature (OP) maintained viability at 30 days (33.3%) and reduced to 1.1 per cent at 75 days. However, seeds in EP at 16^{o}C maintained viability only for 30 days. Descriptive statistics also showed that Germination per cent, GE and GI is highest in seeds at 20^{o}C.

Table 17. Seed germination of *Hopea erosa* under storage

Storage period (days)	OP	EP at room temperature	EP at 16^{o}C	EP at 20^{o}C
15	44.4	39.5	37.7	55.5
30	33.3	38.8	36.6	48.6
45	17.7	31.1	0.0	40.0
60	11.1	25.5	0.0	34.4
75	01.1	23.3	0.0	31.1
90	0.0	07.7	0.0	25.5
105	0.0	2.2	0.0	20.0
120	0.0	0.0	0.0	03.3
135	0.0	0.0	0.0	0.0

***Note**: OP = Seeds in open tray at room temperature; EP = seeds in earthen pot inside wet saw-dust*

Multivariate analysis revealed statistically significant difference (Wilks' Lamda = 0.372, F = 5.104, p-value = 0.0001) among storage conditions (Table 18).

Table 18. Multivariate tests

Effect		Value	F	Hypothesis df	Error df	Sig.
Intercept	Wilks' Lambda	.042	293.645	4.000	51.000	.0001
Group	,,	.372	5.104	12.000	135.225	.0001

Test of between subject effects showed significant difference in mean GI (F (3, 54) = 3.637, *p-value = 0.018*) among germination conditions. Turkey's post-hoc test revealed that GI is significant difference only among conditions at 20^{o}C and 16^{o}C (MD = 0.160, *p-value = 0.010*).

3.2.4.2. *Hopea racophloea*: Seed longevity of *H. racophloea* under different storage condition is shown in Table 19. The results showed that viability of seeds maintained for about 11 months when stored in earthen pot inside wet saw-dust (EP) at 20^{o}C storage temperature. Seed viability decreased from 77.36 per cent germination (fresh seeds) to 68.80 per cent after 15 days of storage and further to 13.30 per cent at 300 days and drastically decreased to 2.20 per cent at 330 days. Viability of seeds maintained for 105 days when stored in EP at 16^{o}C (15.50%) and decreased to 4.40 per cent at 120 days after storage. Similarly, seeds stored in EP at room temperature maintained viability for 60 days (55.50%) and decreased to 2.20 per cent at 75 days after storage. However, seeds in open tray at room temperature maintained viability only up to 30 days (28.80%). The study resulted that seeds storage in EP at 20^{o}Cis the best storage condition.

Descriptive statistics of other parameters also revealed that mean value of germination energy and germination index is highest in seeds stored in earthen pot at 20^{o}C (Table 20). All the parameters were showed highly significant difference among storage conditions (*p-value = 0.0001*). As there is significant difference in mean value of study variables, Turkey's post-hoc test is conducted to test which two pairs of group have significant difference. Table 21 showed that the parameters at each storage condition significantly different with other conditions.

Seed viability of *H. erosa* and *H. racophloea* retained at different storage regimes. Among them, seeds stored in earthen pot inside wet saw-dust at 20^{o}C showed better seed longevity. Critical seed moisture level also maintained in the same storage regime. Hence, it is the most effective storage condition for the two species of *Hopea*. Majority of the Dipterocarp species are unable to withstand below 15^{o}C (Tompsett, 1992). Previous studies reported that seeds of *H. parviflora* extended viability up to 40 days when stored in mud pot at 10^{o}C (Sunilkumar & Sudhakara, 1998). Optimum seed storage conditions for other species like *Syzigium cumini* (plastic container at 20^{o}C), *Calamus longisetus* (polythene bag inside saw-dust at 4°C) and *H. hainanensis* (laminated aluminium foil packet at 15^{o}C & 20^{o}C) which maintain viability for a while (Anandalakshmi *et al*., 2005; Pillai & Menon, 2011; Lan *et al*., 2012). All the studies indicated that storage condition plays an important role in conservation of recalcitrant seeds.

