INTERNATIONAL JOURNAL OF PLANT, ANIMAL AND ENVIRONMENTAL SCIENCES
IJPAES
Volume-5, Issue-1, Jan-Mar-2015 Coden: IJPAJX-USA Copyrights@2015 ISSN-2231-4490
Received: 24th Nov-2014 *Revised: 29th Dec-2014* *Accepted: 30th Dec-2014*
Research article

FLORAL ECOLOGY AND REPRODUCTIVE BIOLOGY OF *PTEROSPERMUM RETICULATUM* WIGHT AND ARN. (STERCULIACEAE): A VULNERABLE TREE SPECIES OF WESTERN GHATS OF INDIA.

Keshavanarayan P, Rajkumar K and V Sivaram*

Laboratory of Biodiversity Conservation and Apiculture, Department of Botany, Bangalore University – 560056
Corresponding Author: sivaram900@gmail.com Tele fax: 91-80-23181443.

ABSTRACT: *Pterospermum reticulatum* Wight and Arn is a vulnerable according to IUCN red list category (1998). In the present study observations were made on floral ecology and reproductive biology of *Pterospermum reticulatum* at Pilikula reserve forest, Mangalore, Karnataka during 2012-2013. The peak flowering was observed during February to April and anthesis occurred in the evening between 18:30 -22:30hrs. Inflorescences in terminal or axillary fascicles, Flowers are white colour and 5 petals and 5 linear sepals. Stamens are 15, in group of five. Fruits are capsule, brown, 5-angled and 5-valved, development of fruit to dehiscence was observed. Pollen production per flower was counted 3, 93499.9 and pollen ovule ratio was 1:393500. Floral biology including the stigma receptivity, pollen viability, pollen germination and pollen tube growth were recorded during the flowering period. Macro and Micro nutrients of the soil collected from the rhizosphere were analyzed. The factors responsible for declining of the population of *Pterospermum reticulatum* are discussed.
Key words: *Pterospermum reticulatum,* IUCN, Stigma receptivity, Pollen viability, Pollen germination, Macro and Micronutrients.

INTRODUCTION

Pterospermum reticulatum Wight and Arn. is considered as rare and threatened species endemic to evergreen forests of Western Ghats [1, 6] belonging to the family Sterculiaceae. The family of tropical and sub tropical plants comprising nearly 70 genera and 1,500 species. Based on floral anatomy and embryology the family Sterculiaceae is considered as most primitive because of Pentacyclic flowers [12]. *Pterospermum* in Greek word refereed to winged seed [19]. According to IUCN Red Data Book it is vulnerable and commonly called as Malavuram in Malayalam and Mulipolavu or Thopuli in Tamil. It is a tree growing up to 25 meters tall, bark is greyish brown; leaves are simple, alternate, oblong, obovate, white tomentose and stellate hair beneath. Flowers are white to yellowish, axillary fascicles, solitary. sepals 5, linear, lanceolate , stellate hairy outside, petals 5,obovate,oblong, stamens 15 in group of 5, ovary globose, densely tomentose, fruits are capsule ,brown,5angled , 5 valved , seed 4 in each cell, winged at one end.

Pterospermum reticulatum is a medicinally important plant; the stem bark of the plant was used by tribes to treat ulcers, wounds and inflammations and also wood has been used for construction of boating. The population from the natural habitat is rapidly declining due to various factors such as habitat destruction and reproductive constrains [18]. The present study is to investigate the floral ecology covering floral biology, anthesis, pollen production, pollen viability, pollen germination, stigma receptivity and to study the possible reasons for its endemic and vulnerable status of *P. reticulatum.*

In this context, study has been undertaken to know the factors influencing the population decline in the natural habitat. The locals extensively used the wood for the ship making and also as fuel wood for cooking purpose. The seedling, flowers and fruits are predative by several predators which may contribute to the decline of the population and lead to vulnerable status according to IUCN, red list category and status of *Pterospermum reticulatum.*

MATERIAL AND METHODS

Study species: *Pterospermum reticulatum* occurs in two localities in the deciduous forests. Twenty individual trees were selected in the natural populations in Pilikula reserve forest, Mangalore and Bandala Ghats, Sirsi, Uttar Kannada district, Karnataka, Western Ghats, India. The diversity of the species and their location was recorded between June 2011- May 2012 by using GPS. The study site lies between N 12˚55,789 Latitude; E 74˚53.91 Longitude, elevation 349m and N 14˚31. 725; E 074˚39.031; Elevation 1678m, above the sea level (Fig. 1a-d).

