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The Indian Institute of Science, Bangalore, and the Department of Biotechnology, Government of India, jointly organized a workshop on rare, endangered and threatened plants in 1996 and opened up a research front and working group for species recovery of the rare, endangered and threatened (RET) plants of India. This project had its origin in this meeting and the support rendered by the Department of Biotechnology, under grant no. BT/PR7056/BCE/08/438/2006 is gratefully acknowledged.

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Mr. VS Ramachandran, Research Fellow in the project conducted the field surveys in the forest areas of Kerala. Mr. SM Sujesh, Research Fellow took up the risk of climbing the 40 m tall trees of *D. bourdillonii* and stayed inside the elephant infested dense forests of the Periyar Tiger Reserve for weeks together, in order to study the reproductive biology of *H. bourdillonii*. The work would not have been accomplished without their dedication and determination.

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Authors

Summary

In 2005, a National Chapter on species recovery of rare, endangered and threatened plants of the country was initiated by the Department of Biotechnology (DBT), Government of India. Species recovery of two threatened tree species in Kerala was taken up jointly by the Kerala Forest Research Institute, Peechi and the Botanical Survey of India, Calcutta.

The species investigated were *Dipterocarpus bourdillonii* (Fam. Dipterocarpaceae) and *Humboldtia bourdillonii* (Fam. Fabaceae). *D. bourdillonii* is a lofty ripicole evergreen tree which suffered from selective logging in the past. Its distribution is discontinuous with a few disjunct patches along the Western Ghats. Earlier seed germination attempts were not successful. The second species, *Humboldtia bourdillonii* is a medium sized tree originally recorded form the Peermade Plateau of Southern Kerala. After the original collections made by Bourdillon in 1894, the species was relocated recently, nearly after 100 years, in the Periyar Tiger Reserve. Thus the objectives of the study were: (i) To generate reliable information on distribution, population structure, and reproductive biology including reproductive constraints, (ii) To explore for evolving macro or micro propagation techniques for the species, (iii) To explore the possibility of enhancing the natural population of the two species by appropriate translocation programmes, and (iv) To evaluate the post-translocation survival and growth of the planted stocks.

Population ecological, reproductive biological, propagation and recovery aspects were attempted of the two species. Distribution of the species was studied through intense field explorations. Phytosociological studies were conducted in releves of 0.1 ha size and tree regeneration enumerated in sub-samples. Reproductive phenology was studied by periodic field visits and observations on different phenophases. Reproductive biological studies included floral biology, study of pollinators, pollen tube growth, controlled self and cross pollinations, and the performance of the seeds originating from these experiments. Propagation of the species from seeds, through air layering, rooting of branch cuttings, and micro-propagation were attempted. The propagated ramets were augment planted in *in-situ* as well as *ex-situ* sites and their survival and growth performance evaluated at 6-moth intervals for a period of two and a half years.

Dipterocarpus bourdillonii is found in forests north of Palghat Gap, where it grows along with species such as *Vateria indica*, *Knema attenuata*, *Polyalthia fragans*, *Syzygium* spp., etc., while south of Palghat Gap, the dominant associates were *Turpinia malabarica*, *Cinnamomum malabaricum*, *Drypetes oblongifolia*, *Baccaurea courtallensis*, *etc.* The total population of *D. bourdillonii* in Kerala is only $\pm$ 240 trees ≥ 30cm gbh. The larger populations of the species are located at Chavachi (Aralam,

Kannur, 52 trees), Pinavurkudi (Urulamthanni, 51 trees) and at Pampa (49 trees). In other places, the species is represented with a few individuals. Two larger populations (Moonnumukku and Pamba) had only larger girth classes, ≥ 60cm gbh, and were deficient of young ones; against a subsample of 180 trees (>= 10cm dbh), there were only 60-70 seedlings. Absence of young ones to replace their older individuals is characteristic of a species moving towards extinction. The small population size, which is a result of low efficiency in reproduction/ establishment is also characteristic of a species which is on its way towards extinction. The non-viable embryo in the mature fruits tells us this story explicit.

The flowers are pollinated by two species of *Apis, A. indica* and *A. dorstata*. Fruits derived from controlled self pollinated flowers failed to germinate confirming self-incompatibility; incompatibility operates in the post-zygotic phase. On the other hand, in control cross pollinated flowers 77% progressed to fruits/seeds and 35% of these germinated. About 32% of the fruit population was found devoured by weevils, which ovipose inside the ovary and the larvae feed on the embryo. Dissection of near-mature fruits showed that ca. 61% had decayed embryos, independent of insect infestation.

Many seed germination trials resulted in total failure. Air layering using different treatments of IBA (400, 600, 3000 and 5000 ppm) was not successful. Cent percent rooting of stem cuttings was obtained when treated with IBA at 6000 and 8000 ppm and ca. 60 % survived after transfer to nursery. Nodal explants with the hairy covering removed, when inoculated into Woody Plant Media supplemented with Benzyl Adenine and Kinetin, the explants responded positively. Sprouting was obtained but growth was extremely slow and no multiple shoots obtained. Immature embryos when cultured on MS media supplemented with auxins (NAA, 2,4-D, Picloram and Dicamba) callus development occurred but failed to differentiate. Embryonic axes cultured on BAP containing medium sprouted but failed to produce multiple shoots. The species appears to be recalcitrant to *in vitro* culture as is the case with a few other Dipterocarps.

A total of 1,200 seedlings/ramets of *D. bourdillonii* were augment planted in eight *in-situ* sites and 150 seedlings planted in *ex-situ* sites. In general, seedlings planted *ex situ* have scored maximum survival compared to that at *in-situ.* Highest survival obtained was 54% (Meenvallam) but low rates of 12-15% were also recorded. Low survival owes mainly to mortality factors of which pest infestation, suppression by weed growth, trampling by wildlife, grazing by wildlife, etc, are the dominant. The three *ex-situ* sites which had a garden environment, the survival rate was high, 60-78%. A maximum height growth of 238 cm was recorded for the species in the 2½ yr period.

Humboldtia bourdillonii is a medium sized rare tree seen distributed in the evergreen forests of the Peermade Ghat at Arjunan Kotta and Poonkavanam area of Pampa valley. The dominant associates of the species are *Vateria indica, Mesua ferrea* and *Palaquium ellipticum*. Field survey provides a picture of patchy distribution; seven discrete patches ranging between 0.1-2.0 ha were located. The area of occurrence was found to be c. 2 km^2 and the area of occupancy 0.07 km^2. The estimated total population is c. 1,100 trees (≥30 cm gbh). The species is found well represented in all life stages.

The flowers open gradually from late morning till early evening and are anemophilous but cross pollinated. Natural fruit set is quite low (17%). Controlled self pollinated flowers did not progress to fruit set whereas c. 40% of the cross pollinated flowers produced fruits inferring self-sterility. Jumping thrips multiply and colonize the young infructescence and suck the sap of young fruits causing to lose a sizable portion of fruit populations. Infestation by squirrels also contributes to immature fruit-fall. Weevils penetrate the fruit wall and lay eggs in the cotyledons of the young embryo. The larvae grow at the expense of the cotyledons and the adults emerge out as the seeds get dispersed. Nearly 13% of the mature seeds are lost this way.

In a germination trial conducted the percentage of germination obtained was 37.5%; in peak periods of germination (May-June) germination is still low (ca. 28%). Branch cuttings treated with 400-800 ppm of IBA solutions rooted. Nodal explants from trees when cultured on Woody Plant medium with various hormones (Benzyl Adenine, Kinetin, and Benzyl Adenine/Kinetin), sprouting response was extremely poor and no shoot growth succeeded. Explants from cotyledonary nodes when cultured on media containing BAP sprouted but again multiplication of shoots could not be achieved. By inoculating cotyledonary explants on a wide range of auxin concentrations, low callus development was obtained but had the same fate. Young embryos when cultured in media containing hormones such as NAA, 2,4-D, IBA and Picloram, scant callusing was obtained but the same could be maintained in subcultures.

A total of 300 seedlings were planted in three *in-situ* sites and 150 seedlings in three *ex-situ* sites. As with *D. bourdillonii*, the observed survival rates of planted seedlings of *H. bourdillonii* were also higher in *ex-situ* sites, in comparison to *in-situ* sites; 64-100% survival was obtained after two years in the protective environment of the *ex-situ* sites. The low rates of survival in *in-situ* sites (20-50%) were due to disturbances such as grazing, wildlife and weed growth. A maximum height growth of 214 cm was recorded for the species in the 2-yr period.

1. Introduction

Plants and animals having cosmopolitan distribution constitute only a very small proportion of the world's biota. The larger proportion, however, remains endemic being restricted by geographic bounds of differing scales. Depending up on rarity (in terms of the population size) and a posited higher risk for extinction, the endemics constitute various categories such as critically endangered (CR), endangered (EN), vulnerable (VU), near threatened (NT), etc. Compared to the temperate, tropical forests abound in endemic biota falling in the risk-some categories mentioned; oceans, islands and mountains are also estimated to contain more concentration of endemics and rare biota. These are the segments of biodiversity that could be lost for the mankind forever, where conservation efforts and species recovery programmes are needed urgently.

India is a biodiversity-rich country with as many as 17,500 flowering plants, 61% of which are endemics (Ramesh and Pascal, 1991). Calling up on the urgent need for the conservation of the rarer segments of the nation's biodiversity, a workshop was organized jointly by the Department of Biotechnology (DBT), Government of India, and University of Agricultural Sciences (UAS), Bangalore, at Bannerghatta National Park (Banglaore) on 19th and 20th July 2005 (Ganeshaiah, 2005). This meeting, attended by a number of organizations doing active research on conservation of RET species (rare, endangered and threatened species), chalked out a National Programme for Species Recovery, where initially a few highly endangered and threatened plants were given priority.

Scientists from KFRI, who attended the Bennerghatta Workshop, proposed investigating the causes of rarity and recovery of two threatened trees of the Southern Western Ghats, *viz.*, *Dipterocarpus bourdillonii* (Dipterocarpaceae) and *Humboldtia bourdillonii* (Leguminosae). The programme was supported by DBT (BT/PR7056/BCE/08/438/2006).

The above scheme was conceived as a collaborative programme between four institutions:

a. The Kerala Forest Research Institute, Peechi
b. The Botanical Survey of India, Calcutta
c. College of Forestry, University of Agricultural Sciences (CF-UAS), Ponnampet, Coorg
d. The Kerala Forest Department (KFD)

A segment of the programme dealing with *Dipterocarpus bourdillonii* in Karnataka was awarded to the College of Forestry (CF-UAS), Ponnampet, through a separate grant by DBT. The Kerala Forest Department being

the custodian of the forest areas where the study areas belonged was more as a facilitator. The scientific expertise of the Botanical Survey of India served to provide the taxonomic expertise required in the context. This report therefore is a summary of the work done at KFRI and the results emerged out of it.

2. Scope of study and species studied

The evergreen forests that cloth the Western Ghats show high degree of endemism and out of 490 trees found inhabiting the low and medium elevation evergreen forests, 308 are endemic (Ramesh and Pascal, 1991, 1997). Looking at the floral elements of Kerala, which retains a larger segment of the Western Ghats, Sasidharan (2003) enlists 497 red-listed species, which includes 151 tree species (Sasidharan, 2003).

The red-listed tree species according to their ecological status are: Extinct (2 spp), Low risk/Near threatened (22), Critically endangered (26), Vulnerable (38), Endangered (63). The order of their representation in various families is given in Table 1 (Sasidharan, 2003). All these species are candidates for effective recovery programmes.

Table 1. Red-listed trees of Kerala across the families*.

Sl. No.	*Families*	*Species*	*Sl. No.*	*Families*	*Species*
1.	Myrtaceae	20	8.	Rubiaceae	8
2.	Lauraceae	16	9.	Symplocaceae	7
3.	Leguminosae	11	10.	Meliaceae	7
4.	Dipterocarpaceae	11	11.	Flacourtiaceae	6
5.	Euphorbiaceae	10	12.	Clusiaceae	5
6.	Anacardiaceae	10	13.	Sapotaceae	4
7.	Annonaceae	8			

* After Sasidharan (2003).

Dipterocarpus bourdillonii (Dipterocarpaceae) and *Humboldtia bourdillonii* (Fabaceae) were the two tree species selected for the study and recovery.

1. ***Dipterocarpus bourdillonii*** Brandis

Within the tree family Dipterocarpaceae the genus *Dipterocarpus* is well known for the timber value of its species. The Peninsular Indian species of the genus are, *Dipterocarpus bourdillonii* Brandis and *D. indicus* Bedd., both being endemic to the Western Ghats. Both are lofty evergreen trees of primary wet evergreen forests growing to height of 40m and attaining a bole diameter of 1-1.5 meter at breast height (Fig. 1). The timber of the

species has long been exploited in the past, as softwood for making plywood.

Figure 1. Two pictures of the ripicole species, *Dipterocarpus bourdillonii.*

While *Dipterocarpus indicus* is widely distributed all through the Western Ghats from Shimoga southwards, *D. bourdillonii* is largely confined to Southern Western Ghats. Although the area of distribution of the species extends from Coorg (Karnataka) southwards, as Ramesh & Pascal (1991) pointed out, it is broken into a few disjunct patches, especially towards the northern half of the distributional area.

Ramesh & Pascal (1991) assigned a '*threatened*' status to *D. bourdillonii* and Ramesh & Pascal (1997) provided a distribution map of the species based on specimens preserved in herbaria. These documentations confirm the discontinuous distribution. Sasidharan (2003) assigned a '*Low Risk/ near threatened'* status to the species.

D. bourdillonii (Dipterocarpaceae) is a large tree of primary forests at low elevations (200-400 m), distributed in Kerala and Karnataka. It is a ripicole species found restricted to areas adjoining river courses only in interior forests. Outside the conventional floristic documentations, no population details of the species are available. An estimate of its population size by the investigators (as part of a different study) was below 500 mature trees and therefore qualified a critically endangered status, mainly due to habitat loss and apparent intrinsic reproductive problems.

Germination studies by the Kerala Forest Department have repeatedly met with failure. Examination of the dissected mature seeds (fruits) shows only dead embryos.

Whether fragmentation of population (See Fig. 2) has contributed to genetic incompatibilities and reproductive failure remains unclear. In programmes aiming to conserve the species, it is worthwhile to explore and understand the role of past forest management (selective logging) and reproductive inefficiency in diminishing the populations. As the species does not produce viable fruits, there is scope for experimenting for potential macro and/or micro propagation (tissue culture) techniques.

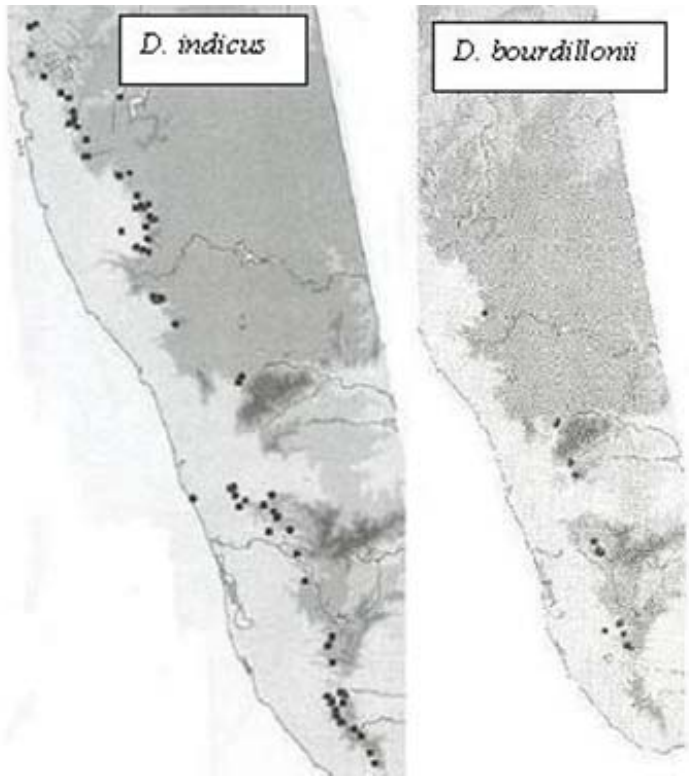

Figure 2. Distribution of *Dipterocarpus indicus* and *D. bourdillonii* (After Ramesh and Pascal, 1997).

The small population of existing juveniles in the wild indicates that a small percentage of the fruits/seeds do contain viable embryo. The tree being a potential plantation species development of mass multiplication techniques is desirable.

The species is of timber value and a flagship species contributing to the forest composition and architecture of the low elevational forest type: *Dipterocarpus bourdillonii – D. indicus – Anacolosa densiflora* type, described by Pascal (1988). The future genetic value, the flagship nature, the discontinuous distribution, and the 'Critically Endangered' status of the species underline the need for conservation of the species through recovery programmes.

2. ***Humboldtia bourdillonii*** Prain (Fabaceae)

Leguminosae (also known as Fabaceae), the bean family is economically very important, with a global species count of 19,327. The family is represented in India by c. 340 species (Sanjappa, 1991). *Humboldtia* is one genus coming under the subfamily Caesalpinioideae having six species; all of them are confined to Southern Western Ghats, except one extending to Sri Lanka (Sanjappa, 1986; Sasidharan, 2004).

Humboldtia bourdillonii Prain is a medium sized tree originally recorded form the Peermade Plateau of Southern Kerala. It is a small tree found in the evergreen forests distributed in Southern Kerala. Growing to a height of 25m, the tree occupies the sub-canopy (Fig. 3).

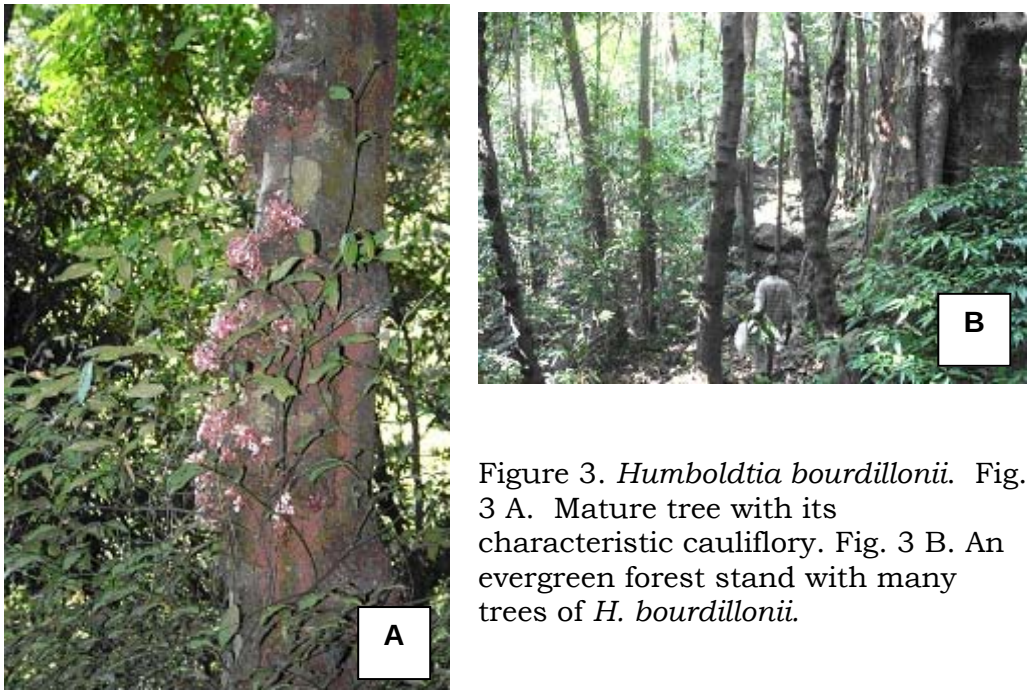

Figure 3. *Humboldtia bourdillonii*. Fig. 3 A. Mature tree with its characteristic cauliflory. Fig. 3 B. An evergreen forest stand with many trees of *H. bourdillonii.*

After the original collections made from Peermade by Bourdillon in 1894, there has been no subsequent report of the species (Sanjappa, 1991). Nearly after 100 years, Sasidharan (1998a, 1998b) relocated it from the Periyar Tiger Reserve (Peermade Plateau) and was again reported from the same area subsequently by Augustine (2002). Sasidharan (2003, 2004) ranked the species as 'endangered'. However, because of its restricted distribution and paucity of representation in regional herbaria, there is every possibility the species is 'Critically endangered'. Apart from this fragmentary information no other details of the species are available including its population size and specific details of distribution.

H. bourdillonii is a medium sized tree seen distributed in the Peermade Ghat of Southern Kerala (Gamble, 1915-1936). The tree grows to a height

of 15 m and diameter of c. 35 cm at breast height. Like *Dipterocarpus bourdillonii*, *Humboldtia bourdillonii* is also believed to be a ripicole species found along the streamlets and watercourses of evergreen forests at about 900 m (Balasubramanyan *et al.*, 1985).

Except for some preliminary investigations on macro-propagation of *H. bourdillonii* (Jose *et al.,* 2011), practically nothing is known about its population size and why the species is rare and restricted in distribution and therefore it merits species recovery.

3. Objectives of the study

- To understand the cause of rarity of *Dipterocarpus bourdillonii* and *Humboldtia bourdillonii*, two critically endangered endemic trees of Western Ghats.
- To study the distribution, population structure, and reproductive biology with special emphasis on reproductive constraints, if any, such that the output from these studies is available for framing sound species conservation programmes.
- To evolve appropriate macro or micro propagation techniques for the species so that ample planting stocks of the species required for the desired species recovery programmes are available.
- To explore the possibility of enhancing the natural population of the two species by appropriate translocation programmes.
- To evaluate the post-translocation survival and growth of the planted stocks.

4. Materials and Methods

The programme had a multi-disciplinary approach. Population ecological, reproductive biological, propagation and recovery aspects were attempted in the project. The study was conducted during the period, 2006-2011.

Population ecological studies

DIVA-GIS and GARP are GIS-based softwares capable of identifying the probable areas of distribution based on climate regimes and pixel matching. Using these softwares and by providing reference points of known distribution, the probable areas of distribution of both the species were generated. The result was not promising. This probably was due to the very restricted distribution of the species, or the highly fragmented populations. Thus, conventional field surveys were conducted based on: a. Previous collection records, and b. Prospective areas identifiable, based on the ecological specificities of the species.

Field surveys: As *Humboldtia bourdillonii* is known only from the High Ranges of Kerala, the field surveys were restricted to that region.

Dipterocarpus bourdillonii, being a ripicole species, the field surveys were restricted to river courses. As the survey of this species was partly covered in an earlier research project, in the present study, only Central and Northern Kerala were surveyed. The areas explored included Waynad and Aralam Wildlife Sanctuaries. With the help of Dr. CG Kushalappa, the Collaborator from CF-UAS, explorations for the species was also conducted in the forests of Subramanya (Karnataka) and adjacent areas.

Exploratory field trips were conducted in the forests of Kerala to locate the two trees. In conducting the field surveys, we adopted the following approach. Geographic information available in herbarium specimens were extracted from three sources: a. Madras Herbarium (MH), Coimbatore, b. Herbarium of the Kerala Forest Research Institute (KFRI), and c. Forest Herbarium (IFGTB). Field knowledge of three categories of people was also utilized in planning the field trips: a. Scientists who collected the species earlier, b. Forest dwellers, and c. People living nearer the forest areas.

Phytosociological studies: Populations of the two species were studied in releves (samples plots) of 0.1 ha (50m x 20m) size. Whenever, the distribution area was found to be small, total enumeration was attempted. All trees having girth range (GR) ≥10cm in the study plots were identified, enumerated and recorded. Tree regeneration with GR<10cm were enumerated only in two sub-plots of 25m^2 (5m x 5m) in each releve of 0.1 ha.