Table19. Seed germination of *H. racophloea* under storage

Storage period (days)	OP	EP at room temperature	EP at 16^0C	EP at 20^OC
15	58.80	64.40	65.00	68.80
30	28.80	61.10	57.70	57.70
45	0.00	62.20	48.80	52.20
60		55.50	45.80	48.80
75		02.20	37.70	48.40
90		0.00	21.10	45.50
105			15.50	43.30
120			04.40	40.80
150			0.00	40.00
180				34.00
210				31.10
240				27.70
270				16.60
300				13.30
330				02.20
360				0.00

***Note**: OP = Seeds in open tray at room temperature; EP = seeds in earthen pot inside wet saw-dust*

Table 20. Descriptive Statistics of *H. racophloea*

Germination parameter	Storage condition	Mean
Germination energy	Open tray at room temp.	01.646 ± 1.215
	Earthen pot at room temp.	11.811 ± 4.235
	Earthen pot 16°C	14.280 ± 3.522
	Earthen pot 20°C	28.765 ± 3.011
Germination index	Open tray at room temp.	00.039 ± 0.027
	Earthen pot at room temp.	00.335 ± 0.108
	Earthen pot 16°C	00.375 ± 0.095
	Earthen pot 20°C	00.916 ± 0.222

Table 21. Parameters that significant different among the following storage conditions

Germination (%)	OT	EP at 16 & 20 °C
	EP at room temp.	EP at 20 °C
	EP at 16°C	OP & 20 °C
	EP at 20°C	OP, EP at room temp., EP at 16°C
Germinationenergy (%)	OP	EP at 16 & 20 °C
	EP at room temp.	EP at 20°C
	EP at 16°C	OP & EP at 20°C
	EP at 20°C	OP, EP at room temp., EP at 16°C
Germination index	OP	EP at 20°C
	EP at room temp.	EP at 20°C
	EP at 16°C	EP at 20°C
	EP at 20°C	OP, EP at room temp., EP at 16°C

Note: OT = Seeds in open tray at room temperature; EP = Seeds in earthen pot inside wet saw-dust

3.2.5. Biochemical studies

3.2.5.1. *Hopea erosa*: Table 22 presents the result of level of reduced Glutathione (GSH) and activity of Glutathione peroxidases (GPx) in seeds of *H. erosa* under storage and compared with fresh seeds. Activity of GSH in fresh seed was 31.448 ± 1.41 μg per gram of seeds and GPx was 31.674 ± 2.026 μg per gram of seeds.

Table 22. Activity of anti-oxidants on stored seeds (mean ± SE)

Storage condition	Storage period			
	1-month		4-month	
	GSH (μg/g)	GPx (μg/g)	GSH (μg/g)	GPx (μg/g)
Earthen pot at 16^{o}C	30.988 ± 3.036	28.996 ± 2.788	26.398 ± 0.756	15.206 ± 0.478
Earthen pot at 20^{o}C	29.322 ± 1.908	24.182 ± 1.000	23.988 ± 1.100	18.244 ± 1.378

Note: GSH = Reduced Glutathione (GSH); GPx = Glutathione peroxidases

A marginal decrease in the glutathione levels were observed in seeds of *H. erosa* during storage as noted in the Table 22. Similarly, glutathione peroxidase also has decreased during storage. However, the assays could not be performed in seeds stored in ambient condition and earthen pots at room temperature as the seeds were decayed in one month and were inappropriate for biochemical assays.

3.2.5.2. *Hopea racophloea*: Result of the level of reduced Glutathione (GSH) and activity of Glutathione peroxidases (GPx) in seeds of *H. racophloea* under storage is presented in Table 23. Activity of GSH in fresh seed was 133.904 ± 3.238 μg/g and GPx was 69.864 ± 3.592 μg/g.

Table 23. Activity of anti-oxidants on stored seeds of *H. racophloea* (mean ± SE)

Storage condition	Storage period			
	1-month		4-month	
	GSH (μg/g)	GPx (μg/g)	GSH (μg/g)	GPx (μg/g)
Ambient	68.5772 ± 2.08	69.04 ± 1.982	-	-
Earthen pot at room temperature	123.238 ± 1.688	56.696 ± 2.292	108.436 ± 2.398	32.42 ± 1.06
Earthen pot at 16^{o}C	77.32 ± 3.2747	59.916 ± 1.48	61.743 ± 0.812	41.210 ± 0.530
Earthen pot at 20^{o}C	74.372 ± 3.176	48.102 ± 3.292	69.218±1.199	33.580 ± 0.710

In *H. racophloea* seeds, a significant decrease in the glutathione levels were observed during storage. Seeds stored in earthen pot showed stable values and was comparable to fresh seeds. All other storage conditions showed significant decrease in glutathione levels. During storage, the glutathione peroxidase levels were only marginally varied from the fresh seeds. The assays could not be performed during 4^{th} month in seeds stored at ambient condition as the seeds were decayed after one-month storage.