Floral biology: Flower phenology was recorded based on flowering initiation, anthesis and anther dehiscence during flowering season of February to March. The anthesis was recorded on a daily basis on the mature floral buds of ten inflorescences. Five trees were marked and twenty five mature buds were observed for recording the time taken (in minutes) to reach various developmental stages from anthesis up to another dehiscence in successive days. Five flowers each from ten trees were selected randomly to study the flower morphology such as sex, shape, size, colour, odour, sepals, petals, stamens and ovary.

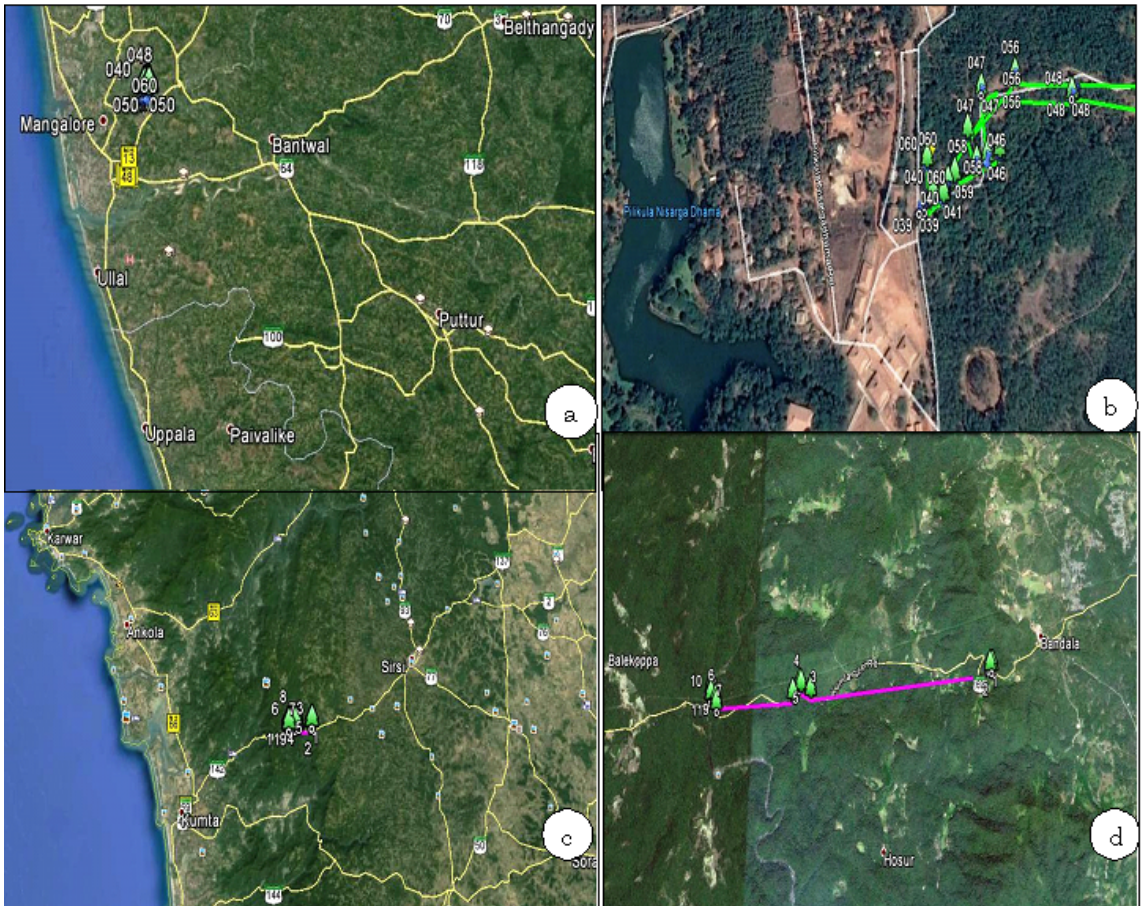

Figure 1.a- Study area; b- Enlarged view by GPS map in Pilikula reserve forest c & d-Map of Bandala Ghats.

Pollen viability and pollen germination: Pollen viability was assessed by 2, 3, 5, -Triphenyl tetrazolium chloride (TTC) tests under different sucrose concentrations. To study of in-vitro pollen germination, pollen was suspended under Brew baker and Kwack media to observe the pollen tube growth and micrometry was used for the measurement of pollen tube length. Pollen germination media was also studied by Hanging drop method. The pollen germination and viability of pollen were observed under light microscope.