Computations of general phytosociological parameters (viz, density, frequency and cover; Muller-Dombois and Ellenberg, 1974) were performed to characterize the natural vegetation that hosted the candidate species.

Density: Number of individuals of a species per unit area gives its density (d). This is usually computed as trees per hectare (tr ha^{-1}).

Frequency: The chance of finding a species in a particular area in a particular trial sample is called its frequency and is expressed as the number of quadrats in which a species is found per total number of quadrats studies.

Cover (Basal area): Cover is usually the area covered by crown or shoot area, or the stem. For trees and shrubs, the area occupied by the stem is taken as the cover and is known as the basal area. Basal area=πr^2; r = gbh/2π.

The Importance Value (or IVI): Important Value Index (IVI) is defined as the sum of Relative Density (RD), Relative Frequency (RF) and Relative Basal Area (RBA, Muller-Dombois and Ellenberg, 1974). This expresses the relative importance of the species in the community. Thus, IVI = RD + RF + RBA, where,

RD = (Density of the species) / (Density of the stand) x 100
RF = (Frequency of the species) / ∑ (frequency of all the species) x 100
RBA = (Basal area of the species) / (Basal area of all species) x 100

The species importance values (Species-IVIs) computed with respect to the stand's total IVI (300) are reduced to percentile values (Species IVI/3; Narayanan & Swarupanandan, 1996) for easy comparison.

Phenological studies: Reproductive phenology of the species was monitored through repeated observations of recognizable phenophases for *Dipterocarpus bourdillonii* at Urulamthanni and Pamba. Phenological observations of *Humboldtia bourdillonii* were conducted at Arjunankotta.

Reproductive biological studies

Reproductive biological studies included field studies as well as laboratory investigations.

Reproductive biology of Dipterocarpus bourdillonii

The field components of the investigations in respect of *D. bourdillonii* were conducted in the Periyar Tiger Reserve at Urulamthanni and Pinavoorkudi area belonging to Urulamthanni, Thattekkad and

Neriamangalam Forest Ranges of Kothamangalam Forest Division, between 2007 and 2009.

As the trees are lofty (35-40 m tall) and devoid of branches on the tall clear bole, a mechanical climbing device was devised with the assistance of a local engineering college (Fig. 4 A), which however did not work. Hence a ladder system made of bamboo and reeds and utilizing the technical expertise of local tribes was utilized for climbing trees (Fig. 4 B).

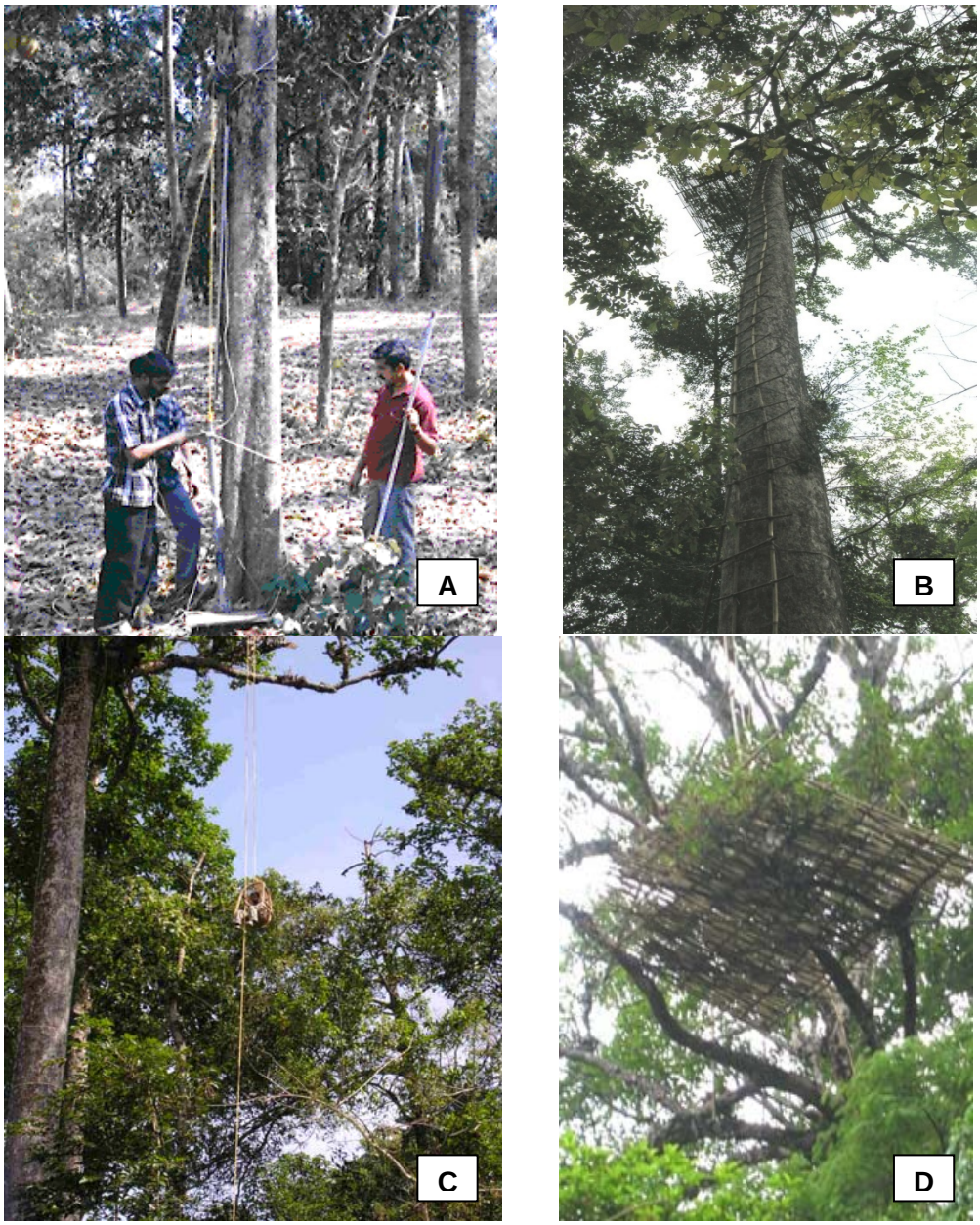

Fig. 4. Managing tree climbing for reproductive biological studies. Fig. 4 A. A mechanical device developed for climbing clear-bole trees, but which failed to work. Fig. 4 B. The stair-climbing system made of reeds and utilizing the technical expertise of local tribes. Fig. 4 C. The pully system for climbing the tall trees of *Dipterocarpus bourdillonii*. Fig. 4 D. Canopy platform.

At a later date, a pully and a cane swing (basket) mechanism was also utilized for climbing the trees (Fig. 4 C). Platforms for moving across the canopy and accessing the flowers for breeding experiments were also made using bamboos and reeds (Fig. 4 D). Altogether, six ladder systems and two canopy platforms were built for the study.

Reproductive phenology and floral biology: Trees were monitored for: a. dormant buds, b. flushing, c. inflorescence initiation, d. flowers, e. flower opening, f. fruiting, g. fruit maturity and diaspore (fruit or seed) dispersal. Inflorescences and flowers selected for the studies were labelled separately and were observed at regular intervals. Details on the morphology of the reproductive organs were also collected in detail. Physical changes during flower opening, anther dehiscence, dehiscence of floral whorls as well as flower size parameters were also observed and recorded.

Insect visitors: Floral visits by insects and their time and frequency of visits were recorded. Observations on insects visiting the flowers and fruits were made round the clock (day and night) and their involvement with floral structures during the visits noted. During the day, insects were collected using insect-nets and at night using lights. The nature and extent of insect attack on flowers and fruits were subjected to study.

Role of insects on pollination: In order to find out the role of insects in pollination and seed set, pollen loads on stigmas of emasculated and non-emasculated flowers were assessed. Two lots of open-flowers were tag-marked and covered with nylon net; one lot contained emasculated flowers and the other non-emasculated. The following day evening, 20 stigmas were excised from each lot, stained with aniline blue and examined under the microscope.

Pollen viability: Pollen viability was tested in the field for a duration of 48 hrs at a 2hr-interval. Viability was tested using tetrazolium chloride (0.5% of 2,3,5-triphenyl tetrazolium chloride in 5, 10, 15 and 20% of sucrose solution). After 30 minutes of staining, pollen grains were examined under a light microscope and viability determined based on staining level.

Stigma receptivity: Stigma receptivity was determined by observing pollen germination on stigmatic surface after controlled pollinations carried out at intervals. Flowers were emasculated in mature flower buds due for opening the next day and bagged. Artificial pollinations were performed at 4-hr interval. Three pistil replicas were kept for experiment. The cleared pistils were then stained using a modified aniline blue staining technique (Shivanna and Rangaswamy, 1992). The specimens were observed under fluorescence microscope for pollen germination.

Study of mating system and development of fruits: Artificial pollination studies were conducted to get an idea of the type of mating system. The

pollination studies included pollen viability, stigma receptivity, pollen tube development, natural seed set ratio, seed setting through apomixes and self- and cross- pollinations. All the inflorescences and flowers used for pollination studies were labelled.

- *Natural pollination:* Experiments were conducted to assess the role of insect foragers in pollination and seed set. The study included three sets of inflorescences: i. Covered with nylon nets, so as to prevent entry of insects. ii. Kept open for natural pollination (ie, without emasculation). iii. Kept open for natural pollination with emasculation. Percentage of seed set was monitored in two treatments: Flowers sprayed with insecticide and fungicide, and control devoid of fungicide application.

- *Apomixis:* 40 mature flower buds due for opening the next day were *e*masculated, tag-marked and bagged so as to monitor apomictic embryo development.

- *Controlled pollinations:* Artificial cross pollinations (in 48 flowers) and self pollinations (in 91 flowers) were conducted at dusk after anthesis. Flowers were emasculated 24 hours prior to anthesis, as anthers dehisce and liberate pollen grains in small quantities 10 hours prior to anthesis. Fresh pollen grains were collected immediately before pollination in Petri plates kept below and tapping the flower. The sticky pollen grains were transferred to the receptive stigma using a fine needle. The experiment involved two treatments: i. The stigmas of the flowers were artificially self-pollinated by dusting with pollen of the same tree at peak pollination time. ii. The stigmas of the flowers were artificially cross-pollinated using pollen from other trees.

When fruits developed from each of the above experiments were one month old, the fruits were tagged and tied to the branches so that the fruits were available for further experimentation.

Pollen tube growth: Immediately after anthesis, self and cross pollinations were carried out separately for analyzing the pollen tube growth. Three pistils each were collected at 2-hr intervals, starting from 2 hrs to 48 hrs after pollination. Pistils were collected and fixed in acetic alcohol and then stored in 70% alcohol. Collected specimens were stained by aniline blue and examined under fluorescent microscope.

Embryo development: Embryo development was monitored from young flowers to mature fruits using longitudinal and transverse hand sections of the flowers/fruits.

Embryo decay studies: Fruits were cut open at various stages of development for identifying for recording live/dead/decaying embryo and occurrence of insect damage and fungal attack. Fruits derived from controlled pollination experiments and naturally fallen fruits not derived from experimental studies were also subjected to similar studies. Many of the fruits were seen decayed even in the absence of insect attack. To analyze the cause of decay, embryos from such fruits were inoculated in microbiological media (Potato Dextrose Media) to study the influence of fungal attack.

Insect infestation: Insect infestation on flowers and fruits were monitored by cutting open flowers and fruits periodically. These insects were collected and preserved in alcohol. Efforts were made to identify the insects.

Seed germination: Fruits derived from different experiments (mentioned above) were sown in trays filled with garden soil enriched with soil from natural habitats of the respective species. 513 fruits collected from five trees and derived from open pollinated flowers were also subjected to germination trial. The non germinated fruits were examined for insect attack.

Reproductive biology of Humboldtia bourdillonii

The ground plan of reproductive biological studies of *H. bourdillonii* followed that of *D. bourdillonii*. In this section, only deviations from that of *D. bourdillonii* alone are provided.

Reproductive biological studies were carried out in the evergreen forests of Arjunankotta (450-800m asl) in Periyar Tiger Reserve (PTR) (Fig. 5).

Figs. 5. Reproductive biological studies in the natural habitat of *Humboldtia bourdillonii*. Fig. 5 A. Performing the breeding experiments. Fig. 5 B. Bagged flowers after self and cross pollination experiments.

Reproductive phenological studies: Trees of six patches of the population at Arjunankotta were observed for phenological data collection. Observations were made at an interval of one month between December 2006 and March 2009.

Control pollination experiments, mating system, fruit development: Since *H. bourdillonii* proved self incompatible, delayed self pollinations and bud pollinations were tried. Bud pollinations were performed in mature flower buds 12 hours prior to flower opening and delayed pollinations were carried out in flowers 25 hours after flower opening. In the first case pollen grains collected from flowers of the same tree opened on the previous day was used and in the second instance fresh pollen grains collected from newly opened flowers of the same tree were used for pollination.

Natural fruit set: Percentage of natural fruit set was evaluated. For estimating the proportion of natural fruit set, a total of 109 randomly tag-marked flowers from nine trees were used.

Propagation studies

Conventional seed germination studies, air layering, rooting of stem cuttings through application of hormones, multiplication of nodal explants and immature embryos in culture media were experimented with.

Propagation studies of Dipterocarpus bourdillonii

Propagation through germination of seeds: Two lots of fruits of *Dipterocarpus bourdillonii* were collected from Urulamthanni (Munnar FD) and Pamba (PTR) and germination trials conducted in the Central Nursery at KFRI, Peechi.

Air layering of D. bourdillonii: Air layering was attempted for *D. bourdillonii* in the field on the accessible lower branches of small trees. Vermiculite was used as the medium and different treatments of IBA (400, 600, 3000 and 5000 ppm) were given.

Rooting of stem cuttings in D. bourdillonii: 5-8 cm thick cuttings were planted in soil and placed under mist to induce sprouts for micropropagation. Cuttings of approximately 30 cm were treated with varying concentrations of IBA to induce rooting. The cuttings were bagged in soil and sand mix (2:1 ratio) and kept in the mist (Fig. 7) for rooting. The concentrations of the hormone applied were: 400 ppm, 600 ppm, 6000 ppm, 8000 ppm, 9000 ppm, 1000 ppm. The experiments were conducted during the months of August-November 2007.

Shoot multiplication of axillary buds in D. bourdillonii: Nodal explants were inoculated into Woody Plant Media supplemented with Benzyl Adenine (1.5 ppm, 3 ppm and 6 ppm), Kinetin (3 ppm) and Benzyl Adenine/Kinetin (3 ppm/3 ppm). The outer hairy covering and the size of the nodal explants were reduced to bring down contamination. Sterile explants showing expansion of the terminal bud and signs of sprouting were subcultured on BAP containing media (liquid with filter paper support). The experiments were conducted during August-November 2007. Despite the application of antibiotics, controlling the contamination was found difficult (Table 2).

In vitro *regeneration from immature embryos in D. bourdillonii:* Embryos from mature fruits were used as explants for *in vitro* regeneration through organogenesis. As the majority of the seeds were infested with insect larvae, only a small percentage was found suitable for *in-vitro* studies. This posed a major constraint for conducting the experiments. Hormones and their concentrations applied in *in vitro* culture are given in Table 3.

Table 2. Sterilization treatment given to nodal explants of *D. bourdillonii.*

No.	*Treatments*
1.	Cefotaxime (200 ppm) + Ampicillin (200 ppm) + 0.1 % $HgCl_2$
2.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 0.1 % $HgCl_2$
3.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 0.1 % $HgCl_2$ + PPM[3]
4.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1% Bavistin (105 mins) + 0.1 % $HgCl_2$ (15 mins)
5.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1 % Bavistin; Exalin (0.1%) + $HgCl_2$

Propagation studies of Humboldtia bourdillonii

Propagation through germination of seeds: Seed lots of *Humboldtia bourdillonii* collected from Periyar Tiger Reserve (PTR) have been germinated (Fig. 9). The seedlings were potted in poly bags and maintained in the nursery for transplantation.

Table 3. Hormones and concentrations used in the *in vitro* culturing of embryos of *D. bourdillonii*.

Sl. No	*Hormones*	*Concen-trations*	*Dates*	*Sl. No.*	*Hormones*	*Concen-trations*	*Dates*
1.	2,4-D	3 ppm	Feb. 2007	5.	NAA	1 ppm	Feb. 2007
2.	2,4-D + BAP	3 ppm + 0.5 ppm	Feb. 2007	6.	NAA	3 ppm	Feb. 2007
3.	2,4-D	3 ppm	Mar. 2008	7.	NAA	3 ppm	Mar. 2008
4.	2,4-D	5 ppm	Mar. 2008	8.	NAA	5 ppm	Mar. 2008
				9.	Picloram	3 ppm	Feb. 2007

Rooting of stem cuttings in H. bourdillonii: Branch cuttings collected from Periyar Tiger Reserve in June, 2007 and March, 2008 were treated with varying concentrations of IBA for 10 minutes, bagged in soil/sand mix (1:1 ratio) and placed in the mist for rooting. The concentrations of IBA used were 400, 600, 800, 6000 and 8000 ppm and each treatment contained 10 stem cuttings.

Shoot multiplication of axillary buds in H. bourdillonii: Nodal explants collected from living trees as well as sprouts from branch cuttings maintained in the mist were placed in Woody Plant media with different hormones at varying concentrations. The hormones used were Benzyl Adenine (I.5 ppm, 3 ppm and 6 ppm), Kinetin (3 ppm) and Benzyl Adenine + Kinetin (3 ppm each). Various culture sterilization treatments were also applied to the cultures (Table 4).

Nodal explants were also cultured on Murashige and Skoog's media. Immature as well as mature seeds were used as source of explants. The

excised cotyledonary nodes were cultured on media containing BAP (5 mg/l).

Table 4. Sterilization treatments applied to nodal explants of *H. bourdillonii*.

No.	*Treatments*
1.	Cefotaxime (100ppm) + Ampicillin (100 ppm) + 0.1 % $HgCl_2$
2.	Cefotaxime (200 ppm) + Ampicillin (200 ppm) + 0.1 % $HgCl_2$
3.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 0.1 % $HgCl_2$
4.	70% alcohol (10 seconds); Cefotaxime (200 ppm) + Tetra-cycline (200 ppm) + 1% Bavistin (60 mins) + 0.1 % $HgCl_2$ (10 mins)
5.	70% alcohol (20 seconds); Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1% Bavistin + 0.1% $HgCl_2$ (10 mins)

In vitro *regeneration of immature embryos of H. bourdillonii:* Seeds at all stages of development were used as explants. Surface sterilization was done with 0.1 % $HgCl_2$ mixed with 0.2 % Extran (surfactant). Embryos were removed and treated with 1 % PVP before inoculation. Different hormones in varying concentrations were applied in the culture media (Table 5).

Table 5. Hormones and their concentrations applied *in vitro* regeneration of embryos of *H. bourdillonii*

No.	*Hormo-nes*	*Concen-trations*	*Dates*	*No.*	*Hormo-nes*	*Concen-tration*	*Dates*
1.	NAA	1 ppm	Jan. 2007	6.	2,4-D	1 ppm	Jan. 2007
2.	NAA	3 ppm	Jan. 2007	7.	2,4-D	3 ppm	Jan. 2007, Mar. 2008
3.	NAA	3 ppm	Mar. 2008	8.	2,4-D	5 ppm	Mar. 2008
4.	NAA	5 ppm	Mar. 2008	9.	2,4-D + BAP	3 ppm	Jan. 2007
5.	IBA	3 ppm	Jan. 2007	10.	2,4-D + BAP	0.5 ppm	Jan. 2007
				11.	Picloram	3 ppm	Jan. 2007

Restoration through re-introduction

Transplantation feasibility experiments

As both the seed germination trials failed with *D. bourillonii*, a few wildlings were collected from Urulamthanni (Munnar Forest Division) and 10 of these were transplanted at KFRI, underneath the moist deciduous forest stand as a test case. Experimental transplantation of seedlings was also conducted likewise for of *H. bourdillonii*.

Transplantation of rooted clones of H. bourdillonii: About 60 ramets derived from rooting of stem cuttings were transferred to soil in polybags and kept in the

nursery. Hardened clones of *H. bourdillonii* rooted in 2007 were transplanted to the field at KFRI for pilot trial.

Restoration of species populations

Species recovery is the core of the programme where conservation of the species through re-introduction of the raised planting stocks to the wild was attempted. Both *in situ* and *ex situ* planting of the species were attempted. Survival and performance over the 5-year period was also conducted.

The objectives involved in two of the major objectives of the research project were: to explore the possibility of enhancing the natural populations of the two target species through appropriate translocation programmes and to evaluate the post-translocation survival and growth of the planted stocks. Sites for transplantations were selected based on ecological characteristics of the species and the safety of the locations, with respect to damage by fire, wildlife, men and browsing. Both these *in-situ* and *ex-situ* plantings were attempted.

Restoration planting involved four steps: i. Identification of *in situ* locations for planting. ii. Transporting the planting stock and planting. iii. Marking the planted area with concrete display board. iv. Monitoring survival and growth of planted seedlings.

Identification of planting sites

Eight natural ecosystems in the Western Ghats of Kerala region were selected (Table 6) for *in-situ* recovery planting. All the locations were natural evergreen forests. Four sites were selected for *ex-situ* planting.

Brief descriptions of the in-situ *planting sites*

Urulamthanni (Site 1) where the original populations of *D. bourdillonii* is growing, is an evergreen forest patch. The area is surrounded by human settlements and subject to encroachment, browsing of domestic animals and illegal firewood collection. Despite the disturbances, this location documented the largest number of regeneration of *D. bourdillonii* than in other natural locations.

Meenvallam (Site 2) is a natural habitat of *D. bourdillonii*; despite the vigorous (rubber) plantation activities in the adjacent areas, *D. bourdillonii* is represented by a few individuals and a good number of regeneration.

Valayamchal (Site 3) situated within the Aralam Wildlife Sanctuary is well protected from human disturbances. The sanctuary is an original habitat of *D. bourdillonii*. Decades back, the forest was subjected to timber

extraction. River courses still contain a few individuals of *D. bourdillonii* and a fair number of regeneration.

Ezhukumon (Site 4, Azhuthakadavu) located at Periyar Tiger Reserve (West) in Pathanamthitta Dt. and Ponnampara (Site 5) and Velithode (Site 6) within the Goodrickal Range of Pathanamthitta Dt. are the other locations selected for planting of *D. bourdillonii*. These forest areas are areas where *D. bourdillonii* grows naturally. Apparently as a result of past logging the species is now limited to a few scattered individuals along the inaccessible steep forest slopes.

Medicinal Plant Conservation Areas (MPCAs) are high diversity forest areas and for which special protection is ascertained by the Forest Department. Because of the protection available, the Kulamavu MPCA (Site 7), Idukki Dt., was selected for the planting of *D. bourdillonii.*

The *ex-situ* site, Karipponi (Site 8), is a moist deciduous forest located adjacent to Kalladikode hills. This area was selected for recovery planting because of the good establishment of *D. bourdillonii* beneath moist deciduous forests in Peechi. The other *ex-situ* sites, 9, 10 and 11, respectively KFRI-Arboretum, KFRI-Subcenter, Nilambur and KFRI-FRC, Velupadam are artificial garden environments with ensured protection (Table 6).

Table 6. Details of planting sites and their status.