Stress is generally defined as all effects that have negative impact on plants (Taiz & Zaiger, 1998). Active plant growth in mesophile organisms is in between 10-40°C. Any temperature which is under and below that range creates heat stress on metabolic activities of plants (Treshow, 1970). Plant growth is a function of biochemical reactions and those reactions are controlled by enzymes. Most of the chemical reactions increase as two fold by every 10°C increase in between 20-30°C. Temperatures above that range, reaction speed decreases since enzymes step by step denatured or inactivated.

In addition, reactions catalyzed by enzymes depend on completeness of tertiary structure of enzymes. Plants have evolved a complex regulatory network to mediate biotic and abiotic stress responses based on synthesis of reactive oxygen species (ROS), a chemically reactive chemical species containing oxygen, scavenging, and signaling. ROS appear to be involved in dormancy alleviation. In dormant barley grains under control condition, gibberellic acid (GA) signaling and ROS content are low, while ABA signaling is high, resulting in dormancy. Exogenous H_2O_2 does not appear to alter ABA biosynthesis and signaling, but has a more pronounced effect on GA signaling, inducing a change in hormonal balance that results in germination (Bahin *et al*., 2011). Various abiotic stresses lead to the over-production of reactive oxygen species (ROS) in plants which are highly reactive and toxic and cause damage to proteins, lipids, carbohydrates and DNA which ultimately results in oxidative stress. The ROS comprises both free radical (O_2), superoxide radicals; hydroxyl radical (OH), perhydroxy radical (HO_2), alkoxy radicals (RO) and non-radical (molecular) forms (H_2O_2), and singlet ($1O_2$) (Sarvajeet & Narendra, 2010).

In order to survive from these toxic oxygen intermediates, plants made a defense systems that include enzymes such as superoxide dismutases, catalases, ascorbate peroxidases (APX), glutathione S-transferases (GST) and glutathione peroxidases

(GPX) that catalyze the scavenging of ROS. The activities of APX, GST and GPX depend on the availability of reduced ascorbate (ASA) and glutathione (GSH) that are maintained by enzymes such as glutathione reductase (GR), dehydroascorbate reductase (DHAR) and mono dehydro ascorbate reductase (MDHAR) using NAD(P)H as an electron donor (Roxas *et al*, 2000).

Previous studies of transgenic tobacco lines that over express plant glutathione S-transferase/ glutathione peroxidases (GST/GPX) showed substantial improvement in seed germination under stressful conditions (Roxas *et al*., 1997). Earlier study on *H. ponga* revealed a decreasing trend of carbohydrates and increased level of reducing sugars and total phenolics during seed storage, while total free amino acids increases with laps of time (Sukesh & Chandrashekar, 2011). They reported that the increased lipid peroxidation and protein modification together are responsible for reduced seed viability.

3.2.6. Seedlings survivability under *in situ* condition

3.2.6.1. *Hopea erosa*: Table 24 represents regeneration dynamics of *H. erosa* during 2014-15 at two study sites. Only 0.5 - 0.8 per cent survivability of seedlings was noticed. Figure 11(a) is the regeneration under *in situ* condition. High seedling mortality was due to *Pythium/Fusarium* sp.

Table 24. Survivability of natural regeneration of *H. erosa* under *in situ* condition

Duration (2014-15)	Seedling count	
	Vanaparvam	Payyanikotta
August, 2014	262	584
September, 2014	128	376
October, 2014	79	289
November, 2014	49	197
December, 2014	31	112
January, 2015	15	77
February, 2015	12	68
March, 2015	9	45
April, 2015	6	34
May, 2015	4	25
June, 2015	4	13
July, 2015	3	7
August, 2015	2	3

3.2.6.2. *Hopea racophloea*: Seedling survivability of *H. racophloea* during 2014-15 at Nadukani and Rosemala is presented in Table 25. Study showed that about 2.5-2.6 per cent seedlings were survived during the observation period. Figure 11(b) is the regeneration under *in situ* condition. Mortality in regeneration of the species was also due to fungal infestation (*Pythium/Fusarium* sp.).

Table 25. Survivability of natural regeneration of *H. racophloea* under *in situ* condition

Duration (2014-15)	Seedling count	
	Nadukani	Rosemala
August, 2014	722	884
September, 2014	528	676
October, 2014	379	489
November, 2014	249	397
December, 2014	131	212
January, 2015	105	177
February, 2015	72	98
March, 2015	59	75
April, 2015	46	64
May, 2015	34	55
June, 2015	24	33
July, 2015	23	27
August, 2015	18	23

a. *Hopea erosa*

b. *Hopea racophloea*

Fig. 11. Regeneration of *Hopea erosa* and *Hopea racophloea*

3.2.7. Restoration

3.2.7.1. *Hopea erosa*: A total of 100 seedlings having 30 cm height were planted in the natural habitat at Payyanikotta in Kozhikode District. Each seedling was tagged and monitored growth and survivability. High level of mortality was observed due to fungal infestation (*Pythium/Fusarium* sp.) and survived only 5% of the planted seedling.