Pollen output/anther/flower: Pollen production was recorded on five individual trees during flowering and the pollen count was made by selecting anthers from randomly selected 10 matured buds of *Pterospermum reticulatum.* The anthers taken and stored in the Petri dish and each anther were taken and squashed to observe total numbers of pollen per anther and per flower under light microscope using Haemocytometer (10, 40,100x).

Pollen ovule ratio: The number of ovules per flower was observed by taking the cross section of ovary to determine pollen ovule ratio. Pollen ovule ratio was calculated following [3] method. The formula for pollen ovule ratio given as follows.

P/O ratio = Pollen count per anther× No. of anthers per flower/No. of ovules per flower.

Stigma receptivity: During Anthesis, 3 flowers each from ten inflorescences were selected to test stigma receptivity. Experiment were conducted with Hydrogen Peroxide, a drop is added to stigma of flower during Anthesis [4]. Observations were recorded if stigma showed pure effervescence producing bubbles resulting in catalase enzyme activity (peroxidase). The time, period of effervescence was calculated at different time intervals to study the duration of stigma receptivity.

Floral predators and threats: predators and pests of flowers and fruits were captured in the field using nets and stored in 70% alcohol and they were identified in department of entomology, GKVK, Bangalore.

Soil Analysis: Soil samples were collected in the study area to determine the pH, Micro and Macro nutrients. Total 10 samples were collected from each of 10 marked tress in this area and the analysis was carried out at Biotechnological centre of Karnataka horticulture department, Bangalore.

RESULTS AND DISCUSSION

The genus *Pterospermum* found in the moist deciduous forest and evergreen forest at low altitudes, similar as in the case of *Eriolaena lushingstonii* and *Helicteres isora. Pterospermum reticulatum* is a vulnerable tree species belongs to the family Sterculiaceae. It is an evergreen tree at low elevations and endemic to Western Ghats of India.

Floral biology: The flowering occurs in the beginning of February and ends in April last week. An individual tree shows flowering for 2 to 3 weeks and in some population level is about 4 to 5 weeks. The flowers are solitary, bisexual flower, few flowered cymes in 5-6 numbers (Fig. 2b), pedicellate, laciniate and measuring about 4 cm. corolla 5 with equal shaped petals measuring 2.8cm with milky white colour, calyx is composed of 5 with equal shape- 2.6cm as long as corolla, densely brown tomentose, stamens 15, in groups of 5 with 4 staminodes and each anther size is 1.5, tetra sporangiate and each staminodes 2.8 cm. The ovary globose, pubescent- 2.7cm, 2-loculed and total number of ovules per ovary is 10; style is 2cm long pubescent standing beyond the length of anthers with staminode, stigma wet and simple, (Fig.3a-o; Table.1).

Table 1. Floral morphological characters of *Pterospermum reticulatum*.

Floral parameter	Observations
Flowering period	February to April
Flower shape	Solitary
Flower colour	White
Odour	Fragrant
Nectar	Present
Flower opening time	18:30 to 22:30hrs
Anther dehiscence time	Just after flower opening time
Pollen per anther	2,6233.33
Mean number of pollen per flower	3,93,499
Pollen ovule ratio	1:393500
Pollen morphology	Kurtz type.

Table 2. Stigma receptivity.

Parameters	Results
Fresh Stigma	ARP
After 3 h	ARP
After 6 h	ARP
After 9 h	ARP
After 12 h	ARP
After 15 h	PRP
After 18 h	PRP
After 21 h	PRP
After 24 h	PRP

(ARP - active receptive period, PRP - passive receptive period

Development of flower bud from initial stage takes approximately 90 days (Fig 3. o) to mature flower. The mature flower buds of *Pterospermum reticulatum* open in the evening between 18:30 to 22:30 hrs (Table 1). During flower opening, sepals and petals unfold exposing the stigma along with female reproductive part (Fig 2d), gradually anther dehiscence and stigma receptivity occur after anthesis. The stages of flower buds are shown in the Figure 2c-s. Flower along with the floral parts opens slowly and completely for 3-4 hrs. At this time stigmatic part is completely opened (Fig 2q), and seen in wet condition and dry at the last day of withering. The stigma remains receptive during anthesis and receptivity is lost after 12 hrs and anther dehiscence occurs completely within 2 days. Receptivity of stigmas was scored as positive for peroxidase activity, vigorous bubbling was observed across the entire surface of the stigma within one minute due to applications of the H_2O_2. Stigma attains receptivity soon after anthesis and remains receptive up to 12hrs (Table 2). The stigmatic exudate was also evaluated for its viscosity and glistening appearance (Fig.4a-d), [8, 6, 10]. Anthesis ends 3-4 days and pollinated and fertilized flowers initiate fruit development to produce mature fruits, sometimes flowers fall off if they are not pollinated (Fig. 4 h-k).