Site No.	*Status*	*Planting sites*	*Geographic coordinates*	*Altitude (m asl)*
1.	*In-situ*	Urulamthanni, Neriamangalam Range, Idukki Dt.	*Lat*: 10° 7' 30" N. *Long*: 76° 45' 15" E.	60 m
2.	*In-situ*	Meenvallam, Mannarkkad Range, Palakkad Dt.	*Lat:* 10° 55' 12.8" N. *Long:* 76° 35' 25.1" E.	187 m
3.	*In-situ*	Valayamchal Aralam WLS, Kannur Dt.	*Lat:* 11°50' N. *Long:* 75° 49' E.	150 m
4.	*In-situ*	Ezhukumon, Periyar Tiger Reserve-West, Pathanamthitta Dt.	*Lat:* 9° 26' 3.2" N. *Long:* 76° 56' 46.7" E.	96 m
5.	*In-situ*	Ponnampara, Goodrickal Range, Pathanamthitta Dt.	*Lat:* 9° 23' 35.9" N. *Long:* 77° 2' 29.4" E.	273 m
6.	*In-situ*	Velithode , Kochukoical, Goodrickal Range, Pathanamthitta Dt.	*Lat:* 9° 19' 40.2" N. *Long:* 77° 1'.8" E.	166 m
7.	*In-situ*	Kulamavu Medicinal Plant Conservation Area (MPCA),	*Lat:* 9° 48.7' 4" N. *Long:* 76° 53.4' 5" E.	763 m

		Nagarampara Range, Idukki Dt.		
8.	*In-situ*	Karipponi, Mannarkkad Range, Palakkad Dt.	*Lat:* 10° 53' 29.2" N. *Long:* 76° 33' 18.7" E.	135 m
9.	*Ex-situ*	KFRI-Arboretum, Peechi, Trichur Dt.	*Lat*: 10° 31' 47" N. *Long*: 76° 22' 7.5" E.	45 m
10.	*Ex-situ*	KFRI-Subcenter, Nilambur, Malappuram Dt.	*Lat*: 11° 17' 28.5" N. *Long*: 76° 15' 36.3" E.	37m
11.	*Ex-situ*	KFRI-FRC, Velupadam, Trichur Dt.	*Lat*: 10° 26' 7.4" N. *Long*: 0076° 21' 39.9" E.	45m

Recovery planting of seedlings

Nursery raised seedlings of both the species were field planted in the translocation sites mentioned. Fully established 2-yr old poly-potted seedlings having an average height of 30-45 cm height were transported from the nursery to the planting sites. Pits of 45cm x 45 cm x 45 cm size were made in canopy gaps in the forest sites. Each seedling was tag-marked for future monitoring. Planting of the seedlings was done during the monsoon, July-September in 2010.

A total of 1,350 seedlings of *D. bourdillonii* were planted in the eight *in situ* (1,200 seedlings) and three *ex situ* locations (150 seedlings). A total of 450 seedlings of *H. bourdillonii* were planted in three *in situ* (300 seedlings) and three *ex situ* locations (150 seedlings).

Recovery planting of D. bourdillonii

H. bourdillonii was planted in all the eight *in-situ* sites and four *ex-situ* sites. The number of seedlings planted is given in Table 7; 1200 seedlings have been planted in *in-situ* sites and 300 seedlings planted in *ex-situ* sites.

Recovery planting of H. bourdillonii

Peermade Ghat, which includes the Periyar Tiger Reserve (PTR) is the only distribution area for *H. bourdillonii* in the Kerala. So, *in-situ* of *H. bourdillonii* was carried out in three *in situ* sites falling in this region: a. Kulamavu MPCA (Site 3), b. Azhutha kadavu, Ezhukumon (Site 5), and c. Ponnampara (Site 6). *Ex-situ* planting of the species was done only in three *ex-situ* sites. The total number of seedlings planted was 300 in *in-situ* sites were 300 and 150 in *ex-situ* sites (Table 8).

Table 7. Seedlings of *Dipterocarpus bourdillonii*, planted in various sites.

Site no.	*Planting site*	*Status*	*Dates of planting*	*Seedlings planted*
1.	Urulamthanni, Neriyamangalam Range	*In-situ*	07-08-2010	100
2.	Meenvellum, Mannarkkad Range	*In-situ*	11-08-2010	50
3.	Kulamavu MPCA Nagarampara Range	*In-situ*	18-08-2010	100
4.	Valayamchal, Aralam WLS	*In-situ*	31-08-2010	150
5.	Azhutha kadavu, Periyar Tiger Reserve-W	*In-situ*	07-09-2010	250
6.	Ponnampara, Goodrikkal Range	*In-situ*	15-09-2010	250
7.	Velithodu, Goodrikkal Range	*In-situ*	16-09-2010	150
8.	Karipponi, Mannarkkad Range	*Ex-situ*	12-08-200	150
		Subtotal: *In-situ* plantings:		**1200**
9.	KFRI-Arboretum, Peechi, Trichur Dt.	*Ex-situ*	27-07-2010	50
10.	KFRI-Subcenter, Nilambur, Malappuram Dt.	*Ex-situ*	02-08-2010	50
11.	KFRI-FRC, Velupadam, Trichur Dt.	*Ex-situ*	20-07-2010	50
		Subtotal: *Ex-situ* plantings:		**150**

Evaluation on post-translocation survival and growth

The survival of planted seedlings in each site was monitored at 6 months intervals after planting. The height increments of each seedling were also taken during each visit.

Signboards for planting sites

The planting sites in the natural forest areas (8 sites) have been permanently demarcated by fixing concrete display boards. The board displayed with relevant information such as title of the project, funding agency, GPS details of the location, date and number of seedlings planted, etc.

Table 8. Seedlings of *Humboldtia bourdillonii*, planted in various sites.

Site no.	*Planting site*	*Status*	*Dates of planting*	*Seedlings planted*
3.	*H. bourdillonii*	Kulamavu MPCA Nagarampara Range	18-08-201(	200
5.	*H. bourdillonii*	Azhutha kadavu Ehukumon, PTR-W	07-09-201(	50
6.	*H. bourdillonii*	Ponnampara, Goodrikkal Range	15-09-201(	50
		Subtotal: *In-situ* plantings	**300**	
9.	*H. bourdillonii*	KFRI-Arboretum, Peechi, Trichur Dt.	27-07-201	50
10.	*H. bourdillonii*	KFRI-Subcenter, Nilambur, Malappuram Dt.	02-08-201	50
11.	*H. bourdillonii*	KFRI-FRC, Velupadam, Trichur Dt.	20-07-201	50
		Subtotal: *Ex-situ* plantings	**150**	

5. Results and discussion

This chapter is organized into two sections, the first dealing with *Dipterocarpus bourdillonii* and the second with *Humboldtia bourdillonii*.

1. Dipterocarpus bourdillonii

Locally known as *Kar anjili* and *Kalpayin* (Malayalam), *Dipterocarpus bourdillonii* is a graceful tree devoid of branches in the bole up to 30 m (Fig. 6). All cited instances of the species are along stream banks; only rarely do they inhabit off-stream non-riparian habitats.

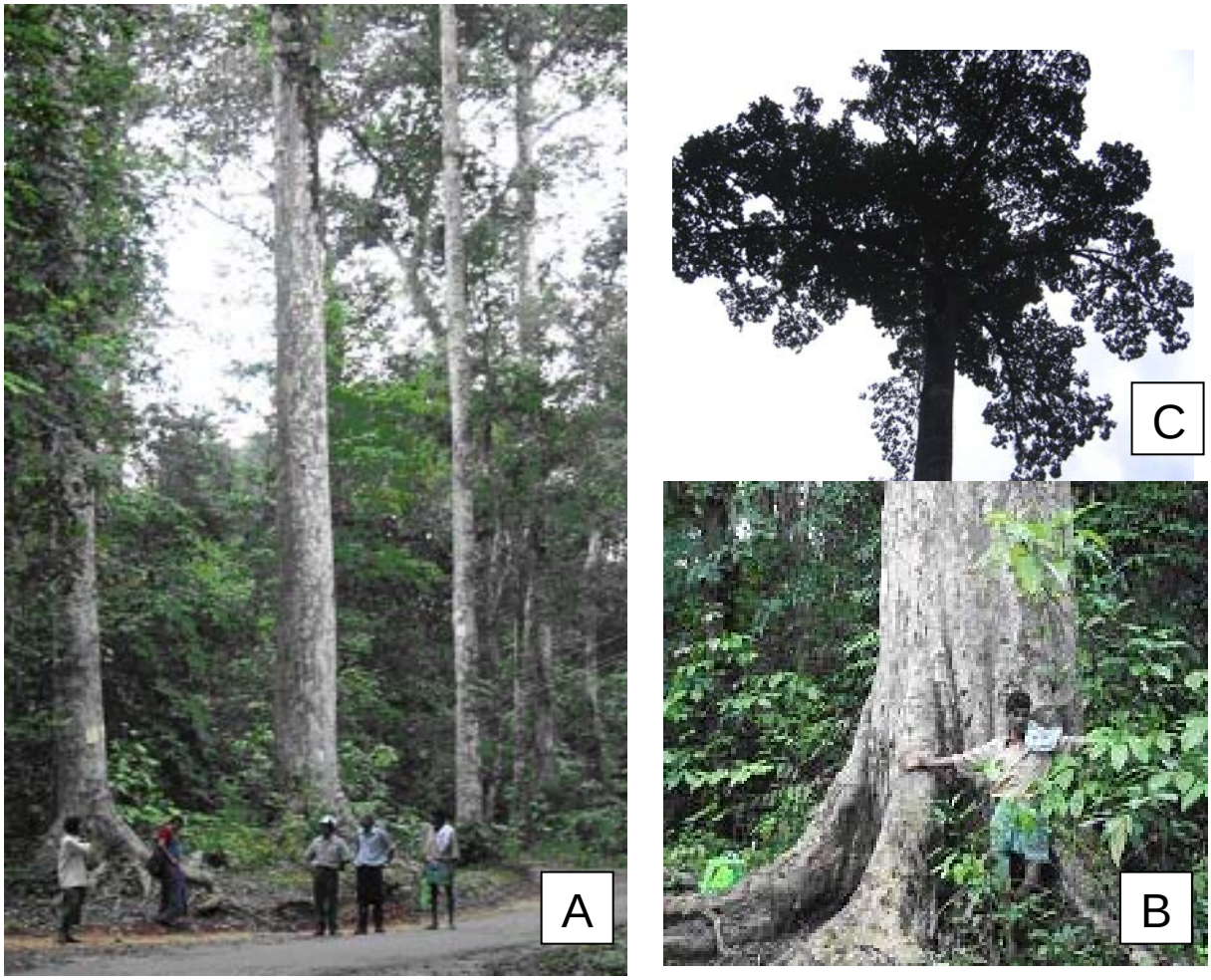

Figure 6. *Dipterocarpus bourdillonii.* Fig. 6 A. A stand showing the gigantic trees of *D. bourdillonii*. Fig. 6 B. A picture showing the large size of the bole. Fig. 6 *ca.* The crown.

A botanical description of the species is given below. Photographs of flowers, floral parts, and fruits are also given in Fig. 7.

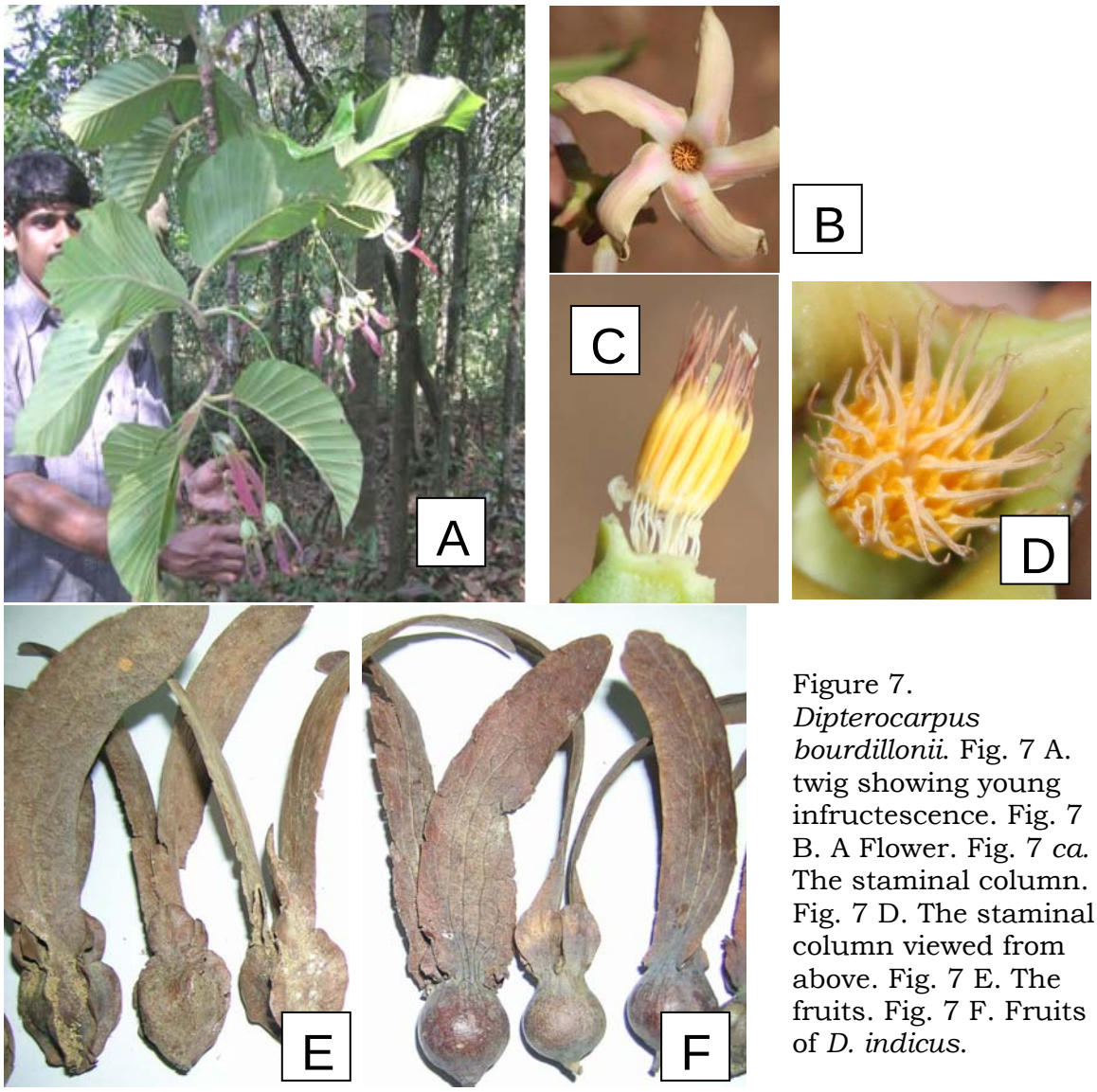

Figure 7. *Dipterocarpus bourdillonii*. Fig. 7 A. twig showing young infructescence. Fig. 7 B. A Flower. Fig. 7 *ca.* The staminal column. Fig. 7 D. The staminal column viewed from above. Fig. 7 E. The fruits. Fig. 7 F. Fruits of *D. indicus*.

Trees: large, 35-40 m tall attaining emergent strata piercing the canopy. *Bole:* around 180 cm in diameter, straight, cylindric, often with plank buttress at the base. *Bark:* pale, exfoliating in thick irregular flakes. *Leaves:* simple, alternate, blade ovate, tip acuminate, base round or sub-cordate, margins entire or wavy, coriaceous; lateral nerves 12-15 pairs, looped and connected by transverse reticulation; leaf-nerves below and young shoots clothed with spreading tomentum; petiole often swollen just below the leaf-blade; stipules large encircling the stem, falling early and leaving an annular scar. *Flowers:* large, white or reddish, in short racemes. Calyx with a free tubular base. Petals valvate, pubescent. Stamens numerous; anthers elongated,

acuminate. Ovary with 3-2-ovuled cells and a filiform style. *Fruit:* 1 seeded nut, enclosed in a 5-winged calyx-tube with 2 erect wings.

Distribution: The known distribution of the species as given by Ramesh & Pascal (1991) is reproduced in Fig. 8 A. The distribution map generated using DIVA-GIS (Fig. 8 B) closely follows the former, but was found not really useful in planning field explorations, because of the small map scale. In both the cases however, the patchy distribution is evident.

The species has been encountered from the following locations: KARNATAKA: Coorg: Mari Gunty. KERALA: Aralam Wildlife Sanctuary: Chavachi thodu; Attappadi: Chittoor Riv.; Mannarkkad: Meenvallam; Urulamthanni: Pinavurkudi; Pooyamkutty: Manikandanchal; Muzhiyar: Moonnumukku; Periyar Tiger Reserve: Pampa; Achenkoil: Vazhaperiyar, Kallar valley.

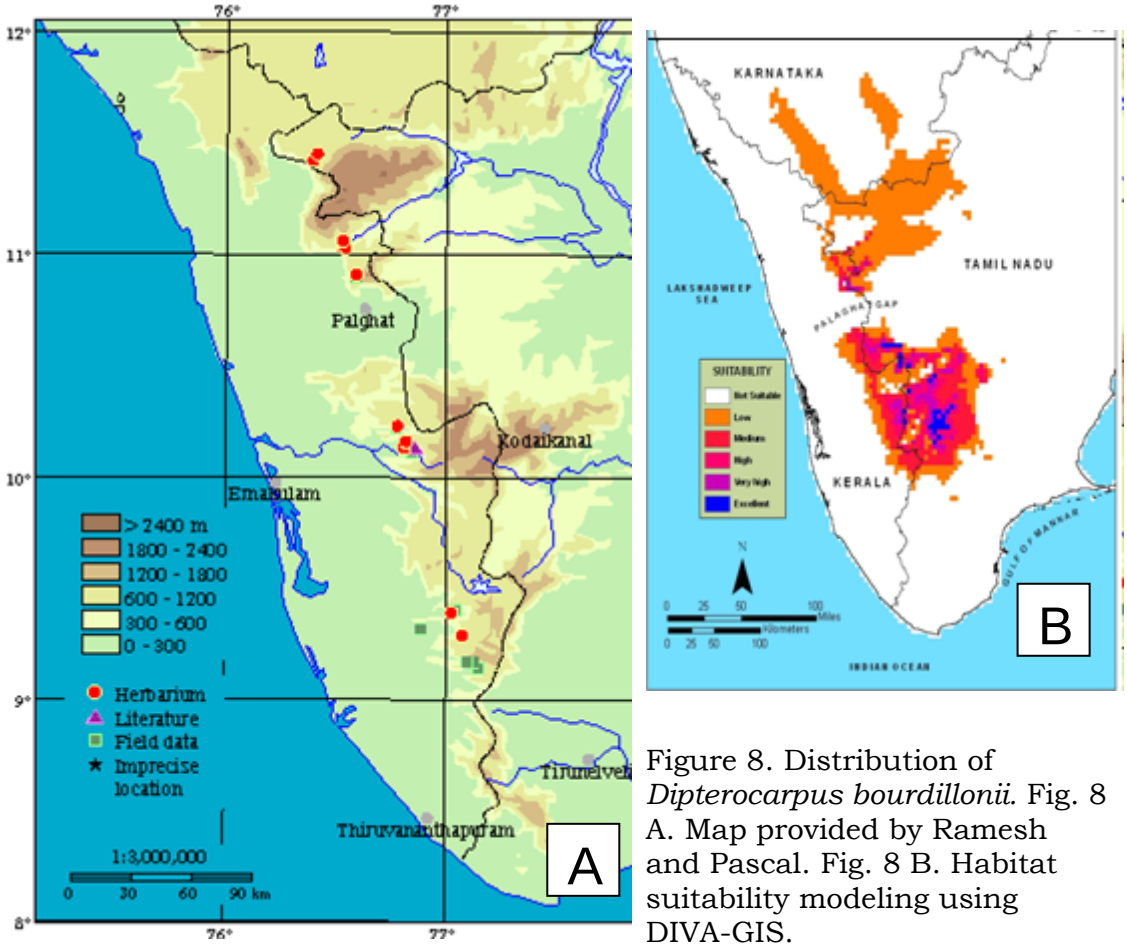

Figure 8. Distribution of *Dipterocarpus bourdillonii.* Fig. 8 A. Map provided by Ramesh and Pascal. Fig. 8 B. Habitat suitability modeling using DIVA-GIS.

Population ecology of *D. bourdillonii*

Communities supporting D. bourdillonii

Structure of the stands where *D. bourdillonii* is found differ from one another. Composition of the stands at two locations, respectively from north of Palghat Gap and south of it (Chavachi and Goodrickal) are given in Table 9. *Vateria indica*, *Knema attenuata*, *Polyalthia fragans*, *Syzygium* spp., *etc.*,., were the common associates of *D. bourdillonii* in the Western Ghats north of Palghat Gap. In the stand at Chavachi, *Vateria indica* and *D. bourdillonii* had almost equal species importance values and the species together occupied *ca.* 35% stand's IVI.

Table 9. Structure of the stand in the decreasing order of dominance at Chavachi in the Aralam Wildlife Sanctuary (North of Palghat Gap).