3.2.7.2. *Hopea racophloea*: Hundred seedlings (30 cm height) of *H. racophloea* were planted in the natural habitat at Rosemala in Kollam District. The seedlings were tagged and monitored growth performance. Twelve per cent of the planted seedlings were survived and high mortality was observed due to fungal infestation. In spite of high mortality, conservation of both the species can be done through augmentation by planting of one-year-old saplings in its natural habitat.

4. CONCLUSIONS

Populations of *H. erosa* were identified at Aralam, Payyanikota, Vanaparvam and Karamanayar. *Hopea erosa* was the 3rd layer species (10-20 m height). It was in 31st position among 32 species at Aralam in terms of dominance, 25th among 32 species at Payyankotta and 23rd among 39 species at Karamanayar. Regeneration of the species was very poor in the study areas. Populations of *H. racophloea* was identified at Nadukani, Rosemala and Thudimaram. It was the 2nd layer species (21-30 m height) and in 19th position out of 46 species at Nadukani and 27th out of 48 species at Rosemala. Regeneration of *H. racophloea* was not bad and most of the saplings were under 50-100 cm height class.

Gregarious flowering and fruit setting was noticed only during 2014 and 2016 in *H. erosa* under the project period (2013-2016). Flower bud initiation commenced from last week of January and about 22 to 26 days were required for blooming the flower buds. Anthesis (*in situ*) was during night and closed by 01.30 hr. Pollen-ovule ratio was 787:1.85 and 90 per cent fertile pollens were in each flower. Pollen size was 543.92 x 1080μ. Pollen viability decreased with increased duration of anthesis. Pollen grain shows 86 per cent viability after 5 hr of collection and decreased to 71 per cent after 17 hr. Pollen germination was highest at the time of anthesis and decreased as to the time of closing. Fruit initiation commenced during March. About four months were required for fruit maturity. Around 50 per cent of the naturally fallen seeds were infested by borer and weevils. Only about 6-7 per cent of the flower bud developed in to mature fruits. Mature seeds have high moisture content (51.8%) and commenced germination soon after shedding. Its critical moisture content was 47 per cent, below which the seeds became non-viable. Fresh seeds had 60 per cent germination under laboratory condition. Naturally fallen seeds had very poor germination (10-15%) due to seed-born pathogen.

No gregarious flowering observed in *H. racophloea* during the project period. However, sporadic flowering noticed in a few huge trees. Hence, couldn't conduct detailed study on floral biology of *H. racophloea*. Flower bud initiation in the species was from

January. Fruit initiation in *H. racophloea* commenced from February and extended up to June. Fresh seeds of *H. racophloea* had about 35 per cent moisture content with 77 per cent germination (de-winged seeds); however, seeds with wings have poor viability with 11 per cent germination. Critical moisture level was 30 per cent, below which seeds were non-viable. De-winging is a pretreatment to improve seed germination. Seed viability of *H. erosa* maintained for 3½ months when seeds stored in earthen pot inside wet saw-dust (EP) at 20^{o}C. Similarly, seed viability of *H. racophloea* maintained for 11 months when stored in EP at 20^{o}C. The study concluded that seeds in EP at 20^{o}C is the most effective storage condition for the two species of *Hopea*. It is indicated that storage condition plays an important role in conservation of recalcitrant seeds. Survivability of natural regeneration as well as restored saplings under *in situ* condition was very poor due to pest infestation.

In general, the study could generate information on population structure of *H. erosa* and *H. racophloea*, and identified factors responsible for its rarity, which will help for developing conservation protocols and management of existing populations through augmentation. Similarly, explored the causes for short viability of seeds and standardized optimum conditions for long-term seed storage that become a reality and thereby large scale seedling production of the species would be possible. This study paved basis of essential database of the species towards their conservation efforts and supports further research. Since the species enjoys both economical and conservational values, the conservation and management practices established through the study would be benefitted in academic, forestry and commercial sectors. Being timber yielding trees, seedling production and augment planting *in situ* would ensure their sufficient supply of resources. Further, being a sub-canopy species in the wet evergreen forest ecosystems, the augmentation of plants would enable the reconstitution of particular ecosystems and thereby improves productivity of the landscape. The data generated through the study could be utilized for the Forest Department to sort out their working plan and future management programmes.

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