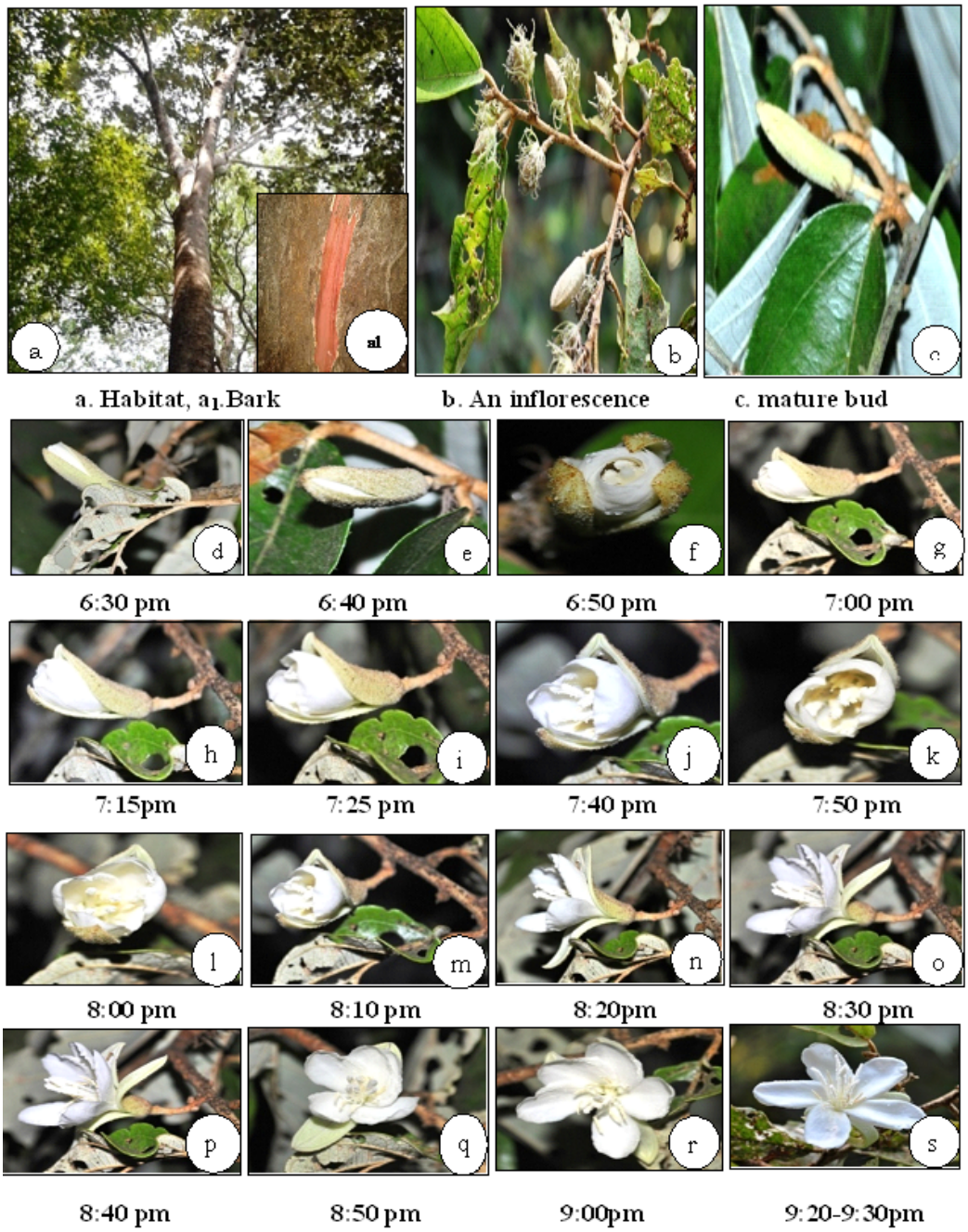

Figure 2. a- Habitat; b.-An inflorescence; c-s. Stages of floral development.