Species	**D**	**BA**	**RD**	**RF**	**RBA**	**IVI**	**RIVI**	**CRIVI**
Vateria indica	78	3.87	20.4	7.3	26.0	53.7	17.9	17.9
Dipterocarpus bourdillonii	18	6.24	4.7	3.6	41.9	50.3	16.8	34.6
Knema attenuata	16	0.55	4.2	3.6	3.7	11.5	3.8	38.5
Syzygium spp	4	1.16	1.1	1.5	7.8	10.3	3.4	41.9
Combretum albidum	16	0.05	4.2	4.4	0.3	8.9	3.0	44.8
Polyalthia fragrans	10	0.33	2.6	3.6	2.2	8.5	2.8	47.7
Atalantia racemos	12	0.04	3.1	3.6	0.3	7.0	2.3	50.0
Myristica beddomei	10	0.11	2.6	2.9	0.7	6.2	2.1	52.1
Pterygota alata	6	0.38	1.6	1.5	2.6	5.6	1.9	53.9
Litsea glabrata	10	0.08	2.6	2.2	0.5	5.3	1.8	55.7
Syzygium laetum	10	0.02	2.6	2.2	0.1	4.9	1.6	57.4
Pterospermum reticulatum	6	0.16	1.6	2.2	1.1	4.8	1.6	59.0
Artocarpus hirsutus	6	0.12	1.6	2.2	0.8	4.5	1.5	60.5
Dimocarpus longan	6	0.11	1.6	2.2	0.8	4.5	1.5	62.0
Nothopegia colebrookeana	6	0.09	1.6	2.2	0.6	4.4	1.5	63.4
Hydnocarpus alpina	6	0.06	1.6	2.2	0.4	4.2	1.4	64.8
unknown species2	6	0.03	1.6	2.2	0.2	4.0	1.3	66.1
Tabernaemontana gamblei	6	0.03	1.6	2.2	0.2	3.9	1.3	67.4
Tetrameles nudiflora	6	0.13	1.6	1.5	0.9	3.9	1.3	68.7
Ixora pavetta	6	0.01	1.6	2.2	0.1	3.8	1.3	70.0
Drypetes venusta	6	0.01	1.6	2.2	0.1	3.8	1.3	71.3
Holigarna grahamii	6	0.08	1.6	1.5	0.5	3.5	1.2	72.5
Gnetum edule	6	0.07	1.6	1.5	0.5	3.5	1.2	73.6
Reinwardt. anamalaiense	4	0.14	1.1	1.5	1.0	3.5	1.2	74.8
Xanthophyllum arnottianum	6	0.04	1.6	1.5	0.3	3.3	1.1	75.9
Mangifera indica	6	0.03	1.6	1.5	0.2	3.2	1.1	77.0
Baccaurea courtallensis	6	0.02	1.6	1.5	0.1	3.2	1.1	78.0
Caesalpinia cucullata	6	0.02	1.6	1.5	0.1	3.2	1.1	79.1
Vepris bilocularis	2	0.23	0.5	0.7	1.5	2.8	0.9	80.0
Alstonia scholaris	2	0.21	0.5	0.7	1.4	2.7	0.9	80.9
Acacia caesia	4	0.02	1.1	1.5	0.1	2.6	0.9	81.7
Dalbergia horrida	4	0.02	1.1	1.5	0.1	2.6	0.9	82.6
Hopea parviflora	4	0.01	1.1	1.5	0.1	2.6	0.9	83.5
Dysoxylum spp	4	0.01	1.1	1.5	0.1	2.5	0.9	84.3
Sarcostigma kleinii	4	0.01	1.1	1.5	0.1	2.5	0.9	85.2

Ancistrocladus heyneanus	4	0.01	1.1	1.5	0.1	2.5	0.8	86.0
Homalium zeylanicum	4	0.01	1.1	1.5	0.1	2.5	0.8	86.8
Humboldtia vahliana	4	0.01	1.1	1.5	0.1	2.5	0.8	87.7
Strombosia ceylanica	4	0.01	1.1	1.5	0.1	2.5	0.8	88.5
Chionanthus mala-elengi	4	0.02	1.1	0.7	0.1	1.9	0.6	89.1
Neolamarckia cadamba	2	0.07	0.5	0.7	0.5	1.7	0.6	89.7
Casearia ovata	2	0.06	0.5	0.7	0.4	1.7	0.6	90.3
Trewia nudiflora	2	0.06	0.5	0.7	0.4	1.64	0.6	90.8
Cinnamomum malabatrum	2	0.05	0.5	0.7	0.3	1.6	0.5	91.3
Toona ciliata	2	0.03	0.5	0.7	0.2	1.5	0.5	91.8
Symplocos racemosa	2	0.02	0.5	0.7	0.2	1.4	0.5	92.3
Flacourtia montana	2	0.02	0.5	0.7	0.1	1.4	0.5	92.7
Meliosma pinnata	2	0.01	0.5	0.7	0.1	1.3	0.4	93.2
Drypetes oblongifolia	2	0.01	0.5	0.7	0.1	1.3	0.4	93.6
Garcinia morella	2	0.01	0.5	0.7	0.1	1.3	0.4	94.0
Otonephelium stipulaceum	2	0.01	0.5	0.7	0.1	1.3	0.4	94.5
Persea macrantha	2	0.01	0.5	0.7	0.1	1.3	0.4	94.9
Strychnos colubrina	2	0.01	0.5	0.7	0.1	1.3	0.4	95.3
Mesua ferrea	2	0.01	0.5	0.7	0.1	1.3	0.4	95.7
Polyalthia coffeoides	2	0.01	0.5	0.7	0.1	1.3	0.4	96.1
Anamirta cocculus	2	0.01	0.5	0.7	0.1	1.3	0.4	96.6
Diospyros candolleana	2	0.01	0.5	0.7	0.1	1.3	0.4	97.0
Mallotus philippensis	2	0.01	0.5	0.7	0.1	1.3	0.4	97.4
Prunus ceylanica	2	0.01	0.5	0.7	0.1	1.3	0.4	97.8
Psychotria nudiflora	2	0.01	0.5	0.7	0.1	1.3	0.4	98.2
Unidentified sp.1	2	0.01	0.5	0.7	0.3	1.3	0.4	98.7
Unidentified sp.2	2	0.01	0.5	0.7	0.3	1.3	0.4	99.1
Unidentified sp. 3	2	0.01	0.5	0.7	0.1	1.3	0.4	99.5
Unidentified sp. 4	2	0.01	0.5	0.7	0.1	1.3	0.4	99.9
Total:	**382**	**14.89**	**99.9**	**99.8**	**100.0**	**299.8**	**99.9**	**99.9**

D=Density, BA=Basal area, RD=Relative density, RF=Relative frequency, RBA=Relative basal area, IVI=Importance value index, RIVI=Relative importance value index (IVI/3), CRIVI=Cumulative relative importance value index.

At Goodrickal, south of Palghat Gap, the common associates of *D. bourdilloii* were *Turpinia malabarica*, *Cinnamomum malabaricum*, *Drypetes oblongifolia*, *Baccaurea courtallensis etc.,*. (Table 10). Here, principal species of the forest type such as *Vateria indica* do not appear in the list of dominant species; this is indicative of the degraded nature of the forest. Though low in density and frequency, *D. bourdillonii* shows high dominance by way of its large basal area.

Table 10. Structure of the stand in the decreasing order of dominance at Pannikunnu in the Goodrickal Forest Range (South of Palghat Gap).

Species	**D**	**BA**	**RD**	**RF**	**RBA**	**IVI**	**RIVI**	**CRIVI**
Dipterocarpus bourdillonii	28	36.87	5.5	6.1	47.6	59.1	19.7	19.7
Turpinia malabarica	65	1.66	12.9	9.1	2.1	24.1	8.0	27.7
Cinnamomum malabatrum	50	3.78	9.9	8.1	4.9	22.9	7.6	35.3
Drypetes oblongifolia	55	1.40	10.9	7.1	1.8	19.8	6.6	41.9
Baccaurea courtallensis	48	0.60	9.4	7.1	0.8	17.3	5.8	47.7
Strombosia ceylanica	33	2.37	6.4	6.1	3.1	15.6	5.2	52.9

Knema attenuata	30	2.75	5.9	5.2	3.6	14.5	4.9	57.7
Tetrameles nudiflora	3	10.03	0.5	1.0	12.9	14.4	4.8	62.5
Polyalthia fragrans	28	0.80	5.5	6.1	1.0	12.6	4.2	66.7
Dysoxylum malabaricum	15	3.99	3.0	4.0	5.2	12.2	4.1	70.8
Hopea parviflora	15	3.83	3.0	4.0	5.0	12.0	4.0	74.8
Elaeocarpus serratus	25	0.94	5.0	4.0	1.2	10.2	3.4	78.2
Macaranga peltata	15	1.52	3.0	5.1	2.0	10.0	3.3	81.5
Hydnocarpus macrocarpa	18	0.37	3.5	4.0	0.5	8.0	2.7	84.1
Vateria indica	10	1.03	2.0	4.0	1.3	7.4	2.5	86.6
Dillenia pentagyna	8	3.27	1.5	1.0	4.2	6.7	2.2	88.8
Artocarpus heterophyllus	13	0.21	2.5	3.0	0.3	5.8	1.9	90.8
Persea macrantha	8	0.64	1.5	2.0	0.8	4.3	1.4	92.2
Actinodaphne malabarica	10	0.11	2.0	2.0	0.1	4.1	1.4	93.6
Xanthophyllum arnottianum	8	0.11	1.5	2.0	0.1	3.7	1.2	94.8
Litsea floribunda	5	0.27	1.0	2.0	0.4	3.4	1.1	95.9
Drypetes venusta	5	0.06	1.0	2.0	0.1	3.1	1.0	96.9
Myristica beddomei	5	0.27	1.0	1.0	0.4	2.4	0.8	97.7
Sterculia guttata	3	0.36	0.5	1.0	0.5	2.0	0.7	98.4
Holigarna beddomei	3	0.22	0.5	1.0	0.3	1.8	0.6	99.0
Dimocarpus longan	3	0.02	0.5	1.0	0.1	1.5	0.5	99.5
Garcinia wightii	3	0.02	0.5	1.0	0.1	1.5	0.5	100.0
Total:	**505**	**77.51**	**100.1**	**100.0**	**100.0**	**300.0**	**100.0**	**100.0**

D=Density, BA=Basal area, RD=Relative density, RF=Relative frequency, RBA=Relative basal area, IVI=Importance value index, RIVI=Relative importance value index (IVI/3), CRIVI= Cumulative relative importance value index.

Population size of D. bourdillonii

The areas explored and the trees (≥30cm gb) encountered are summarized in Table 11. The species has been found restricted to areas adjoining river courses only in interior forests. Major outcomes are also summarized subsequently.

Table 11. Number of trees (individuals >= 30 cm gbh) of *Dipterocarpus bourdillonoii* recorded during field visits.

No.	Locations	Trees *
1.	Mari Gunty, Coorg, Karnataka	1
2.	Chavachi thodu, Aralam Wildlife Sanctuary	52
3.	Chittoor Riv., Attappadi, Mannarkkad	2
4.	Meenvallam, Mannarkkad	1
5.	Urulamthanni, Pinavurkudi	51
6.	Manikandanchal, Pooyamkutty	19
7.	Moonnumukku, Muzhiyar	28
9.	Pampa, Periyar Tiger Reserve	49
10.	Vazhaperiyar, Kallar valley, Achenkoil	25
	Total:	**228**

The total number of trees encountered in Kerala was 228 trees. Joint explorations together with Dr. CG Kushalappa in Coorg (Karnataka), succeeded in locating one tree of the species in the forests of

Subrahmanya. Thus the total population is 229 trees. Giving a 5% allowance for the probable omissions in sampling, the total population would not go beyond 240 trees.

The largest population so far encountered is composed of 52 trees at Chavachi (Aralam, Kannur), followed by 51 trees at Pinavurkudi (Urulamthanni) and 49 trees at Pampa (Table 11).

An earlier study conducted by the Kerala Forest Research Institute (unpublished) indicated that the species has only sparse and widely separated populations. Field explorations conducted during the implementation of the current study confirmed the patchy distribution of the species (Fig. 8). It also confirmed that the species does not extend north of Coorg, (Karnataka).

A '*threatened*' status was assigned to the species by Ramesh & Pascal (1991) and Sasidharan (2003) assigned a *'Low Risk/ near threatened'* status. Outside the conventional floristic documentations, no population details of the species based on field surveys are available except for the current study. According to the IUCN criteria, the species thus qualifies a 'Critically Endangered' status.

Population structure of D. bourdillonii

Population structure of a major section of the population was analyzed. Of the 179 trees (≥30cm gbh) encountered, 78 were Very Large trees (Girth Class, GC: >270 cm), 37 Large trees (GC: 180-270 cm), 48 Medium trees (GC: 90-180cm). There were only 9 Small trees (GC: 60-90 cm) and 7 Poles (GC: 30-60 cm) recorded. Against this there were only 20 saplings (GC 10-30cm), which is a very low figure.

The life stage structure of five larger populations and the composite (average) population structure are given in Fig. 9. Individual populations of the species differed in their life stage structure.

Two out of the four populations (Moonnumukku and Pamba) had only larger girth classes, ≥60cm gbh, *i.e.*, they did not have young ones to replace their older trees. On the other hand, Pinavurkudi and Chavachi had all the life stages including established seedlings, Saplings, Poles and mature trees. Very interestingly, at Chavachi (Aralam) the seedling population was rich, in comparison to its mature tree population. Against the population of 179 mature trees (>= 10cm dbh) in Kerala, we have come across only 60-70 seedlings.

At Moonnumukku and Pamba seedlings were almost wanting and at Pamba, both seedling and young tree-populations were also wanting. The skewed population structure at Pamba can be due to the impact of intense pilgrimage (to Sabarimala) on the local vegetation.

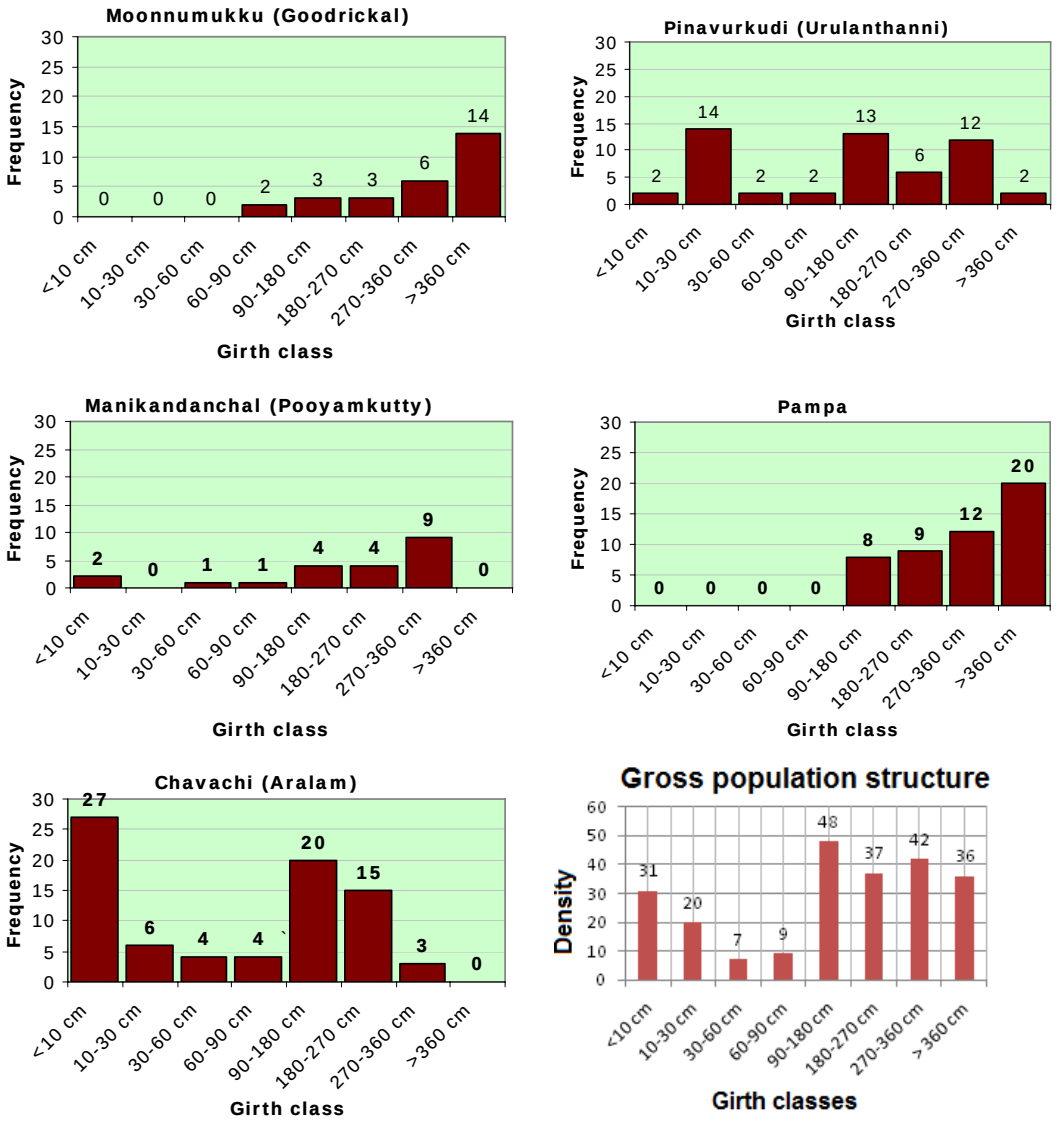

Figure 9. Population structure of *Dipterocarpus bourdillonii* (continued).

At Pinavurkudi, there are a few scattered seedlings and advanced seedlings. Surprisingly, the Chavachi (Aralam) population showed some what good representation both in the seedling and advanced-seedling populations. Unlike in other sites, the trees here were of smaller size (in terms of GBH).

Absence of young ones to replace their older individuals is characteristic of a species moving towards extinction. The small population size, which is a result of failure to reproduce efficiently, is also characteristic of such a species. The non-viable embryo in the mature fruits essentially tells the same.

Reproductive biology of *D. bourdillonii*

Reproductive biological studies of the species was conducted in order to: (a) identify prevailing reproductive constraints if any, (b) identify

the life stages at which the reproductive anomalies operate, and (c) to generate knowledge useful for the effective propagation of the species.

As details on the breeding system and reproductive constraints in *D. bourdillonii* were not subject to detailed study earlier, floral biology, breeding behavior, seed dispersal, germination capacity, survival of seedlings and adults, *etc.,* were subjected to study (Fig. 10). The findings from the above studies are discussed below.

Figure 10. *Dipterocarpus bourdillonii*. Fig. 10 A. A cane swing and pulley system for accessing the canopy. Fig. 10 B. A reed bamboo platform built in the canopy for studying the reproductive biology.

Reproductive phenology of D. bourdillonii

Flowering of the species spanned between mid-November to late-December. Sporadic flowering occurred during early March. Trees at Muzhiyar and Pamba flowered between the second week of January till the end of March. Leaf fall occur immediately before the initiation of flowering and flushing followed it. Mature fruits are available by May. Flowering, fruit development and dispersal were found synchronous, *ie*, without much time lag across the individuals of the population.

Inflorescence initials originated from axillary positions and matured to produce flowers in 14–16 days. Each pendant inflorescence contains 5-7 flowers arranged alternately (Fig. 11 A) and the tree remains flowering for 14-20 days. One flower in each inflorescence opens on alternate days but rarely they also open in consecutive days or two flowers together on the same day.

Flowers are complete, actinomorphic and scented. Calyx is penta-sepalous, valvate and connate. Sepals are green and red-tinged,

particularly two of them which grow and exceed the others to become the wings of the fruit.

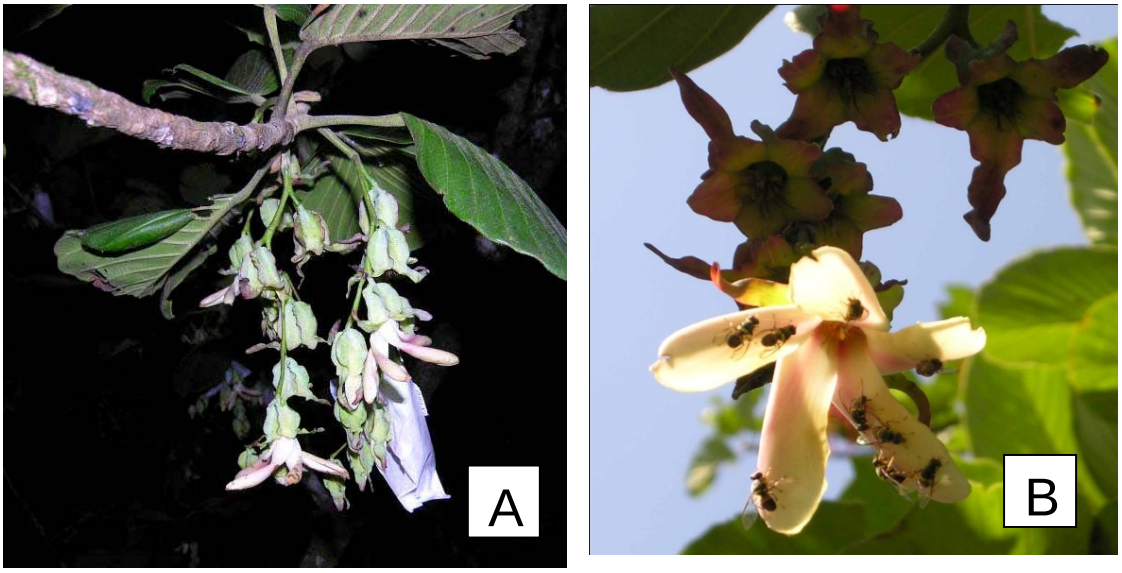

Figure 11. *Dipterocarpus bourdillonii.* Fig. 11 A. A pendant panicle. Fig. 11 B. A flower.

Corolla is penta-petalous; cyclic and showy (Fig. 11 B). Petals are elongated and off-white in colour. Stamens are short, golden yellow, *ca.* 30, arranged in three whirls. Each anther is subtended by thin flat filament and ends in hairy apical appendage (Fig. 7 C & D). Pollen grains are golden yellow and sticky.

Gynoecium is tri-carpellary with *ca.* 12 mm long style, simple stigma and dome-shaped ovary. The syncarpous ovary has two ovules in each carpel. Fruit is a single seeded nut enclosed within the calyx-cup. The connate margins of the calyx-cup develop into five ridges on the surface of the fruit, to become the distinguishing feature of *D. bourdillonii*. The fruit is dispersed by wind and the two wings developed from the two accrescent sepals assist in dispersal.

Floral Biology

Flower opening begins at dusk around 6.45pm and fully opens around 7.15pm. By the time the flowers are open, pollen grains are viable and stigma is receptive. In another species of *Dipterocarpus, viz, D. obtusifolius*, flower opening is again reported to be shortly after sunset at *ca.* 7 pm and complete by 8 pm (Ghazoul, 1997). Pollen grains were observed on the stigma of open pollinated flowers protected by insect nets and paper bags.

Pollen grains were viable 12 hr prior to flower opening. At flower opening pollen grains are 100 % viable; thereafter viability decreases.

After 12 hrs viability is reduced drastically, by 18 hrs, 50% loses viability and by 24hrs, 95% becomes non-viable.

Stigma was receptive even 9 hours prior to flower opening but the pollen grains take 3-4 hrs for germination on the stigma. Stigma receptivity continued even 20 hrs after flower opening. Ghazoul (1997) also reported long stigma receptivity (16 hr) in *Dipterocarpus obtusifolius*.

Pollination: The pendant flowers hardly allow wind pollination. Pollen grains are heavy and much sticky. Pollen cannot be transported by wind when they are heavy and sticky (Krezdorn, 1986; Sanford, 1992; Schneider, 1968). Breeding experiments proved that fruits derived from flowers deprived of insect visits failed to germinate. This, coupled with the high pollen load on the insect's body infers that *D. bourdillonii* is adapted to entomophily. Open flowers also emit a pleasant fragrance; this invites the insects to visit the flowers (Bell, 1985; Webber, 2000).

Pollinators: Insects visiting the flowers include bees, butterflies and sunbirds. Butterflies were rare and often they visited flowers in the outer canopy only. Sun bird visited very rarely. Bees are the pollinators which visit in the morning (7am- 11am) and at dusk. They penetrate to the base of the flowers for nectar and move from flower to flower and from tree to tree. There are two species of bees, *Apis indica*, smaller in size and seen in large numbers during peak hours. They visit flowers one after other, spend more time on the same tree, have low flight speed and thus facilitates more self pollination. The larger bee *Apis dorsata* though seen rarely, has a high flight speed, visits only a few flowers of a tree but travels across many trees, thus facilitating cross pollination.

Pollen grains of *D. bourdillonii* are seen on the hind limbs of both the honey bees, under microscope. Bees are reported to be the pollinators in a few other Dipterocarp species too. Dayanandan *et al.* (1990) report bees (*Apis* spp.) as pollinators in *Shorea megistophylla* and *Vateria copallifera* and Momose *et al.* (1994) reported stingless bees (*Trigona* spp.) as pollinators in *Dryobalanops lanceolata.* Medium sized bees (*Apis* spp.) are the pollinators in *Stemonoporous oblongifolius* and *Shorea trapezifolia* (Bawa, 1998). In many species of *Dipterocarpus* in Sarawak (Malaysia), flowers open in the evening and *Apis dorsata* is an important pollinator (Momose *et al.*, 1998). Another bee-like insect (Fig. 11 B) stay on petals, suck the sap of the petal and never tend to reach the base of the flower. Though the flowers are scented, insect visitors are not found at night hours, except weevils visiting for oviposition.

Growth of pollen tube and self incompatibility: In both self- and cross-pollinated flowers of *D. bourdillonii*, most of the pollen tubes (*ca.* 90%)

lost their direction, meshed within the stigmatic exudates and failed to penetrate the stigma (Fig. 12). Pistils generally carried more than 100 germinated pollen grains and out of the pollen tubes, only less than 10 pollen tubes penetrated the stigma and most of them reached the ovary. It takes *ca.* 12 hrs for the pollen tubes to reach the ovary and 16-21 hours to reach the ovule. Pollen tubes in cross pollinated pistils reached the ovary a little earlier and apparently the entry into the ovary is limited to a few which arrive earlier. Self-incompatibility caused by defective pollen-tube guidance and delayed abortion of selfed flowers reported in *Dipterocarpus tempehes* (Kenta *et al.*, 2002) go along with our findings.

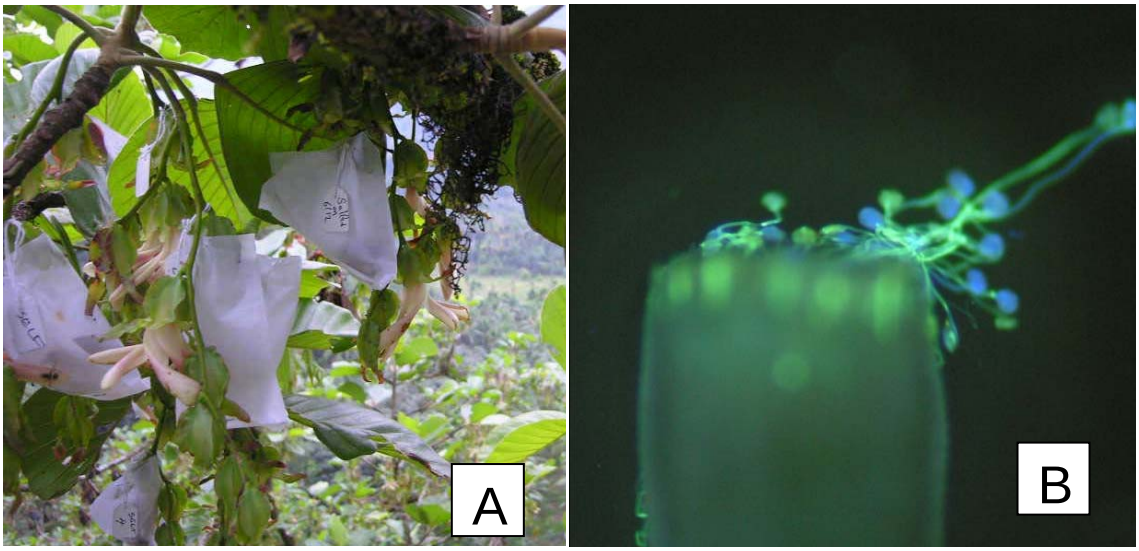

Figure 12. *Dipterocarpus bourdillonii.* Fig.12 A. Controlled pollination experiments on the tree. Fig. 12 B. Pollen tube growth on the stigma.

Self incompatibility in *D. bourdillonii* is of 'post-micropyle entry' type, *ie*, expressed after the pollen tube enters into the micropyle, and is a kind of late-acting self-incompatibility (LSI) comparable to the ovarian self-incompatibility (OSI). Here fruits develop but at a later stage the embryos degenerate. Seavey and Bawa (1986) pointed out that late acting self-incompatibility is characteristic of many woody plants. In a few other dipterocarps, there are reports of self-incompatibility expressed even later, during the course of seed and fruit development, and sometimes many days after fertilization (Chan, 1977, Ghazoul, 1997, Ghazoul *et al.,* 1998, Kenta *et al.,* 2002, Nakagawa *et al.*, 2005).

Fruit development and seed set in D. bourdillonii

Out of the six ovules only one grows into the seed and the remaining ovules fail to grow and thus degenerate (Fig. 13). Degeneration of the ovules is not due to lack of pollination; apparently it is an adaptation, a mechanism for resource economization.

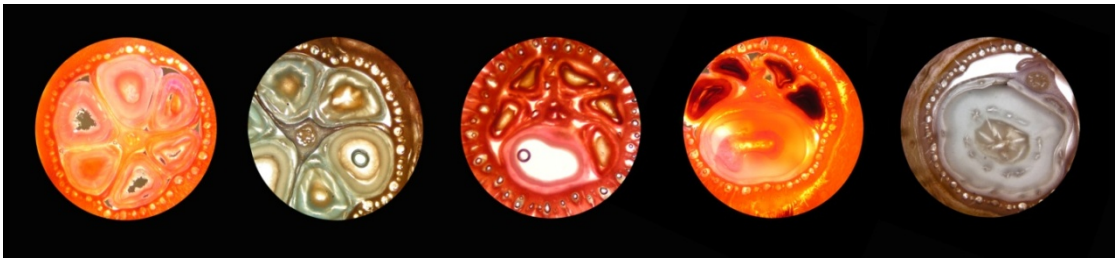

Figure 13. Stages in the abortion of ovules and development of the seed.

Development of fruits progresses till 60-65 days after flower opening and then the fruits undergo browning. Thereafter, drying of fruits takes place on the tree itself. Mature fruits are available on the tree 75-80 days after flower opening.

Natural seed set: Out of the 228 flowers observed for seed set, in flowers which were not protected from insects, 88% fruit set was observed; nearly an equal amount of flowers set fruits (87.5%) which were protected from insects using insect nets. In flowers protected by application of insecticide and fungicide 93.7% fruit set was observed.

Germination trials on fruits/seeds derived from flowers in which insect visits were excluded by emasculation and insect nets, the seeds failed to germinate. At the same time fruits/seeds derived from open-pollinated flowers germinated; this shows the importance of insect pollinators for successful reproduction of the species. Similar dependence on insects for effective reproduction was also documented for *Dipterocarpus obtusifolius* in Malaysia (Ghazoul (1997); there again, fruits derived from flowers protected from insects failed to germinate.

Germination failure of apomictic embryos: Out of the 40 flowers bagged (Fig. 6) for testing the presence of apomixis, 77.5 % developed into fruits and contained embryos but only 50% survived to maturity (Table 1). Presence of apomictic embryogeny is obvious from the experiment but none of the fruits germinated. A few earlier workers have also reported apomictic development in other dipterocarpacean members (Kaur *et al.*, 1986; Chan, 1981; Murawski *et al.*, 1994).

Germination failure of fruits/seeds derived from self pollination: Out of the 91 flowers artificially self-pollinated, 98% progressed with fruit development but only 56% of the total survived to maturity. None of the mature fruits/seeds germinated showing self incompatibility. A similar instance of null selection of selfed progenies was pointed out in *Shorea leprosula* (Lee *et al.* (2000).

Seed setting in cross pollinated flowers: Ninety-eight percent of the cross pollinated flowers developed into fruits and 77% progressed to

maturity (Fig. 14). Thirteen out of 37 fruits (35.1 %) derived from cross pollination experiments germinated (Table 12).

Table 12. Fruit set in artificially self- and cross- pollinated flowers.

Period after pollination	Successful fruit set (%) in artificial breeding experiments		
	Apomixis	Controlled self pollination	Controlled cross pollination
1-10 days	100	100	100
11-20 days	77.5	97.8	97.9
21-40 days	72.5	95.6	97.9
41-55 days	50.0	93.4	91.7
56-75 days	50.0	56.0	77.0

Figure. 14. *Dipterocarpus bourdillonii.* Collection of flowers and fruits for microscopy.

The above experiments clearly demonstrate that *D. bourdillonii* is a self-incompatible species expressing incompatibility in the post-zygotic phase. Existence of post-zygotic self incompatibility system has also been reported for another Dipterocarp, *Dryobalanops lanceolata* (Inoue-personal observation). Nagamitsu *et al.* (2001) also reported self-incompatibility in *Shorea leprosula* (Dipterocarpaceae) and argued that inbreeding depression causes to reduce the proportion of inbred embryos.

Factors affecting fruit/ seed populations in D. bourdillonii

Insects interfere the reproductive cycle of *D. bourdillonii* in two stages, flower buds and fruits. Two groups of insects, the Dipteran and the Lepidopteran, attack the growing flower buds. Larvae of an unidentified Dipteran, grow inside the flower buds and cause abscission (Fig.15). Out of the fallen flower buds, 90 % were due to the attack of the insect. Larvae of another unidentified Lepidopteran were

also found in buds eating up the floral parts inside the corolla. Often, the buds fall off but the whole metamorphosis take place inside the fallen bud and adult emerges out. Insects were reared in the laboratory, but could not be identified.

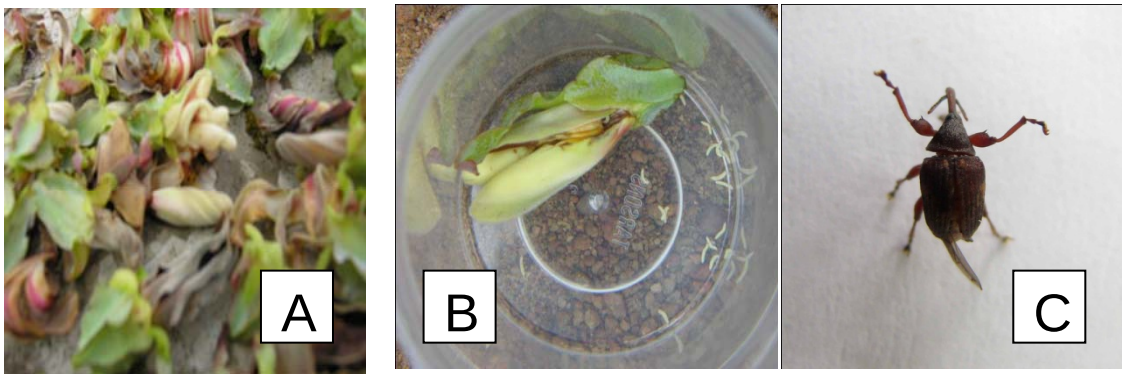

Figure 15. *Dipterocarpus bourdillonii*. Fig. 15 A. Flower buds abscised due to Dipteran attack. Fig. 15 B. Insect larve emerging from the flower bud. Fig. 15 *ca*. Weevil.

As noted above, the fruits destroyed by weevils accounted to 31.9%. The percentage of fruit decay by causes other than insects was 60.7% which included 7% by fungal infection. The percentage of viable fruits was only 7.4.

Insects attacking the fruits are weevils which ovipose inside the ovary near the base of the style, within two days after anthesis (Fig. 16). The larvae grow inside and eat up the embryo but fruit development and dispersal go routine. Before or after dispersal, the mature insects emerge out of the ovary. Around 32% of fruits were damaged by weevils. Tested insecticides were not successful in preventing weevil attack. Sathish *et al*. (2006) observed that most of the seeds of *Dipterocarpus indicus* were also infested by insects. *Nanophyes* (Coleoptera, Apionidae), a similar weevil has been reported to infest seeds of dipterocarps in Malaysia (Daljeet-Singh, 1974). *N. shoreae* a fruit predator in the South-East Asian rain forest was reported by Toy and Toy (1992).

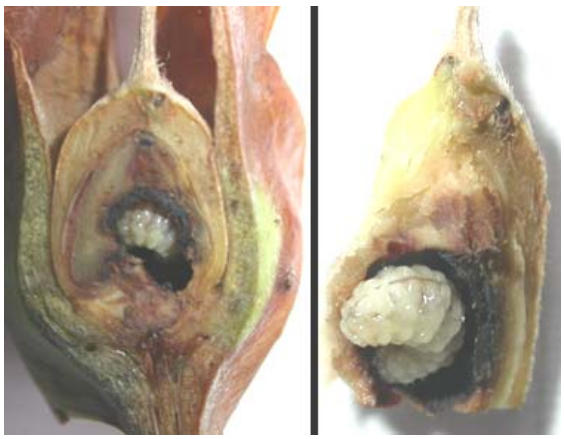

Figure 16. Longitudinal sections of the nut of submature fruits of *Dipterocarpus bourdillonii* showing the insect larvae.

Flower damage by insects is roughly 10-12%. The flower and fruit damage together account for 40-45% damage of the reproductive units.

Pre-mature degeneration of embryos in D. bourdillonii

Dissections conducted 10-20 days prior to dispersal in open pollinated fruits have shown that around 61% of fruits have decayed embryo but without any trace of insect infestation. The decay of embryo was complete and was converted to a brown amorphous mass within the seed (Fig. 17 B).

Growth of the fruits and embryo development in apomicts and self pollinated flowers were normal as in cross pollinated ones till 10-20 days before maturity, when the embryos start decaying. In fruits derived from apomixis and self pollination, drying of fruit wall and browning of embryo inside the ovary was simultaneous. In cross pollinated fruits, viable embryos (Fig. 17 C) and decayed embryos were found in the ratio 4:1.

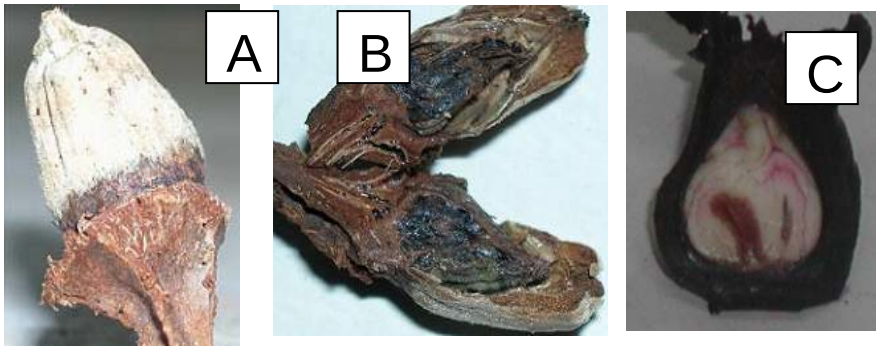

Figure 17. *Diptero-carpus boudillonii*. Fig. 17 A. A nut excised from the calyx cup. Fig. 17 B. A mature fruit split open to show the decaying embryo. Fig. 17 *ca.* A viable embryo inside the fruit.

Embryos inoculated in Potato Dextrose Medium revealed the presence of pathogenic fungi, *Fusarium* sp., *Phomopsis* sp. along with non pathogenic fungi like *Alternaria* sp., *Phoma* sp., *etc.,*. About 10% of the plates contained *Phomopsis* and 70% plates contained *Fusarium*. Whether the fungal infection was primary or post-degeneration of the embryo is unclear.

Fruit dispersal: Supported by the two alar calycine wings, *Dipterocarpus bourdillonii* is basically entomophilous. Low wind currents take the winged fruits to shorter distances. Studies in *Shorea contorta* in the Philippines indicated that fruits were carried as far as 20-40m from the mother tree (Tamari and Jacalne, 1984).

Results from germination experiments

The details on germination of the fruits produced through various experiments are given in Table 13.

Table 13. Germination of fruits from controlled pollination experiments.

No.	*Type of Pollination*	*Fruits sown*	*Insect infested*	*Germi-nated*	*Germination (%) **
1.	Cross pollinated (artificially)	37	11	13	35
2.	Self pollinated (artificially)	51	10	—	—
3.	Apomixis (Emasculated and net covered)	20	3	—	—
4.	Net covered (without control pollinations)	16	—	—	—
5.	Open pollinated	513	140	15	3

* Percentage of uninfected fruits; figures rounded off.

Fruits produced through control pollinations: None of the fruits derived from the following experimental set ups germinated: (a) fruits derived from apomictic development (20 fruits). (b) fruits developed from flowers covered for exclusion of insects (14). (c) fruits set through artificial self pollination (51 fruits). Interestingly enough, 50 % (13/26) healthy, non-infested fruits developed from cross pollination germinated.

Fruits produced through open pollinations: The percentage of germination obtained from different experiments varied. In a germination trial containing 513 seeds collected from five trees, 15 seeds germinated. The un-germinated 140 fruits (27%) were found infested by insects. Thus, out of the healthy 373 seeds, the actual germination percentage obtained was 4%. In two other trials containing 600 and 500 seeds each, 1.5 and 2.4 percent germinations were obtained respectively.

In many tree species including Dipterocarps a large proportion of developing fruits abort midway through development. Inbred fruits abort through late-acting self incompatibility. These seeds act as seed predator sinks and thereby increasing the survival probabilities of out-crossed seeds (Ghazoul and Satake, 2009).

Fruit dissection studies of open pollinated flowers showed that the percentage of fruits with viable embryo at maturity was only 7.4% and the germination rate only 4%. Taking account of the 50% germination in seeds derived from artificial cross pollination, the rate of cross pollination in the natural condition would be *ca.* 8%. This also points to the lack of enough pollinators. In a study from Indonesia, Lillesoe (1996) reported that logging of non-commercial species in a forest area has reduced the population size and efficiency of specific pollinators, which in turn has severely reduced the reproductive capacity of some of the commercially very important *Dipterocarps*.

In addition to the low seed germination, post-germination mortality from grazing is also significant.

Destruction of forests may have many reciprocal effects on its biodiversity. These include changes in: (a) competitors among plant species, (b) alteration in pollen and seed dispersal patterns, and (c) contraction of effective population size (Nason *et al.*, 1997). As most tropical trees are animal pollinated (Bawa *et al.*, 1985; Bawa, 1990), changes in plant density and the destruction of pollinator habitats may have critical effects on the reproductive success of individual trees in fragmented landscapes (Aizen & Feinsinger, 1995).

To sum up, the main reasons for the low population size of *D. bourdillonii* are: (a) logging in the past, (b) self incompatibility, (c) insufficient population of pollinators, (c) insect infestation of flowers and fruits, and (d) human interventions in the form of grazing.

Propagation studies in *D. bourdillonii*

Seed propagation in D. bourdillonii

Two seed germination trials conducted in nursery beds resulted in complete failure; only one seed germinated out of the hundreds of seeds sown (Fig. 18).

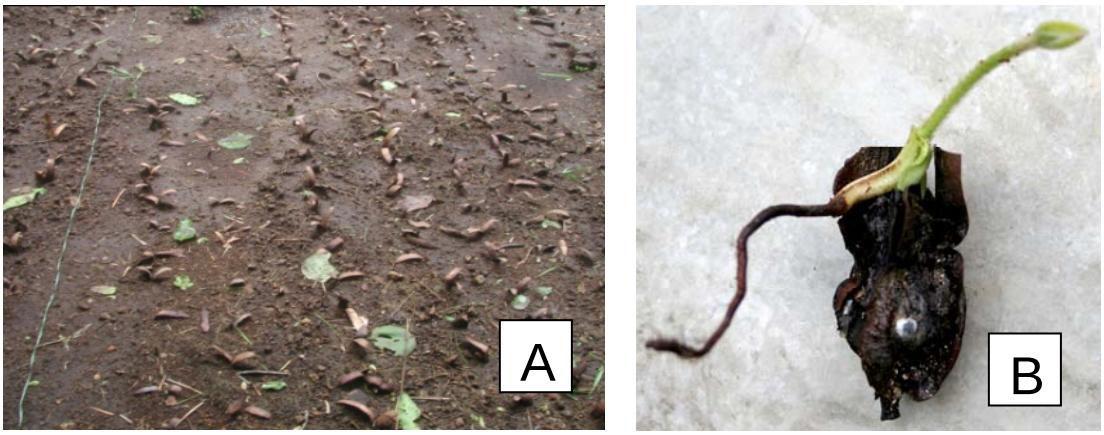

Figure 18. *Dipterocarpus bourdillonii*. Fig. 18 A. Germination trial. Fig. 18 B. A geminating seed.

Germination studies conducted by the Kerala Forest Department have repeatedly reported total failures. Nevertheless, the small population of existing juveniles indicates that a small percentage of the fruits/seeds do contain viable embryos. In an earlier study we had obtained a few seedlings in germination trials carried out with seeds from Achenkoil.

Air layering using IBA in D. bourdillonii

Air layering was attempted for different treatments of IBA (400, 600, 3000 and 5000 ppm) and vermiculite as the medium (Fig. 19). Callus formation was observed in the branches but root formation could not be obtained.

Figure 19. Air layering in *D. bourdillonii.*

Rooting of stem cuttings of D. bourdillonii *using IBA*

Branch cuttings 5-8 cm thick were treated with varying concentrations of IBA to induce rooting. The cuttings were bagged in soil and sand mix (2:1 ratio) and kept in the mist (Fig. 20). The results are provided in Table 14.

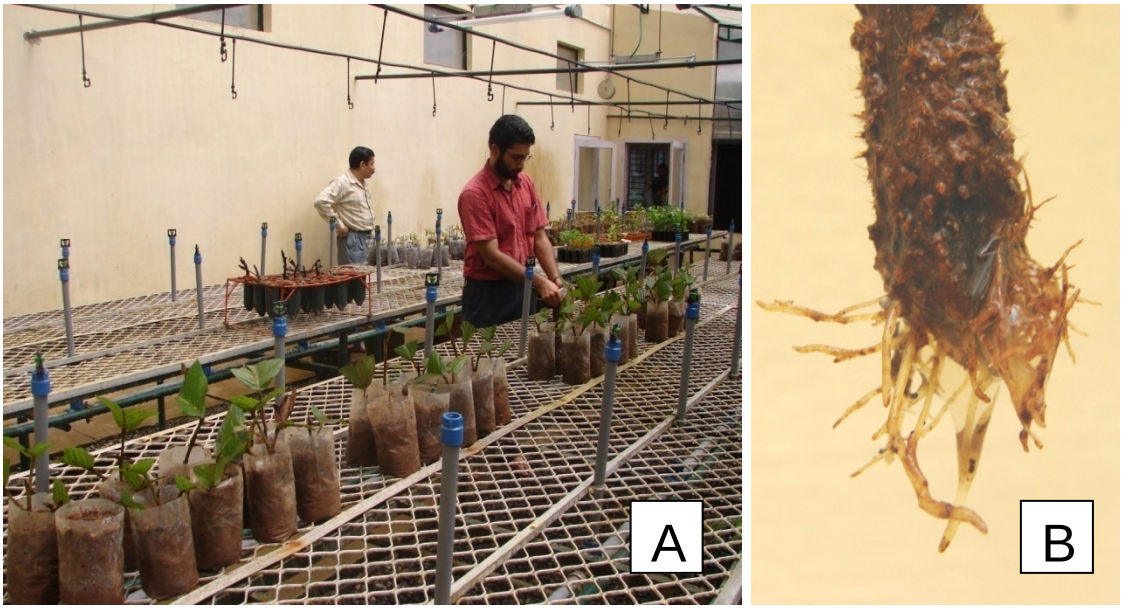

Figure 20. *Dipterocarpus bourdillonii*. Fig. 20 A. Rooting of stem cuttings in the mist chamber. Fig. 20 B. Roots of a rooted cutting.

Lower concentrations of IBA, 400 ppm and 600 ppm although initiated some callusing, no root formation took place and all the cuttings eventually died off. However, cent percent rooting of stem cuttings was obtained for the treatments, 6000 and 8000 ppm. About 60 % survival of the shoots was seen after transfer to nursery. Nevertheless, when the experiment was repeated during different seasons, the results were not consistent (Table 14).

Table 14. Response of *D. bourdillonii* sprouts to treatment with varying concentrations of IBA.

Sl. No.	*Treatments*	*Result (%)*					*Dates*
		Total	*Callus**	*Rooting**	*Dead*	*Survival*	
1	400 ppm	10	30	-	100	-	Aug. 2007
2	600 ppm	10	20	-	100	-	Aug. 2007
3	6000 ppm	10	100	100	40	60	Aug. 2007
4	8000 ppm	10	100	100	60	40	Aug. 2007
5	8000 ppm	10	40	20	60	40	Sept. 2007
6	8000 ppm	10	30	10	70	30	Oct. 2007
7	8000 ppm	10	10	-	80	40	Nov. 2007
8	9000 ppm	10	30	10	70	40	Nov. 2007
9	10000 ppm	10	40	10	60	30	Sept. 2007
10	10000 ppm	10	40	10	60	20	Oct. 2007
11	10000 ppm	10	30	-	70	20	Nov. 2007

* Percentage of rooted cutting

Shoot multiplication from axillary buds in D. bourdillonii

Nodal explants when inoculated into Woody Plant Media supplemented with Benzyl Adenine (1.5 ppm, 3 ppm and 6 ppm), Kinetin (3 ppm) and Benzyl Adenine/Kinetin (3 ppm/3 ppm), the explants with the hairy covering removed responded better than those with the hairy covering left intact.

Better contact of tissues with the media was probably the reason. Nodal explants showed no sprouting response in any of the media. The extreme shoot tip with the bracts removed showed the best response. A pre-treatment with antibiotics (200 ppm Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1 % Bavistin) before surface sterilisation with 0.1% $HgCl_2$ gave the best results. Sprouting was obtained on all BAP containing media but growth was extremely slow and no multiple shoots were obtained even when sprouting of axillary bud occurred in a few of the explants (Fig. 21).