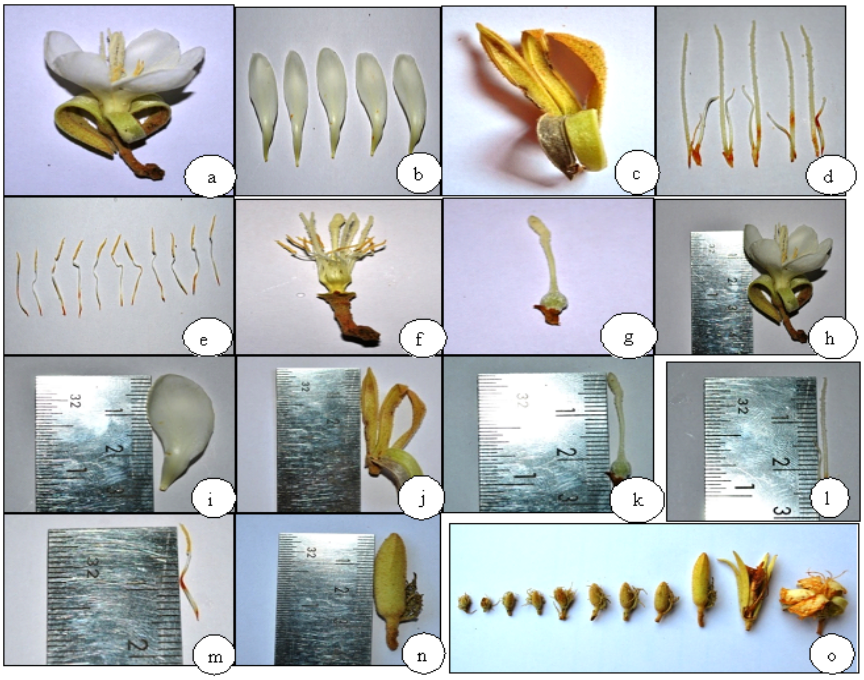

Figure 3. Floral morphological characters of *Pterospermum reticulatum*: a-Bisexual flower; b-petals; c-sepals; d-staminodes; e-anthers; f-bisexual flower parts; g-ovary exposed with pistil and stigma; h-n-floral measurements; o- Development of flower from bud initiation.

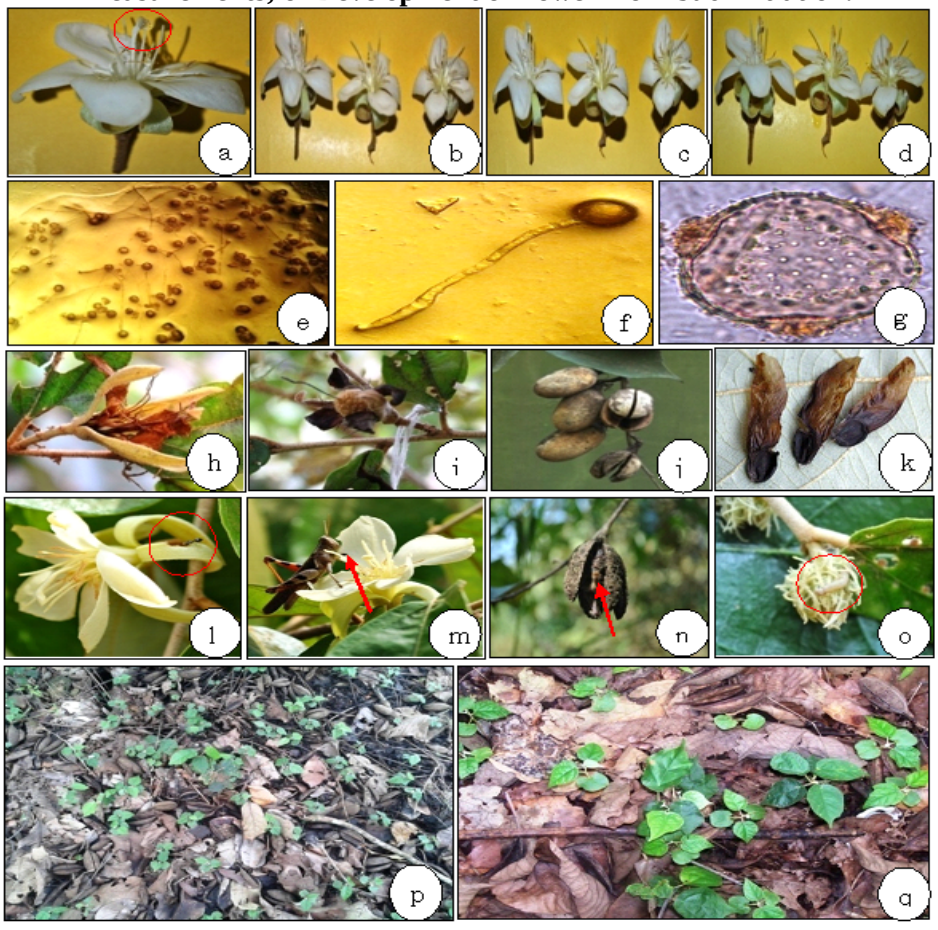

Figure 4. a-d, Stigma receptivity showing effervescence at different hours; e-Germinated pollen grains; f-pollen tube growth; g-pollen grain; h- Dried flower starts to fruit initiation; i- Immature fruit Mature fruit; j-Mature fruit; k- Winged seeds; l- Ant feeding on sepals; m- Grass hopper feeding on sepals; n- Moth; 0-Beetle larvae; p & q- Natural regeneration Growth of seedlings nearby mother tree.

Pollen production, pollen viability and germination: The pollen per anther is 2, 6233.33 and pollen per flower is 3, 93,499. TTC test was confirmed that 94.23% pollen showed maximum viability under 0.5% concentration during first day of anthesis and viability is lost in successive days. In-vitro pollen germination revealed that 96.87% pollen grains showed rich germination under gradual increase in 20 % sucrose concentration and tube length is 162.86±105.86 and viability remained for 12hrs. The pollen grains are having pores circular, spines short with blunt tips and without bulbous base or base very rudimentarily developed, inter spinal area finely baculate and negatively reticulate (Table. 1 and 3 ; Fig. 4 e, f & g) , [7,9,18].

Table 3. Pollen viability, pollen germination and pollen tube growth.

Sucrose Concentration (%)	No of pollen counted in the field	Germination of pollen percentage	Pollen tube length(µm)
5%	35	16	72.86± 45.36
10%	15	10	448.13±122.50
15%	52	49	427.50±111.39
20%	32	31	162.86±105.86

Floral predators and threats: A Beetle larvae (Fig.4 o) belonging to family Noctuidae feeds on anthers and flower buds and enters the bud via pedicel and eaten the bud parts which leads to bud abortions and in turn contributing decrease in the formation of mature flowers. Some of the ants and grass hoppers were seen feeding on the sepals, petals and flower buds (Fig. 4 l, m). It was observed that, the fruits are usually predated by moths (Fig. 4n) and is one of the factors responsible for insufficient quantities of seed production for regeneration of *Pterospermum reticulatum* in the natural habitat.

Soil analysis: *Pterospermum reticulatum* is rarely seen in coastal areas of Western Ghats in Southern India. The chemical profile indicated that soil is slightly acidic. Some of the micronutrients, their salinity such as iron, zinc and copper was high, manganese is ideal in soil samples. Macronutrients such as potassium and phosphorous is deficient and also organic matter is high. The details of chemical analysis of soils samples are shown (Table.4; Fig.5). Soil plays an important role growth and distribution of tree species [5].

Table 4. Soil sample analysis of Macro and Micronutrients in *Pterospermum reticulatum.*

	Macro nutrients					Micronutrients			
Soil sample	pH	EC	Organic matter (%)	P (k.g/acre)	K (k.g/acre)	Fe (ppm)	Mn (ppm)	Zn (ppm)	Cu (ppm)
Mean of 10 samples	6.39	0.042	1.971	4.17	13.59	17.28	3.20	1.20	0.88