Contamination rate of the cultures was very high resulting in low survival percentage. The different surface sterilization treatments given and the results obtained are given in Table 15.

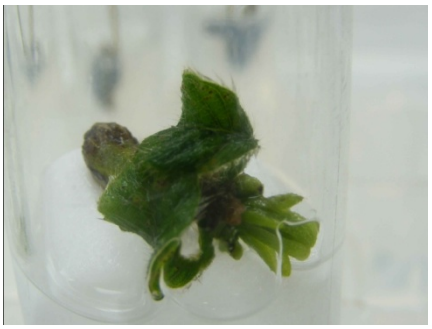

Figure 21. *Dipterocarpus bourdillonii.* Performance of nodal explants/apical buds in culture media.

In vitro *regeneration from immature embryos in* D. bourdillonii

In vitro *culture of D. bourdillonii from immature embryos:* Embryos from mature fruits were used as explants for *in vitro* regeneration through organogenesis. Except for one collection, the heavy incidence of insect larve in the fruits made it difficult to get live and sterile explants.

Embryos isolated from the immature fruits were cultured on MS media supplemented with different auxins (NAA, 2,4-D, Picloram and Dicamba at 1 and 3 ppm) but no morphogenesis has been observed in any of the experiments (Fig. 22).

Table 15. Sterilization treatments given to nodal explants of *D. bourdillonii*

No.	*Treatments*	*Contamination (%)**			*Survival (%)*
		Bacterial	*Fungal*	*Total*	
1.	Cefotaxime (200 ppm) + Ampicillin (200 ppm) + 0.1 % $HgCl_2$	78	97	100	-
2.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 0.1 % $HgCl_2$	26	87	100	-
3.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 0.1 % $HgCl_2$ + PPM[3]	7	83	89	11
4.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1% Bavistin (105 mins) + 0.1 % $HgCl_2$ (15 mins)	9	83	89	11
5.	200 ppm Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1 % Bavistin ; Exalin (0.1%) + $HgCl_2$	10	81	87	13

* Percentage of explants affected.

When embryo axes were cultured on BAP containing medium sprouting of axillary buds and multiple shoot formation was observed (Fig. 21) . Development was however very slow.

It is concluded that the species is recalcitrant to *in vitro* culture as has been observed earlier by researchers in other Dipterocarps.

Figure 22. Embryo ex-plants of *Dipterocarpus bourdillonii* in culture.

Species recovery through augment planting of *D. bourdillonii*

Initial transplantation experiments in D. bourdillonii

As enough seedlings could not be obtained from germination trials and micro- and macro- propagation efforts, 10 wildlings of *D. bourdillonii* were collected and transplanted underneath the moist deciduous forest patch at KFRI, in order to test the feasibility of field transplantation (Fig. 23 A). These wildlings have established well and have grown to a height of about 60 cm within one year.

The seedling survival and growth performance at the end of two years after transplanting into the recovery sites are discussed below.

Survival of D. bourdillonii *seedlings at Urulamthanni (Site-1, In-situ):* The seedlings showed *ca.* 40% survival. The mean height of seedlings was 54 cm and the maximum height recorded 93 cm (Figs. 23 B & 24). Human and wildlife interventions are the major causes for seedling mortality in the site. Browsing by domestic cattle and wild animals (Sambar), removal of the ground flora for fodder and green manure, encroachment of forest land, *etc.,* seem to have affected the seedling survival. The leaves of the natural and planted seedlings of the species were found infested severely by insects.

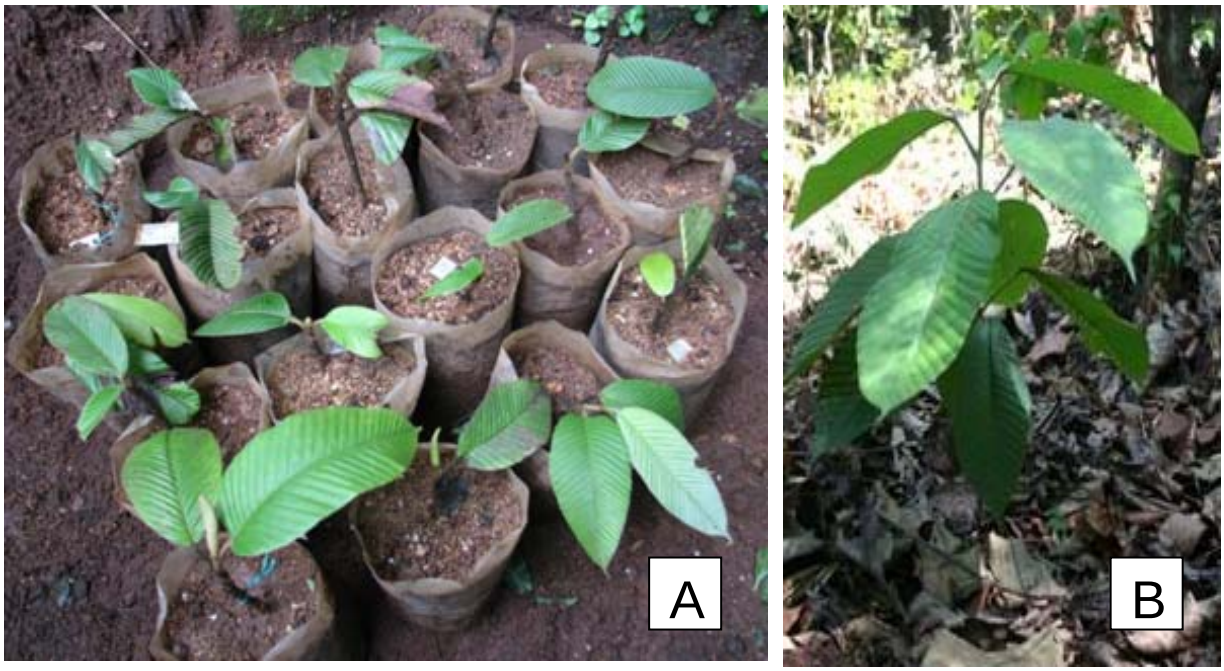

Figure 23. *Dipterocarpus bourdillonii.* Fig. 23 A. Wildlings collected for transplantation. Fig. 23 B. Transplanted seedling after one year.

Figure 24. Restoration of *Dipterocarpus bourdillonii* at Urulamthanni.

Survival of D. bourdillonii *seedlings at Meenvallam (Site-2, In-situ;* Fig. 25): This site recorded 54% survival for the seedlings. The average height recorded for the seedlings was 70 cm; maximum height observed was 186 cm. Overgrowths of weeds was found to have suppressed seedling growth. Elephant trampling also has contributed to the mortality of seedlings in the site.

Figure 25. Restoration of *Dipterocarpus bourdillonii* at Meenvallam. Planting sites, planted seedlings and display pigds.

Figure 25. Restoration of *Dipterocarpus bourdillonii* at Meenvallam (Continued).

Survival of D. bourdillonii *seedlings at Valayamchal (Aralam WLS; Site-3, In-situ*; Fig. 26)*:* This site recorded poor survival rate, *ca.* 31%. The site also recorded a poor growth rate, 34 cm but some individuals recorded a maximum height growth up to 72 cm. Browsing by Sambar, damage by wild elephants are regular in the area affecting survival of the planted seedlings. Another major threat to the saplings was *Sahyadrassus malabaricus,* an insect pest, a stem borer, which was the major cause for the poor survival and growth of the seedlings.

Figure 26. Restoration of *Dipterocarpus bourdillonii* at Aralam Wildlife Sanctuary. Planting sites and the saplings.

Survival of D. bourdillonii *seedlings at Azhuthakadavu (PTR West; Site-4, In-situ;* Fig. 27): This site also recorded a poor survival rate of 30%. The mean height attained by planted seedlings was 41 cm and the maximum height recorded, 138 cm. Damage caused by the stem borer, *S. Malabaricus,* was quite serious; *ca.* 50% seedlings were found infested by the stem borer. Browsing by the Sambar and damage by wild pig were other factors contributing to mortality.

Figure 27. Restoration of *Dipterocarpus bourdillonii* at Azhuthakadavu. Planting sites and planted seedlings.

Survival of D. bourdillonii *seedlings at Ponnampara (Site-5, In-situ):* Ponnampara recorded the lowest survival rate of 12%. The growth rate was also extremely low; the average height of planted seedlings after 2 years were only 25 cm, displaying stunting. Heavy weed growth in the area suppressing the seedlings was responsible for this. The maximum seedling height recorded however was 66 cm. About 40% seedlings were found killed by the stem borer, *S. Malabaricus*. Elephant trampling and guars also seemed to have affected the site and the survival of the seedlings.

Survival of D. bourdillonii *seedlings at Velithode (Site-6, In-situ;* Fig. 28): The site displayed a very low survival rate of seedlings, *ie*, 15%. The average seedling height two years after planting was 79 cm and the maximum height recorded was 175 cm. The stem borer, *S. malabaricus* was the major cause of seedling mortality, but exuberance of weed growth retarded the growth rate. Damage from wild animals such as elephants, gaur, wild pig, also affected the survival.

Figure 28. Restoration of *Dipterocarpus bourdillonii* at Velithode. Planting sites, Weed overgrowth,. Weeding and an insect infected seedling.

Survival of D. bourdillonii *seedlings at Kulamavu MPCA (Site-7, In-situ;* Fig. 29)*:* Twenty-five percent seedlings survived at the end of two year. The site had a low mean seedling height of 25 cm, and the maximum recorded was 37 cm. The stem borer *S. malabaricus* was the major mortality factor; *ca.* 50% of the seedlings was found killed this way. Browsing by Sambar also contributed to seedling loss.

Figure 29. Restoration of *Dipterocarpus bourdillonii* at the MPCA, Kulamavu. Planting sites, planted seedlings and display pigds.

Survival of D. bourdillonii *seedlings at Kariponi* (*Site-8, Ex-situ;* Fig. 30): This site had a vegetation different from the natural vegetation of the species. It also did not enjoy any kind of protection that was available for the sites 9, 10 and 11. Despite these, survival of planted seedlings was not bad. Forty-five percent survival of the planted seedlings was recorded at the site. The average seedling height obtained was 72 cm and the maximum, 148 cm. Overgrowth of weeds and unexpected fire in the adjacent areas has caused severe damage to the seedlings. Damage by wild elephants, sambar, was also notable.

Figure 30. Restoration of *Dipterocarpus bourdillonii* at Kariponi. Planting sites, planted seedlings, weed infestation and seedling damaged by stem borer.

Survival of D. bourdillonii *seedlings at KFRI Arboretum, Peechi* (*Site-9, Ex-situ*; Fig. 31): The seedlings offered a high rate of survival, 78%. The average seedling height was 74 cm and the maximum height recorded was 238 cm. The site being a moist deciduous forest, a few seedlings wilted in the dry months.

Figure 31. Restoration of *Dipterocarpus bourdillonii* at the KFRI Arboretum, Peechi. Planted seedling and growth monitoring of the planted seedlings.

Survival of D. bourdillonii *seedlings at KFRI Subcentre, Nilambur (Site-10, Ex-situ):* The survival percent of the seedlings obtained was 60%. The averages seedling height was 50 cm and the maximum 145 cm. Shade of the canopy was found limiting the growth. A few seedlings wilted during the summer months.

Figure 32. Restoration of *Dipterocarpus bourdillonii* at the KFRI Subcentre, Nilambur. Planting site and seedlings protected with fishnet fence.

Survival of D. bourdillonii *seedlings at KFRI-FRC, Velupadam (Site-11, Ex-situ*; Fig. 33)*:* A somewhat high seedling survival rate was observed: 64%. The mean seedling height was 72 cm and maximum 214 cm. Browsing by the spotted was prevalent and was the major threat to the survival seedlings.

Figure 33. Restoration of *Dipterocarpus bourdillonii* at the Field Research Centre of KFRI at Velupadam. Planted seedlings and growth monitoring.

Stem borer infestation on D. bourdillonii *seedlings*

The insect *Sahyadrassus malabaricus* (Moore) (Lepidoptera, Hepialidae) is a moth which happens to be a stem borer and is one of the major threats in most augment planted sites. Seedling mortality due to the pest ranged up to 60%. The larva of the insect bores into the sapling stem and tunnels through the pith. The opening of the tunnel is covered by a thick mat of wood particles woven with silk, underneath which the larva feeds on the callus tissue that grows (Fig. 34).

The early larval instars survive on weedy ground vegetation and eventually migrate and feed on tree saplings. The larval population gets established in August and is replenished by subsequent instars. Forty species of woody shrubs and trees, including many plantation species such as teak, eucalypt, *Gmelina, Anthocephalus a*nd *Albizia* are susceptible to the moth. *Trema orientalis* and *Clerodendrum viscosum* are two of the most favoured hosts (Nair, 1982). The insect though confined to Peninsular India is widely distributed there and seedlings and saplings of *D. bourdillonii* are affected by the insect.

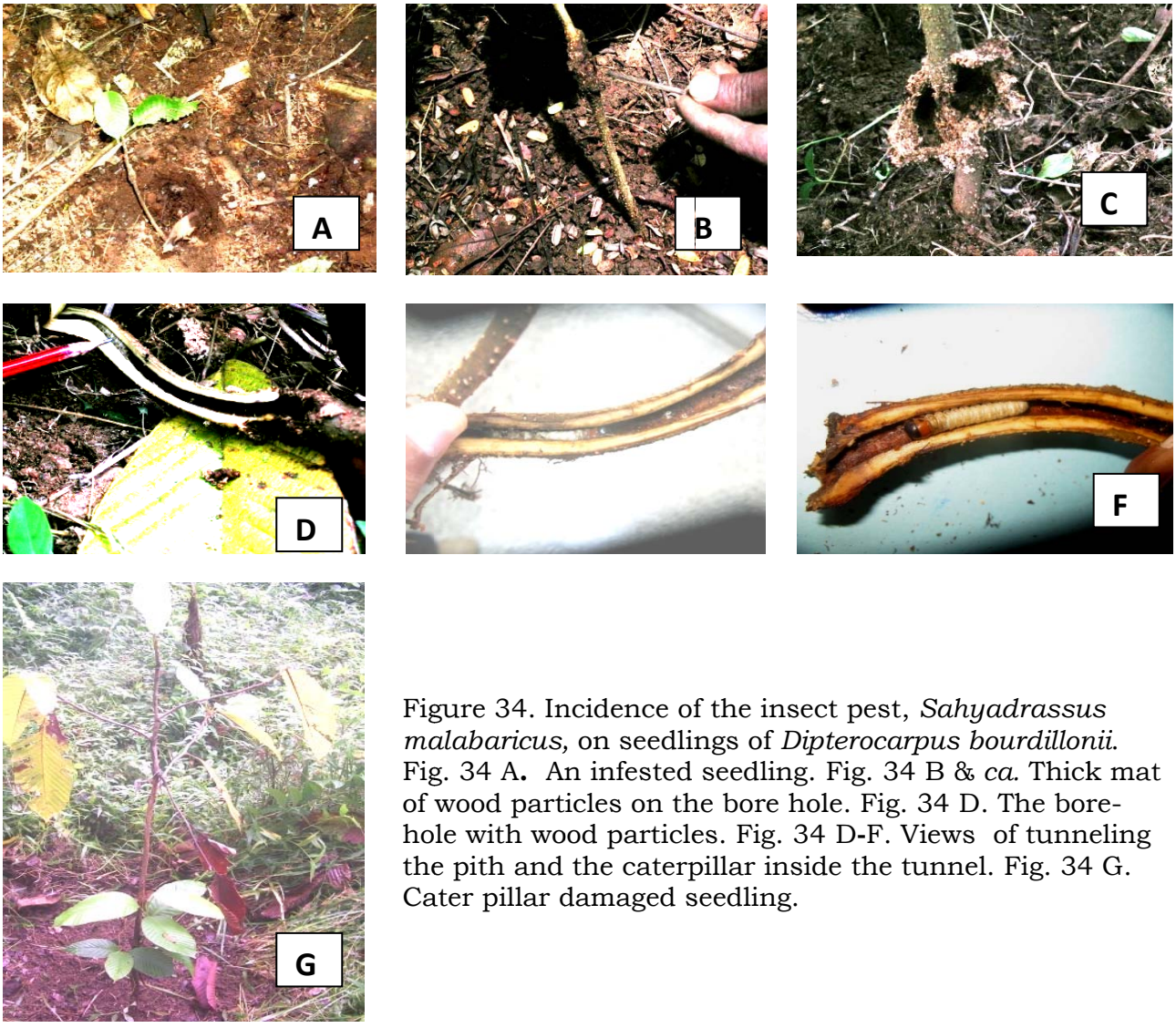

Figure 34. Incidence of the insect pest, *Sahyadrassus malabaricus,* on seedlings of *Dipterocarpus bourdillonii.* Fig. 34 A**.** An infested seedling. Fig. 34 B & *ca.* Thick mat of wood particles on the bore hole. Fig. 34 D. The bore-hole with wood particles. Fig. 34 D-F. Views of tunneling the pith and the caterpillar inside the tunnel. Fig. 34 G. Cater pillar damaged seedling.

Survival, mortality and growth of augment planted stocks of D. bourdillonii

Table 16 describes the observed survival rates, mean and maximum height of saplings recorded 2 years after planting.

In general, seedlings planted *ex-situ* have scored maximum survival performance compared to that of *in situ.*

The highest survival rate recorded for any of the *in situ* sites was 54%, at Meenvallam. A few of the sites recorded very low survival rates also, 12% and 15%. The low rates of survival owe mainly to mortality factors contributed by many factors interfering with the seedlings. The major constraints *in situ* are pest infestation, suppression by overgrowing weed growth, trampling by wildlife, grazing by wildlife. A small percent of the planted seedlings perished due to excessive rain and summer droughts.

Table 16. Survival, growth and factors affecting mortality of the restoration plantings of *D. bourdillonii.*

Site no.	*Planting sites*	*Seedlings planted*	*Survival after 2 yrs (%)**	*Mean height (cm)*	*Max. height (cm)*	*Factors affecting mortality*
I. In-Situ sites						
1.	Urulamthanni	100	40	54	93	Grazing, Sambar, green manure collection
2.	Meenvallam	50	54	70	186	Weeds, Elephants
3.	Kulamavu MPCA	100	25	25	37	Stem borer, Sambar
4.	Valayamchal	150	31	34	72	Sambar, Stem borer
5.	Azhutha kadavu	250	30	41	138	Stem borer, Sambar
6.	Ponnampara	250	12	25	66	Stem borer, Elephant, Gaur
7.	Velithodu	150	15	79	175	Weeds, Elephants, Gaur, Wild pig
8.	Karipponi	150	45	72	148	Weeds, Fire, Elephants, Sambar, Drought
II. Ex-Situ sites						
9.	KFRI-Arboretum	50	78	74	238	
10.	KFRI-Subcenter	50	60	50	145	
11.	KFRI-FRC	50	64	72	214	Spotted

* Rounded off to whole digits.

The three *ex-situ* sites (KFRI Arboretum, KFRI Subcenter and KFRI-FRC) which had a garden environment and the associated protection, the survival rate was high, ranging between 60 and 78.

Evidently because of the protection, care and manuring the saplings receive in *ex-situ* garden sites, both mean height and maximum height also proved to be high there. On the whole, both the above parameters were found low in *in-situ* sites.

Dipterocarpus bourdillonii is a tree belonging to the low level evergreen forests. Nevertheless, saplings planted in the degraded forest (Karipponi, Site 8) provided 45% survival without any additional protection and input. The seedlings also had good growth (mean height after 2 years: 72 cm and maximum height of 148 cm). This is indicative of the fact that restoration of the species is feasible, even in degraded evergreen forests, where the species has disappeared or where the population has gone low.

2. *Humboldtia bourdillonii*

The genus *Humboldtia* comprises of six species, all confined to Southern Western Ghats, except one extending to Sri Lanka (Sanjappa, 1986; Sasidharan, 2004). Distributed in the Peermade Ghats, *H. bourdillonii* grows to a height of 15 m. Like *Dipterocarpus bourdillonii*, *H. bourdillonii* is also a ripicole species found along streamlets and watercourses of the evergreen forest at about 900 m (Balasubramanyan *et al.*, 1985).

A botanical description of the species is given below. Photographs of trees, leaves, flowerws, floral parts and seeds are given in Fig. 35 & 36.

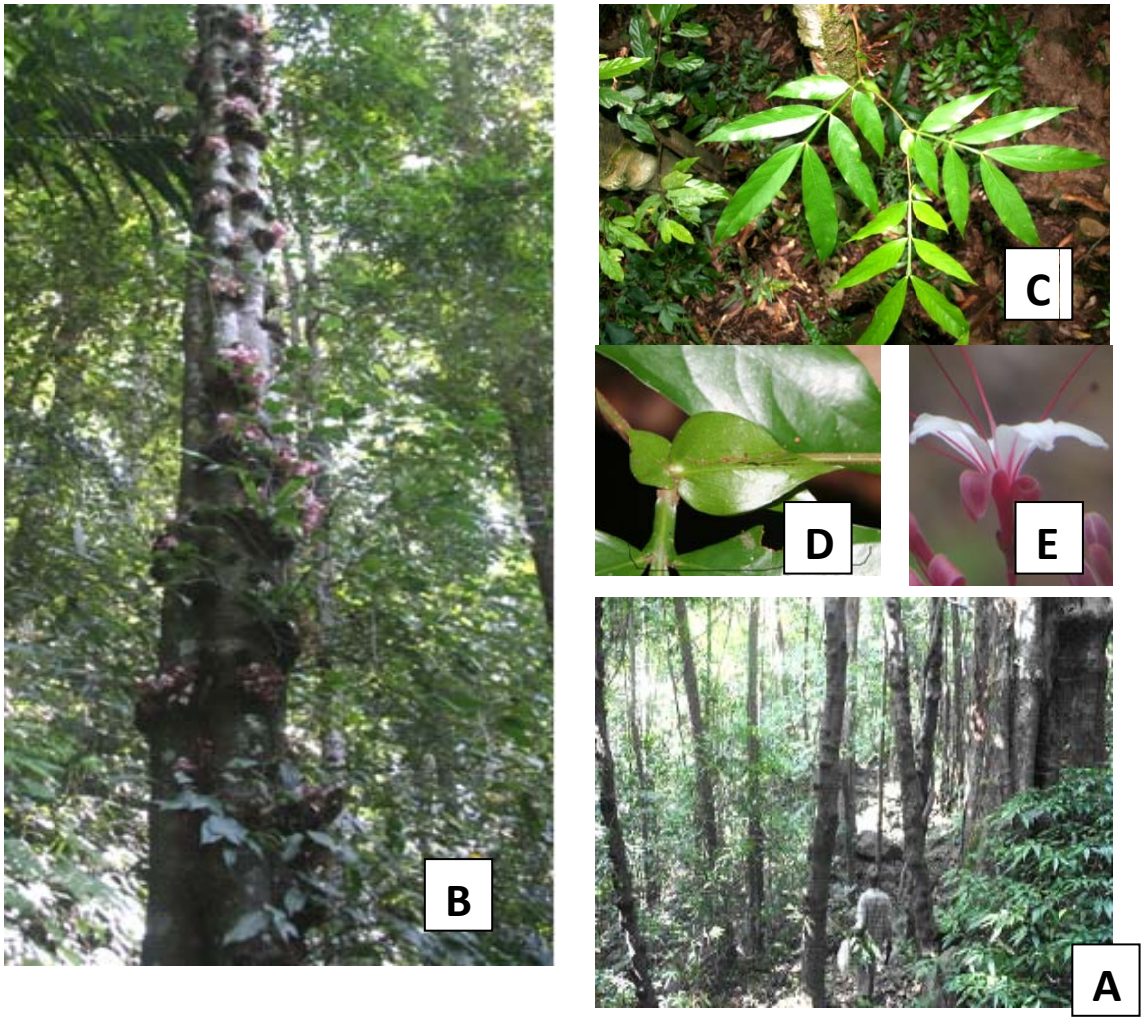

Figure 35. *Humboldtia bourdillonii.* Fig. 35 A. An evergreen forest stand with many trees of *H. bourdillonii.* Fig. 35 B. The bole of a mature tree showing the characteristic cauliflory. Fig. 35 *ca.* A leaf. Fig. 35 D. Stipule enlarged.

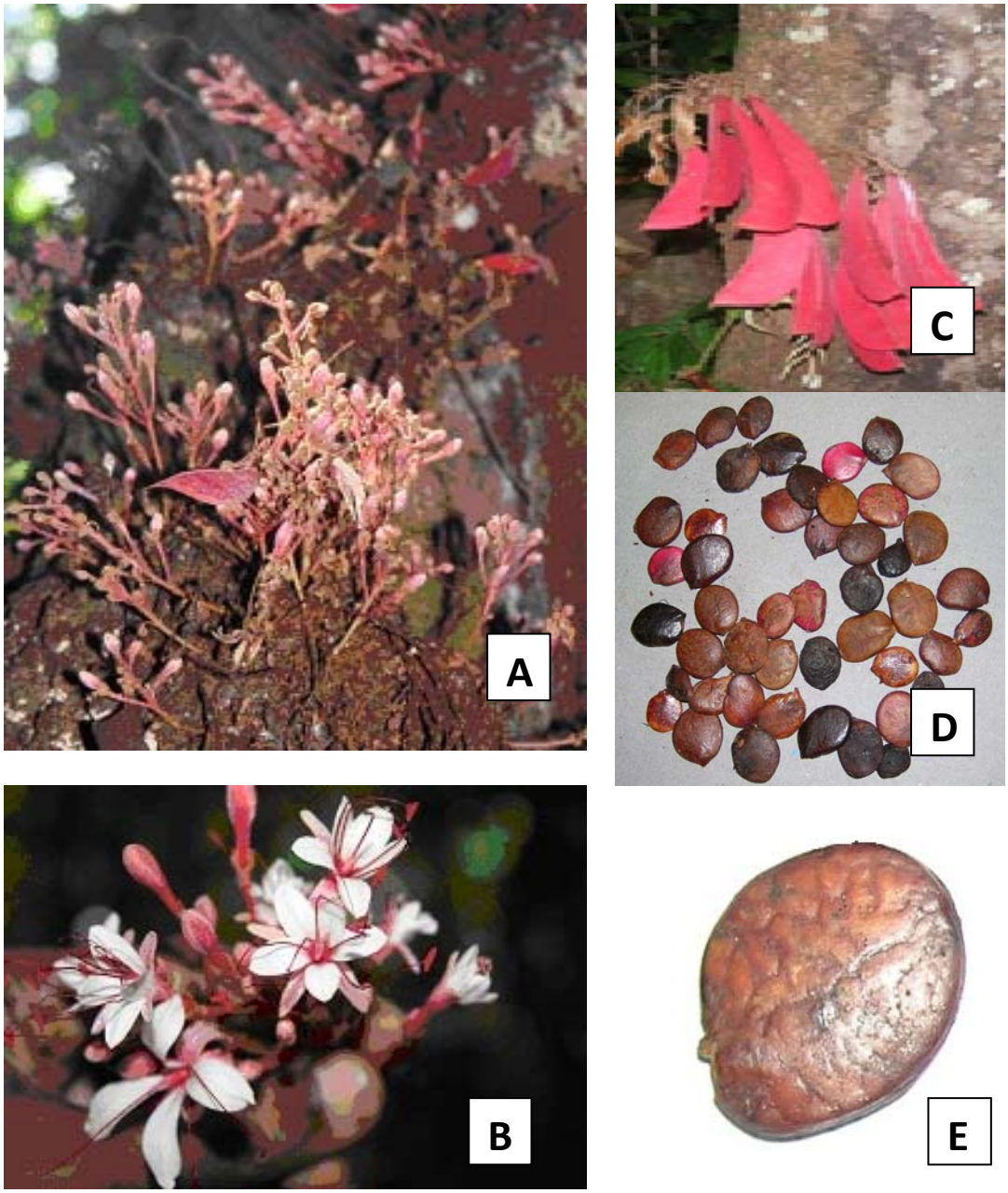

Figure 36. *Humboldtia bourdillonii.* Fig. 36 A. The tubercles on the stem showing the copious production of inflorescences. Fig.36 B. An inflorescence. Fig. 36 *ca.* Open flower in side view. Fig. 36 D. Submature fruits. Fig. 36 E. Mature seeds. Fig. 36 F. A seed enlarged.

Tree: Medium sized, growing 25 m high, occupying sub-canopy strata (Tier II), bole - 65 cm in diam. *Branchlets:* solid, glabrous. *Bark:* reddish-brown, smooth. *Stipules:* ovate, acute, prominently veined, glabrous, appendages falcate, rounded, veined, persistent. *Leaf-rachis:* winged between the petiolules, wings auricled; *leaflets:* 2-4 pairs, sub-sessile, oblong or linear lanceolate, acuminate, base obtuse or truncate, glabrous; lateral veins 6-8 pairs, reticulations faint. The leaves associated with inflorescence are small with 2 pairs of leaflets. Cauliflorous- showing both trunkiflory and branchiflory, producing

flowers on stem as well as in all branches. *Inflorescence:* a mixture of cyme and raceme, produced on tubercles brown tomentose. *Bracts:* ovate, acute; bracteoles connate, splitting unequally, lobes ovate-oblong, tomentose, gland-dotted. Calyx tube oblong, tomentose, crimson. Petals 5, unequal, oblong or obovate, white with pink veins. Stamens 5, filaments pilose; athers oblong. Ovary 4-6 ovuled; style long with a tuft of hairs at base, otherwise glabrous; stigma papillose. Pods oblong, velvety brown pubescent, bright red or crimson coloured, prominently veined. *Flowering:* Middle of November to April. *Mature seeds:* available from March to May.

Population ecology of *H. bourdillonii*

Distribution

The results of simulation of the distribution of the species using DIVA-GIS and GARP softwares have not been helpful in identifying newer areas of distribution of the species. Hence field investigations for finding out the distribution and estimating the population size of the species was carried out, based on the data available with the herbarium specimens and utilizing the field experience of the local people in Periyar Tiger Reserve.

Through repeated field investigations (Fig. 37 & 38), seven populations of the species have been located. All these populations inhabit the Periyar Tiger Reserve (Idukki Dt.). They were discrete and discontinuous, although occupied nearby areas. The species, which was thought to be riparian as its kin species, *H. vahliana*, proved to be non-riparian. The species occupied areas in between streamlets and the ground rich in boulders. How the populations got distributed as discontinuous patches, is yet to be known.

This species is seen in the wet evergreen forest of Arjunan Kotta, Poonkavanam area of Pampa valley in the Peermade plateau. Contrary to the earlier notion that the species is a riparian element, it is seen between 450m and 800m altitude, along or within the flood area of the watercourse to 10-60m away from it. This species prefers medium to heavy slope with boulders.

Fig. 37. Exploration for *H. bourdillonii* in the Periyar Tiger Reserve.

Sample survey of the species showed a patchy distribution; seven discrete patches were identified in the Arjunan Kotta-Poongavanam-Chittamada Ar region (Sabarimala Thodu draining Poongavanam, Uppupara and Arjunan Kotta). Individual patch size varied from 0.1 ha to 2 ha. The largest patch was having an area of almost 2 ha and it being a narrow strip traversed a length of 1.2 km. The patches were found distributed at distances ranging from 177 m to 638 m from each other. The area of occurrence of the species in the above two areas was found to be approximately 2 km^2 and the area of occupancy (area sampled plus non-sampled area) 0.07 km^2 (Fig. 39).

Figure 38. Exploration for *H. bourdillonii* in the Periyar Tiger Reserve. Fig. 38 A. The Sastha temple at Sathram and the evergreen forest stand behind it. Fig. 38 B. The touring party.

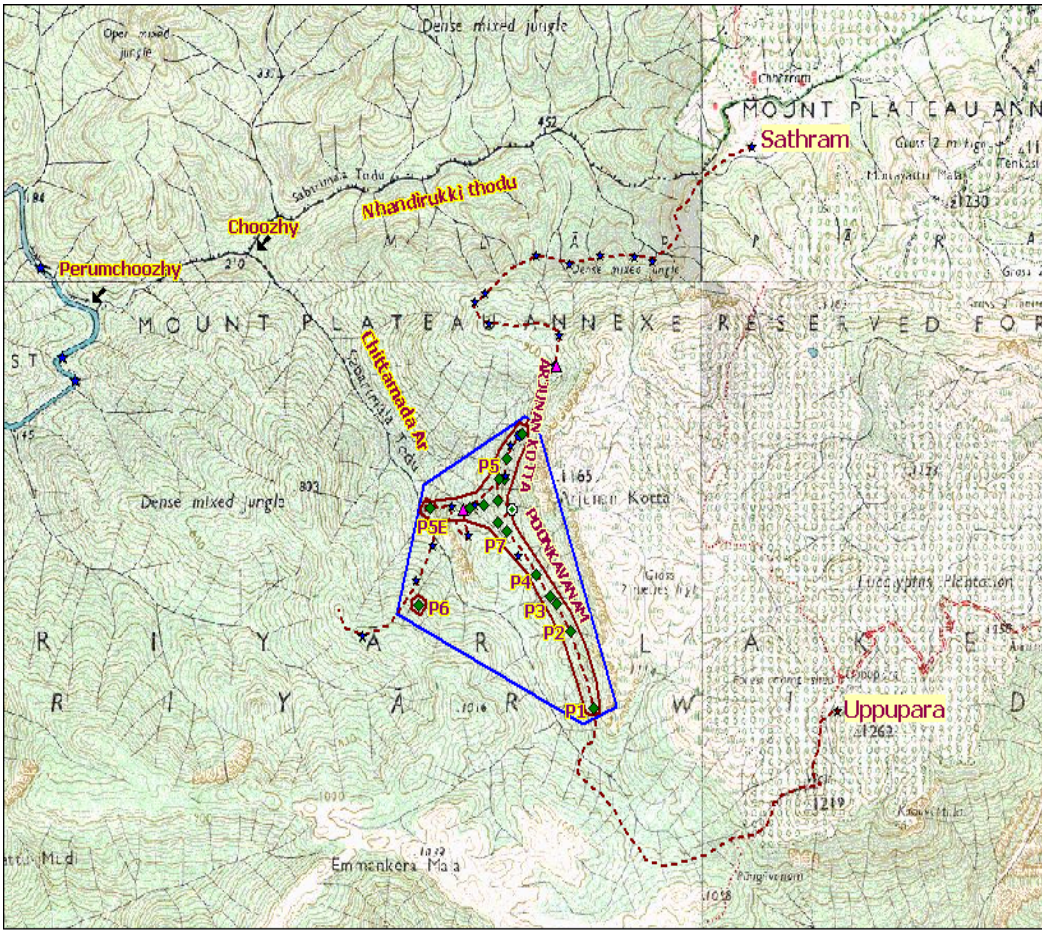

Fig. 39. Distribution of *Humboldtia bourdillonii* in Periyar Tiger Reserve showing the area of occurrence and area of occupancy.

Table 17. Community structure of the evergreen forest in which *H. bourdillonii* is found (condensed table).

No.	*Species*	*D*	*BA*	*RBA*	*IVI*	*RIVI*	*CRIVI*
1	*Humboldtia bourdillonii*	878	25.0	24.6	68.9	23.0	23.0
2	*Vateria indica*	135	19.8	19.5	30.2	10.1	33.0
3	*Mesua thwaitesi*	95	16.3	16.1	25.0	8.3	41.3
4	*Palaquium ellipticum*	133	7.7	7.6	18.2	6.1	47.4
5	*Drypetes confertiflora*	100	6.4	6.3	15.4	5.1	52.5
6	*Sageraea laurifolia*	168	1.0	1.0	13.2	4.4	56.9
7	*Orophea erythrocarpa*	103	1.7	1.7	10.9	3.6	60.5
8	*Reinwardtio. anamalaiense*	55	1.8	1.7	8.4	2.8	63.3
9	*Mastixia arborea ssp.metzi*	50	2.4	2.4	8.3	2.8	66.1
10	*Drypetes elatus*	18	4.8	4.7	8.3	2.8	68.9
11	*Myristica beddomei*	58	0.7	0.7	6.5	2.2	71.0
12-56	Other 45 species (together)	435	14.0	13.7	87.0	29.0	100.00
	Total	**2213**	**101.5**	**100.0**	**300.0**	**100.0**	**100.0**

RBA – Relative Basal Area; *IVI* – Importance Value Index; *RIVI* – Relative Importance Value Index; *CRIVI* – Cumulative Relative Value Index

Communities supporting Humboldtia bourdillonii

The stands supporting *Humboldtia bourdillonii* were sampled and the structure of the vegetations studied. The structure of the forest where *H. bourdillonii* is found is as given below. The species inhabits evergreen forests of the *Humboldita – Vateria – Mesua - Palquium –* community, a variant of the *Mesua – Cullenia - Palaquium* association (Tables 17, 18).

Table 18. Structure of the stand in the decreasing order of dominance at Arjunan Kotta-Poonkavanam region (Periyar Tiger Reserve).

Species	**D**	**BA**	**RD**	**RF**	**RBA**	**IVI**	**RIVI**	**CRIVI**
Humboldtia boudillonii	523	14.3	40.0	14.3	25.1	79.5	26.5	26.5
Vateria indica	103	18.4	7.9	9.0	32.4	49.2	16.4	42.9
Palaquium ellipticum	78	5.8	5.9	6.8	10.2	22.9	7.6	50.5
Sageraea laurifolia	110	0.7	8.4	9.3	1.2	18.9	6.3	56.8
Drypetes confertiflora	53	3.9	4.0	6.1	6.9	17.0	5.7	62.5
Mesua thwaitesii	45	5.0	3.5	4.7	8.8	16.9	5.6	68.2
Orophea erythrocarpa	68	1.1	5.2	7.2	2.0	14.3	4.8	72.9
Myristica beddomei	40	0.6	3.1	5.4	1.0	9.5	3.2	76.1
Reinwardtio. anamalaiense	30	0.9	2.3	3.6	1.7	7.6	2.5	78.6
Polyalthia fragrans	30	0.9	2.3	3.6	1.5	7.4	2.5	81.1
Knema attenuata	18	1.2	1.3	2.2	2.0	5.5	1.8	82.9
Syzygium spp	10	1.6	0.8	1.4	2.9	5.1	1.7	84.6
Mastixia arborea	18	0.2	1.3	2.5	0.4	4.3	1.4	86.0
Gomphandra tetrandra	20	0.1	1.5	2.5	0.1	4.2	1.4	87.4
Oreocnide integrifolia	15	0.3	1.2	2.2	0.5	3.8	1.3	88.7
Xanthophyllum arnottianum	15	0.1	1.2	2.2	0.2	3.5	1.2	89.8
Syzygium laetum	15	0.1	1.2	2.2	0.1	3.4	1.1	91.0
Baccaurea courtallensis	13	0.3	1.0	1.4	0.5	2.9	1.0	91.9
Dipterocarpus indicus	13	0.1	1.0	1.8	0.1	2.9	1.0	92.9
Aporusa lindleyana	13	0.1	1.0	1.4	0.2	2.6	0.9	93.8
Hiptage benghalensis	10	0.0	0.8	1.4	0.1	2.3	0.8	94.5
Bhesa indica	5	0.4	0.4	0.7	0.7	1.8	0.6	95.1
Dendrocnide sinuata	10	0.1	0.8	0.7	0.2	1.7	0.6	95.7
Syzygium gardneri	5	0.3	0.4	0.7	0.6	1.7	056	96.2
Strombosia ceylanica	8	0.0	0.6	0.7	0.1	1.4	0.5	96.7
Kunstleria keralensis	5	0.1	0.4	0.7	0.1	1.2	0.4	97.1
Cayratia pedata	5	0.0	0.4	0.7	0.1	1.1	0.4	97.5
Cinnamomum malabatrum	5	0.0	0.4	0.7	0.1	1.1	0.4	97.9
Garcinia morella	5	0.0	0.4	0.7	0.1	1.1	0.4	98.2
Aphanamyxis polystachya	3	0.1	0.2	0.4	0.1	0.7	0.2	98.5
Ventilago bombaiensis	3	0.0	0.2	0.4	0.1	0.6	0.2	98.7
Eugenia bracteata	3	0.0	0.2	0.4	0.1	0.6	0.2	98.9
Aglaia barberi	3	0.0	0.2	0.4	0.1	0.6	0.2	99.1
Croton malabaricus	3	0.0	0.2	0.4	0.1	0.6	0.2	99.2
Holigarna nigra	3	0.0	0.2	0.4	0.1	0.6	0.2	99.4
Prunus ceylanicus	3	0.0	0.2	0.4	0.1	0.6	0.2	99.6
Garcinia spicata	3	0.0	0.2	0.4	0.1	0.6	0.2	99.8
Nothopegia colebrookeana	3	0.0	0.2	0.4	0.1	0.6	0.2	100
Total:	**1305**	**56.8**	**100**	**100**	**100**	**300**	**100**	**100**

D=Density, BA=Basal area, RD=Relative density, RF=Relative frequency, RBA=Relative basal area, IVI=Importance value index, RIVI=Relative importance value index (IVI/3), CRIVI=Cumulative relative importance value index.

Population structure of H. bourdillonii

The estimated population size in the sampled areas is as follows: Mature trees 1,008, and Saplings (≥10 cm and <30 cm gbh) 478. The probable total population (including a small patch which was not accessible) is *ca.* 1,100 trees (≥30 cm gbh). Number of trees across the seven populations ranged between 32 and 447.

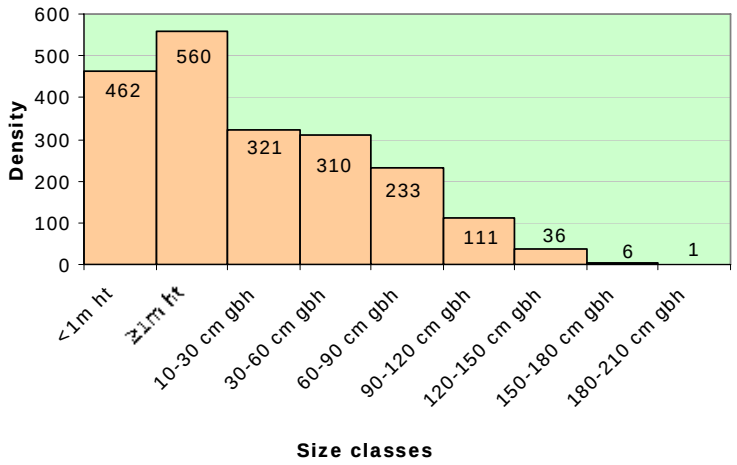

Fig. 40. Population structure of *Humboldtia bourdillonii* in the Periyar Tiger Reserve.

The species is found well represented in all life stages, seedlings, saplings and trees (Fig. 40 & 41). There were 462 seedlings, 560 advanced seedlings, 320 saplings (10-30 cm gbh), and 700 trees (≥ 30 cm gbh) in the total sampled area. The inter-population variation in population traits was not large (Fig. 41).

Reproductive biology of *H. bourdillonii*

Reproductive biological studies on the species were conducted in the natural stands of the species at Arjunankotta (Fig. 42).

Phenology of H. bourdillonii

H. bourdillonii, being an evergreen tree, flushes throughout the year. They flower profusely during November to February. A few trees flower sparingly in late October and late March. A mature tree may flower, twice or thrice, within a flowering season. In the peak period of flowering, most trees produce inflorescence initials synchronously. Inflorescences arise mainly from the tree trunk and to a lesser extent on the branches, particularly in young trees.

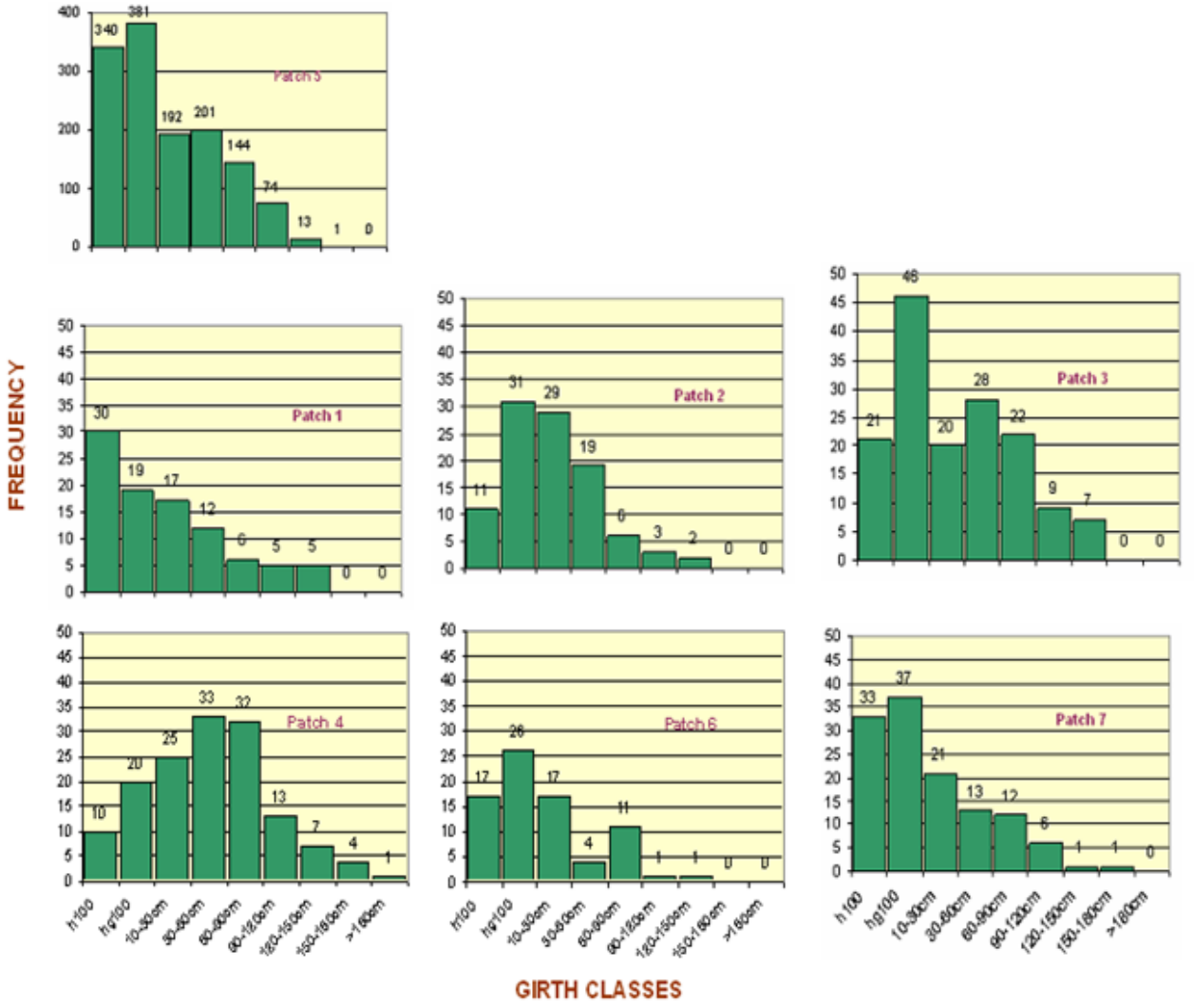

Fig. 41. Stand structure of the seven populations of *Humboldtia bourdillonii.*

A normal inflorescence bud takes about a month to produce mature flowers and the flowers take two and a half months for production of mature pods. Thus, the total reproductive cycle takes *ca.* 3-4 months from inflorescence bud to mature fruit. Mature dry fruits are generally available in late January and early May. Smaller trees usually do not follow this pattern and flower irregularly and produce fruits in accordance. Smallest tree found to produce flowers was with a GBH of 12cm. A good number of flowers tend to degenerate in the acutely damp environment prevalent in the flowering season.