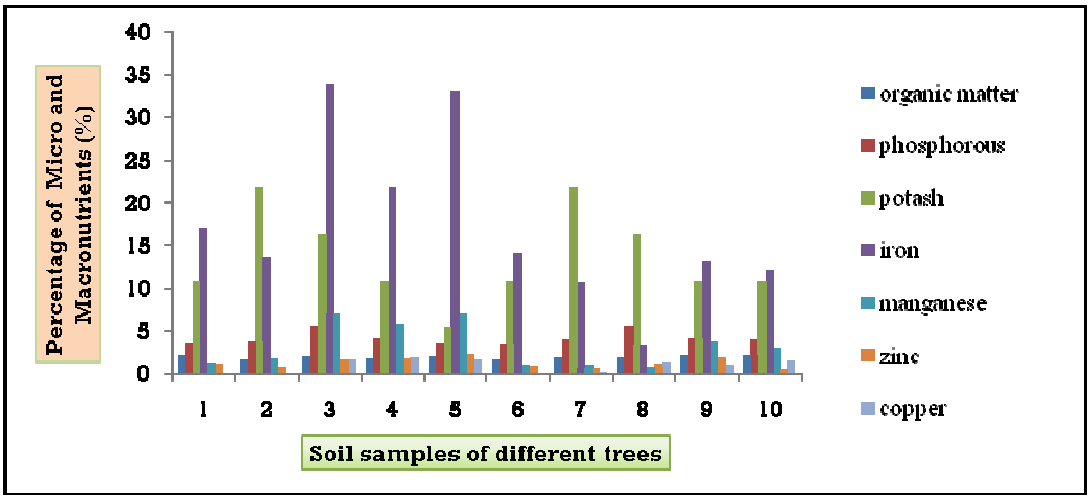

Figure 5. Macro and Micronutrients of *Pterospermum reticulatum.*

The present works provides information on floral biology, pollen germination, viability and factors responsible for decrease of population in wild. *Helicteres isora* flowering occurs for 3 days has been recorded and during anthesis, corolla turns to bluish grey and it turns red in the same evening. The next day it shows red coloration in the morning and by evening it is dark red while in the third day it begins to wither in the evening. Only on the fourth day the pollinated ones begins to fruit initiation [2]. The time of anthesis up to flower withering of *Pterospermum reticulatum* was observed for 3-4 days; the white colour flowers in the initial stage change into yellowish brown at the time withering. Flowers of *E. lushingtonii* [17] are 2-3 flowered cymes borne in the leaf axils and the flowers of *Sterculia chichi* [12] occurs as axillary paniculate clusters and staminate flowers and their position is terminal and is same in the case of *Pterospermum reticulatum* with terminal flowers and few flowers cymes. Author [14] stated that in *Pterospermum* species, most of the pollen characters are like Kurtz type but differs in having large, acutely tipped spines and prominent bulbous base. In *H. isora,* in-vitro pollen germination recorded as 50% and viability is up to 12hrs [2]. Whereas in *Pterospermum reticulatum* the pollen germination has been recorded as 96.87% in 20% Sucrose solution in Brewbaker media and remains viable even after 12hrs. Authors [17] reported that in *Eriolaena lushingtonii* the fruits mature in a short duration. Further, the fruit dehisce explosively to disseminate seeds into the air and hence the seeds are characteristically wind-dispersed for which the dry season is very effective because of low humidity at that time and the reproductive phenological events like flowering, fruiting and seed dispersal of this species occurs in dry season. In the present investigation, *Pterospermum reticulatum* flowers were observed during dry season and fruit maturation occurs during the late winter season, and also seed dispersal during dry season. The fruits dehiscence explosively liberating and winged seeds (Fig.4k) disperse by wind and hence, the plant is anemochorous [11] (Fig.4 i, j). The seeds disperse by wind in the surrounding area to several meters. The regeneration was observed in the rocky region and seedlings of *Pterospermum reticulatum* had successive regeneration capacity in Pilikula reserve forest (Fig.4 p, q) due to the presence of sufficient macro and micro nutrients in the soil. Slightly acidic pH and high organic matter in the soil may cause limitation of micronutrients.

Flower buds, flowers and fruit predation are one of the major reasons in declining of populations of *Pterospermum reticulatum.* Habitat destruction due to invasion of alien species, pathological causes threatening the life cycle is main reasons for vulnerable status of *Pterospermum reticulatum.* Locals harvest wood from this species for commercial purposes, housing and for making boats by local fisherman. They are largely used in ship making in the coastal regions. The crude extracts obtained from the leaf and stem of *P. reticulatum* used to treat bacterial and fungal diseases by the local tribes. The populations of *P. reticulatum* are being threatened because of continuous habitat destruction, anthropogenic pressures, leading to decline of populations in the wild. There is an urgent need to formulate conservation strategies including in-situ and ex-situ conservation programme to protect this valuable tree population in the Western Ghat forests of India.

ACKNOWLEDGEMENTS

The authors are thankful to the Ministry of Environment and Forests, Government of India, New Delhi for extending financial assistance under the All India coordinated research project on Reproductive biology of RET Tree species vide F No.22/10/2010-RE) and to the Chairperson of Botany Department, Bangalore University for providing basic facilities. The authors are also thanking Dr. Ramakrishna Marati, Scientific officer Pilikula Nisarga Dhama Society, Mangalore for extending help during field studies.

REFERENCES

[1] Ahmedulla, M, Nayar, M. P. 1987. Endemic Plants of the Indian region.Vol.1.Peninsular India, Botanical Survey of India, Calcutta, pp: 262.

[2] Atluri, J. B., Purnachandra Rao, S, Subba Reddi, C. 1999. Pollinator ecology of *Helicteres isora* Linn. (Sterculiaceae). Research communications. Current science, Vol. 78. No.6, 25 March.

[3] Cruden, RW.1977. Pollen- ovule ratios: a conservative indicator of breeding system in flowering plants Evolution.31, 32-46.

[4] Dafni, A, Kevan, P.G., Husband, B.C. 2005. Practical Pollination Biology Enviroquest Ltd, Canada, 590pp.

[5] Doran, J. W, Parkin, T.B. 1996. Quantitative indicators of soil quality: A minimum data set, pp.25-37.In: Doran, J.W. and A.J. Jones (eds). Method for Assessing Soil Quality. Soil Science of America, Special Publication No.49, Madison, Wisconsin, USA.

[6] Kaveriappa, Shetty, B.V. 2001. Biodiversity of the Western Ghats with special reference to Conservation of plant Diversity at Kaibab. An International Journal of Nuclear Power – Vol. 15, No. 1to 4 (2001).

[7] Kakali Biswas., Subrata Mondal, Sudhendu Mandal. 2013. Floral biology and pollination of *Solanum Sisymbrifolium* Lamk. International Journal of current Research Vol. 5, Issue, 06, pp. 1429-1433.

[8] Kearns, C. A, Inouye, D. W. 1993. Techniques for Pollination Biologists. University Press of Colorado. Niwot, Colorado, USA.

[9] Kulloli, Sreekala, S. K, Pandurangan, A.G. 2009. Floral biology of *Impatiens trichocarpa* Hook.f, (Balsaminaceae), an endemic Balsam of Western Ghats. Indian Journal of Science and Technology. Vol. 2. No .2.

[10] Marcio Silva de Souz., Giorgini Augusto Venturieri. 2010. Floral Biology of Cacauhy (*Theobroma Speciosum*-Malvaceae). Brazilian Archives of biology and technology. An international Journal, Vol.53, No.4: pp 861-872.

[11] Maury Lechon, G., Curtet, L.1998. Biogeography and evolutionary systematic of Dipterocarpaceae, pp5-44, In Appannah, S. & Turnbull, J.M. (eds). A Review of Dipterocarps: Taxonomy, Ecology and silviculture .Centre for International Forestry Research, Indonesia.

[12] Neusa Taroda, S. P., Brazil, Gibbs, P. E. 1982. Floral Biology and Breeding system of *Sterculia Chicha* St.hil (Sterculiaceae). New Phytol. 90, 735-743.

[13] Oliur Rahman, M, Abdul Hassan, M. D, Manzuruk Kadir Mea, M. D, Ahmed Mozahinurul Huq.2012. A synoptic account of the Sterculiaceae in Bangladesh. Bangladesh J. plant taxon.19 (1):63-78.

[14] Sharma, B.D. 1967. Studies of Indian pollen grains in relation to plant Taxonomy: Sterculiaceae. Proceedings of Indian National Science Academy 35:320-359.

[15] Shivanna, K. R., Rangaswamy, N. S. 1992. Pollen biology: A laboratory manual, Narosa publishing House, New Delhi.

[16] Solomon Raju, A. J, Venkata Ramana, K, Hareesh Chandra, P. 2011. Reproductive Ecology of *Shorea roxbughii* G. Don (Dipterocarpaceae), an Endangered Semi evergreen tree species of peninsular India. Jo TT Communication 3(9):2001-2070.

[17] Solomon Raju, A. J, Venkata Ramana, K., Hareesh Chandra, P. 2013. Floral Ecology and pollination in *Eriolaena Lushingtonii* (Sterculiaceae), An Endemic and Threatened Deciduous Tree species of Southern Peninsular India. Journal of threatened taxa.5 (9):4359-4367.

[18] Sreekala, A. K, Pandurangan, A.G, R. Ramasubbu, R, Kulloli, S. K. 2008. Reproductive Biology of *Impatiens Coelotropis* Fischer, a critically endangered balsam from the Southern Western Ghats. Current Science, Vol.95, No.3.

[19] Srivastava, Saxena, R. K., Gaurav Srivastava. 2012. *Pterospermum Carpon*, a new malvalean fruit from the Sindhudurg formation (Miocene) of Maharashtra, India, and its significance. Indian academy of Sciences J. Earth Syst. Sci 121, NO.1February, pp.183-193.

[20] World Conservation Monitoring Centre. 1998. *Pterospermum reticulatum*. In: IUCN 2014.IUCN Red List of Threatened Species. Version 2014.2. <www.iucnredlist.org>. Downloaded on 03 November 2014.