Figure 42. Fieldwork for reproductive biological studies of *Humboldtia bourdillonii*. Fig. 42 A. Camp shed inside the forest at Periyar Tiger Reserve. Fig. 42 B. Breeding experiments being done on a tree of *Humboldtia bourdillonii*.

Floral biology of H. bourdillonii

An inflorescence remains for a week or less. Usually one or two flowers open in a peduncle a day, but rarely all the flowers of the peduncle open simultaneously. Flowers open in the order of their arrangement in the inflorescence. The corolla is free with five petals, white in colour with pink median veins.

The five stamens alternate with the petals, the filaments are hairy and the anthers basifixed and versatile. The pollen grains are off-white, powdery, non sticky and tricolpate. Ovary is superior, monocarpellary, unilocular and is covered by tuft of hairs. The style is 1.6-3.5cm long,

pubescent, slender and dark red and the stigma capitate. The rather long style is coiled in the bud.

The flowers open gradually, starting from late morning to early evening. Complete opening of the flowers take place during 4.30pm to 5.30pm, and the style and filaments get uncoiled. Style and stigma show a spatial separation among themselves before anther dehiscence. Flowers show size variations. Pistils are shorter than stamens. Anthers dehisce longitudinally around 8pm; on dehiscence they change from dark pink to black, liberating creamy white powdery pollen. Stigma secretes a thin layer of exudate at anthesis. Petals fall off 24 hrs after anthesis. Sepals remain a few more days and the stamens and the style dry out after a week. Bracteoles persist in the developing fruits. The ovules are 4-6 in number.

Among the 17 stigmas observed for presence of pollen, 70% of stigmas carried pollen grains. The maximum pollen load was 16 grains per stigma.

Pollination in H. bourdillonii

In open pollinated inflorescences flowers set fruits normally. Inflorescences kept away from insects by encasing in insect nets also set fruits informing that insects are not necessary for pollination. The pollen grains being very light and powdery, they are carried away by wind. Insect pollinators were not seen except in February when a few bees (*Apis indica*) were found visiting some flowers during early morning and evening hours in a few trees. These bees visit the flowers for nectar. They however do not carry pollen as the anthers and stigma are located high and away from the corolla tube. Moreover, flowers set fruits even during months when bees were absent. Thus *H. bourdillonii* appears to be amemophilous. Not many studies are available on other species of *Humboldtia* except *H. brunonis,* where small allodapine bees are said to pollinate the flowers (Shenoy and Borges, 2008).

Pollen viability and stigma receptivity: Indirect pollen viability and stigma receptivity tests using aniline blue have shown that the pollen grains are viable from the time of anther dehiscence up to 36 hrs after flower opening. After 24 hrs of anther dehiscence, pollen viability is above 50%. The stigmas were found receptive from flower opening to 24 hrs post opening. Assessed from pollen germination on stigma, the stigma was found receptive for 24 hrs.

Pollen tube growth: In pollinated pistils pollen grains germinated for 2 hrs after pollination; during this period, pollen tube growth was limited within the stigma. After 4 hrs, it was 4-5mm long in the 30-36mm long pistils, the pollen tube entry into the ovary takes place by

about 12 hr and into the ovules between 13-14 hrs. In longer pistils (40-49 mm), pollen tube entry in to the ovary takes place at about 16 hr and into the ovules between 17-18 hrs (Fig.43). Pollen entry via the micropyle was observed at 20 hr after flower opening. The rate of pollen tube growth was found the same across cross- and self-pollinated pistils.

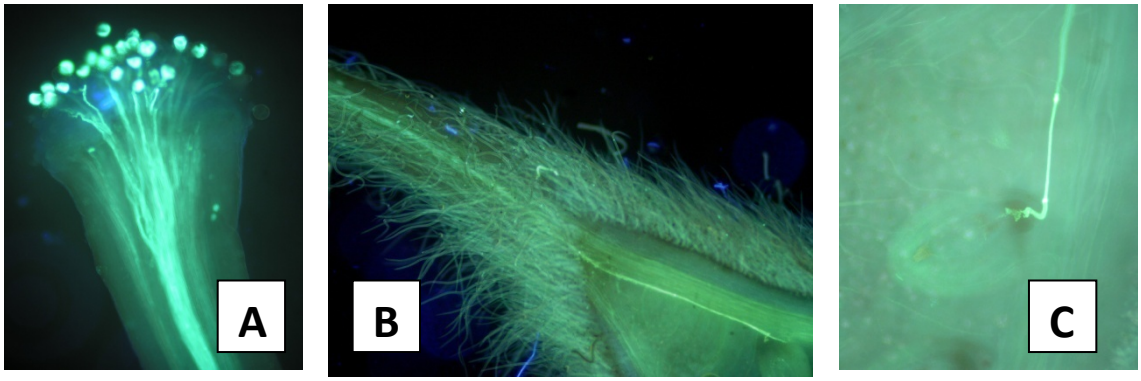

Figure 43. Fluorescence microscopic pictures of pollen tube growth in *Humboldtia bourdillonii*. Fig. 43 A. Pollen germination. Fig. 43 B. Penetration of pollen tube into the style and ovary. Fig. 43 *ca.* Penetration of pollen tube into the ovule.

Mating system in H. bourdillonii

Natural fruit set: Natural fruit set was found to be low; only 18/109 (17%) open pollinated flowers progressed to bear fruits (Table 3). Apomixis does not contribute to embryo development as bagged emasculated flowers (Fig. 44) did not set fruits. The absence of fruit set indicates the absence of apomictic fruit development in this species.

Fruit setting in self-pollinated flowers: Fruit setting did not materialize in controlled self pollinated flowers: (a) 32 flowers subjected to autogamy (pollinated by pollen of the same flower), and (b) 73 flowers set for geitonogamy (pollinated by pollen from the same tree) (Table 3). In both these cases, ovary dried off in the post-pollination phase. In order to overcome the limitations of self incompatibility, delayed self pollinations and bud pollinations were also tried but no fruit set could be obtained.

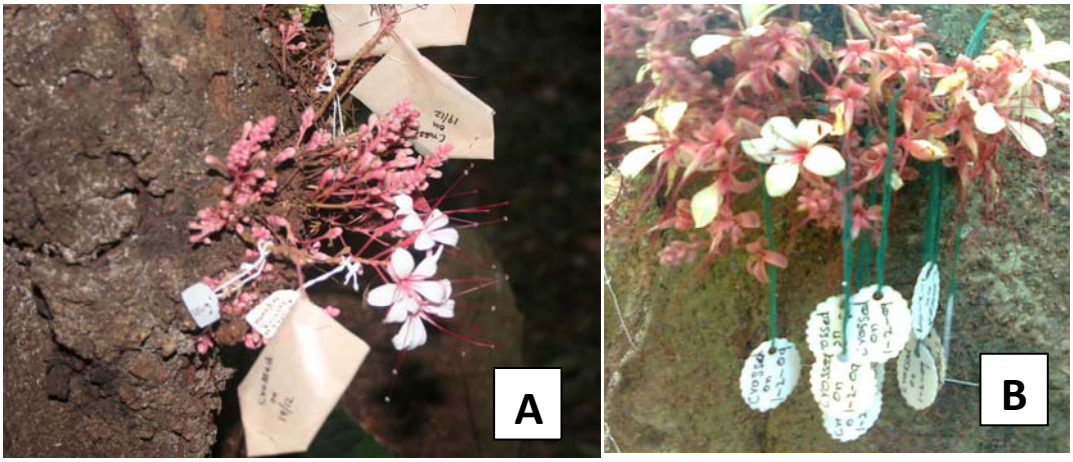

Figure 44. Controlled pollinations in *Humboldtia bourdillonii*.

Fruit setting in cross-pollinated flowers: Contrary to the self pollinated flowers, fruit setting occurred in cross pollinated flowers. Nearly 40% of the 91 cross pollinated flowers progressed to fruit setting (Table 19). The above experiments show that the species is self- incompatible or self-sterile. Pollen tube growth in self pollinated flowers was normal and they penetrated the ovules through the micropyle as in cross pollinated flowers. However, beyond the entry of pollen tubes into the ovules self incompatibility operated and is of the type 'post micropyle entry'.

Table 19. Fruit set in different modes of pollinations in experimental studies.

No.	*Mode of pollination*	*Flowers used*	*Fruits set*	*Percentage of fruit set*
1.	Natural pollination	109	18	16.5
2.	Artificial self pollination	105	-	-
3.	Artificial cross pollination	91	36	39.6
4.	Test-apomixis flowers	9	-	-

The related species, *Humboldtia brunonis,* is self-incompatible as pollen movement across short distances from related trees apparently reduces fruit and seed set (Charpentier *et al.,* 2000; Charpentier, 2002; Reisch *et al.,* 2007). In *H. bourdillonii* also, self incompatibility is one of the reasons for low fruit set and seed output. Though pollen viability and stigma receptivity are sufficiently long (over 24 hrs), wind pollination seems to be inefficient in transferring required quantities of pollen across flowers and trees and hence the low (23%) seed set.

Factors affecting fruit/ seed populations in H. bourdillonii

Fruit development takes two to three months for complete maturation. The mature pod is 9-13.5 x 3-4 cm, flat and tomentose. Young pods are crimson red, pubescent and velvety with thick sutures which turn brown and rigid on drying (Fig. 20 & 21). Seeds per pod varies from 4-6, 2.26 cm x 1.85 cm x 0.60 cm, brown, biconvex and exalbuminous.

Premature fruit-fall: Premature fall of young fruits were also observed in some trees (Table 20). Parts of the fruit wall were found stripped off in some fruits apparently by giant squirrels (Fig. 45 B).

Table 20. Premature fruit fall in *H. bourdillonii*.

Tree No.	Fruits fallen		
	Total	Damaged (%)	Undamaged (%)
1.	23	82.6	17.4
2.	21	71.4	28.6
Total:	**44**	**77.3**	**22.7**

Insect infestation of seeds: In the reproductive cycle of *H. bourdillonii*, insects interfere with reproduction in two stages, one in the inflorescence-stage and in the young-fruit stage and the other at fruit maturing stage. Jumping thrips colonize and multiply inside the ramifying space of the branched inflorescence and they suck the sap of the young fruits. A sizable portion of the fruits is lost this way (Fig. 45).

Insects that attack the seeds inside the developing young fruits are the weevils. The adults pierce the fruit wall and lay eggs in the cotyledons of each young embryo. The entire metamorphosis of the insect takes place inside the seed. The larvae devour the cotyledons and grow. As the seeds get dispersed the adults emerge out.

Weevil attack was prominent during April-May. During May-June, the peak period of seed damage, 13% (40/300) was found damaged by weevils. Out of the remaining 260 apparently healthy seeds, only 27.3% germinated. In germination trial 43% germination was obtained (Fig. 46). The poor germination percentage may be due to the high recalcitrance of the seeds (moisture content 62%) and high percentage of infestation by insect larvae (Fig. 45 A).

Figs. 45. *Humboldtia bourdillonii*. Fig.45 A. Seeds attacked by the weevil. Fig. 45 B. Young fruits spoiled by Malabar giant squirrel.

Fig. 46. Fruits, seed dispersal and young seedlings of *Humboldtia bourdillonii.*

Ishtiyak and Puni (2008) reported another instance of heavy infestation on the seeds of *H. vahliana* by the beetle *Cryptorhynchus indicus* Motsch. (Coleoptera: Curculionidae) causing 50-60% seed loss. The beetles lay eggs on the surface of the seeds and the larvae hatching out bore the young seeds and feed on the endosperm.

Seed dispersal in H. bourdillonii

Dry fruits explode to split open into two halves, separating along both sutures (Fig. 46). Dry dehiscent pods blast open dispersing the seeds just below the mother tree as the explosive mechanism is weak. *H. brunonis* also has the same ballistic dispersal mechanism scattering the seeds around the mother tree and indicates short distance gene flow (Shenoy and Borges, 2008). All the seeds produced from January onwards remain beneath the mother tree and get washed away by the flooding monsoon. Only heavy and continuous rains can produce sufficient flooding of water to carry the seeds. Dev *et al.* (2010) report that seeds of *H. brunonis* also get dispersed this way through seasonal monsoon- ravines.

Germination in H. bourdillonii

The mode of germination is epigeal. Early germination of the seeds in the pre-monsoon rains helps the seeds to anchor to the ground before intense rains and attach to the ground and escape from washing off.

In a germination trial conducted the percentage of germination obtained was 37.5%. Nearly 28% of the seeds germinated in May-June, which is the peak period of seed damage. In experimental germination trials germination spans between 44-185 days. Seeds show a maximum dormancy period of 3 months before germination.

Reproductive constraints in H. bourdillonii

The annual seed output in *H. bourdillonii* is low (17% of the total flower production) due to self incompatibility and unavailability of compatible pollen grains leading to reduced rates of cross pollination and fruit set. Decay of flowers and flower initials during irregular rains reduce seed set. Though flowering and fruiting periodicity is regular each year, a good amount of fruits is spoiled by insects or other agents in the immature stage itself. After dispersal, seeds are also subject to weevil attack causing reduction in viability.

Propagation studies in *H. bourdillonii*

Seed propagation in H. bourdillonii

As viable seeds are available for the species seed propagation was found feasible. The planting stock used for species recovery through augment planting employed the seedlings derived from seeds.

Rooting of stem cuttings of H. bourdillonii

Branch cuttings collected from Periyar Tiger Reserve in June, 2007 and March, 2008 were treated with varying concentrations of IBA for 10 minutes, bagged in soil/sand mix (1:1 ratio) and placed in the mist. Sprouts started appearing on the large cuttings 3-4 weeks after bagging. The rooting response of the cuttings is given in Table 21 (Fig. 47 A).

Treatment with 400-800 ppm of IBA solutions provided rooting; high concentrations like 1000-8000 ppm were not successful in initiating rooting. Ramets derived were transferred to polybags, hardened in the nursery for three weeks and later used for transplantation.

Table 21. Response of the branch cuttings of *H. bourdillonii* to varying concentrations of IBA.

Dates	*Treatment*	*Result (%)*				
		Total	*Callus**	*Rooting**	*Dead*	*Survival*
June 2007	400 ppm	10	60	40	10	80
June 2007	600 ppm	10	70	60	10	80
March 2008	600 ppm	10	60	11	10	-
March 2008	800 ppm	10	70	5	20	40
March 2008	1000 ppm	10	70	-	20	-
June 2007	6000 ppm	10	-	-	100	-
June 2007	8000 ppm	10	-	-	100	-

* Percentage survival of rooted cuttings.

Micropropagation of nodal explants in H. bourdillonii

Nodal explants from trees and sprouts derived from branch cuttings when cultured on Murashige and Skoog's and Woody Plant medium with various hormone combinations, sprouting response was extremely poor (< 5 %). The hormones used were (Benzyl Adenine I.5 ppm, 3 ppm and 6 ppm, Kinetin 3 ppm, and Benzyl Adenine/Kinetin 3 ppm/3 ppm). However, further development of the shoots was not obtained. Endogenous contamination was also a serious hurdle to carrying out experiments (Table 22).

Explants from cotyledonary nodes of seedlings when cultured on media containing BAP (1- 5 mg/l) resulted in sprouting of axillary bud (up to two shoots) but no multiplication of the shoots was observed (Fig. 47 B). Cotyledonary explants inoculated on a wide range of auxin concentrations, sparse callus development was seen. The brown callus that developed on mature cotyledons however did not show any signs of morphogenesis when subcultured to fresh media.

Table 22. Response of nodal explants of H. bourdillonii in culture to various sterilization treatments

No.	*Treatments*	*Contamination (%)**	
		Bacterial	*Fungal*
1.	Cefotaxime (100ppm) + Ampicillin (100 ppm) + 0.1 % $HgCl_2$	47	98
2.	Cefotaxime (200 ppm) + Ampicillin (200 ppm) + 0.1 % $HgCl_2$	23	94
3.	Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 0.1 % $HgCl_2$	12	83
4.	70% alcohol (10 seconds); Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1% Bavistin (60 mins) 0.1 % $HgCl_2$ (10 mins)	27	95
5.	70% alcohol (20 seconds); Cefotaxime (200 ppm) + Tetracycline (200 ppm) + 1% Bavistin + 0.1 % $HgCl_2$ (10 mins)	23	97

* Percentage of explants affected.

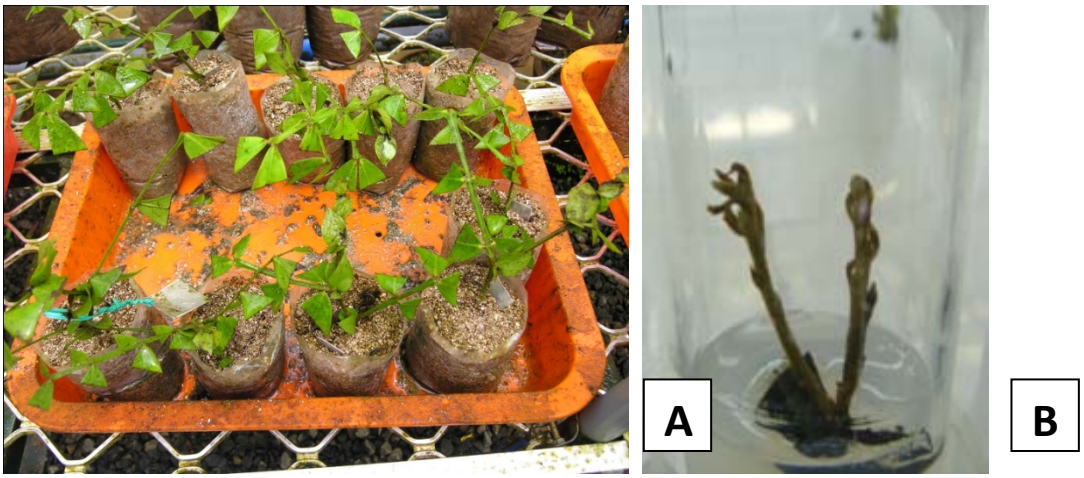

Fig 47. *Humboldtia bourdillonii.* Fig. 47 A. Rooting of branch cuttings. Fig. 47 B. Sprouting of axillary buds in culture medium.

In vitro *regeneration from immature embryos in* H. bourdillonii

Embryos from different stages of seed development when cultured in culture media containing hormones (NAA (1 ppm, 3 ppm, 5 ppm), 2,4-D (1 ppm, 3 ppm, 5 ppm), IBA (3ppm), Picloram (3 ppm), 2,4-D (3ppm) + BAP (0.5 ppm) callusing was scant. Although early stages of zygotic embryos showed some signs of differentiation further development was not obtained in any of the media tested. None of the treatments induced calli that could be maintained by subculture. Morphogenesis was also not observed in any of the treatments.

Species recovery through augment planting of *H. bourdillonii*

Seedlings were augment planted in three *in-situ* sites and three *ex-situ* sites. The *in-situ* sites were, Kulamavu MPCA (Site 3), Azhutha kadavu (Site 5), Ponnampara (Site 6), and the *in-situ* sites, KFRI-Arboretum (Site 9), KFRI-Subcenter (Site 10), and Field Research Center-KFRI (Site 11). The method of planting was the same as described for *D. bourdillonii.*

Transplantation experiments in Humboldtia bourdillonii

The seedlings obtained from propagation trials have been potted in poly bags, maintained in the nursery (Fig. 48), and used for testing transplantation feasibility. Experimental transplantation of seedlings of *H. bourdillonii* underneath the moist deciduous forest patch at KFRI has shown positive results. All the seedlings planted have survived confirming the amenability for augment planting for species recovery.

Survival of seedlings of H. bourdillonii in in-situ sites

Kulamavu-MPCA: The survival rate was 41%, mean height of seedlings 37 cm, and maximum height of 110cm (Fig. 49). A few seedlings were found decayed due to flooding in the planted site.

Figure 48. Transplanted seedling of *H. bourdillonii* in the nursery.

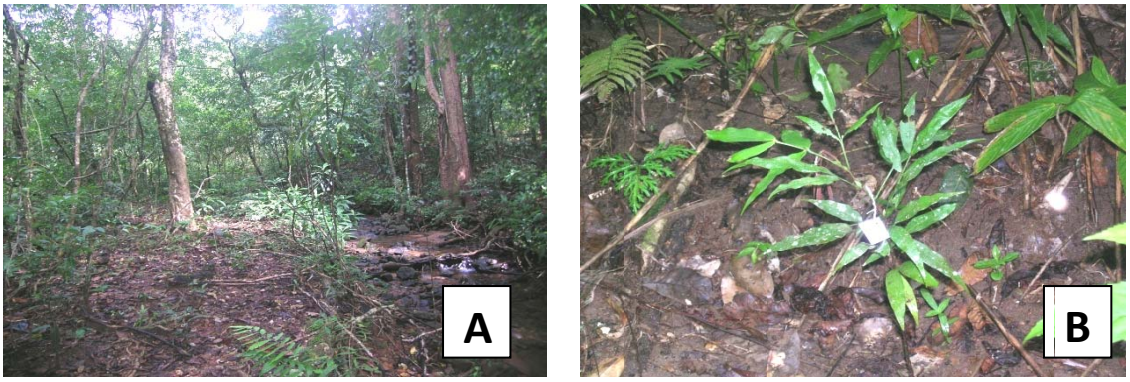

Fig.49. Restoration of *Humboldtia bourdillonii* at Kulamavu MPCA. Fig. 49 A. Planting site. Fig. 49 B. Planted seedling after a period.

Azhuthakadavu (PTR-W): The seedlings recorded a low success, 20%. The average height of seedlings 2-yr after planting was 39 cm and the maximum height 79 cm (Fig. 50). Browsing by domestic cattle, seedling damage/uprooting by wild pig, overflow of river water during peak monsoon, *etc.,*. were found to be major threats.

Fig. 50. Restoration of *Humboldtia bourdillonii* at Azhuthakadavu. Fig. 50 A & B. Planting sites. Fig. 50 *ca.* Planting work in progress. Fig. 50 D & E. Planted seedlings after a period.

Ponnampara: The seedlings showed a moderate survival rate, 50%. The average seedling height was 34 cm and maximum height 75 cm (Fig. 51). In this site, the weed primarily of *Strobilanthes* sp.,

overgrowing the planted seedlings to a height of 5-7' was a problem. Damage to seedlings from wild elephants and gaur was also noted.

Figure 51. Restoration of *Humboldtia bourdillonii* at Ponnampara. Fig. 51 A. Planting site. Fig. 51 B. Planting and monitoring seedling growth.

Survival of seedlings of H. bourdillonii in ex-situ sites

KFRI-Arboretum, Peechi: The seedlings exhibited 100% survival. The average seedling height was 75cm and maximum height 120 cm (Fig. 52).

Figure 52. Restoration of *Humboldtia bourdillonii* at the KFRI-Arboretum, Peechi. Fig. 52 A. Planting site. Figs. 52 B & *ca.* Planted seedlings after a time.

KFRI Sub centre, Nilambur: Here also, high survival rate was obtained, 98%. The mean seedling height was 55 cm and the maximum 140 cm (Fig. 53). Wild pig was found a threat in the site. Canopy shade was also found limiting the growth of seedlings.

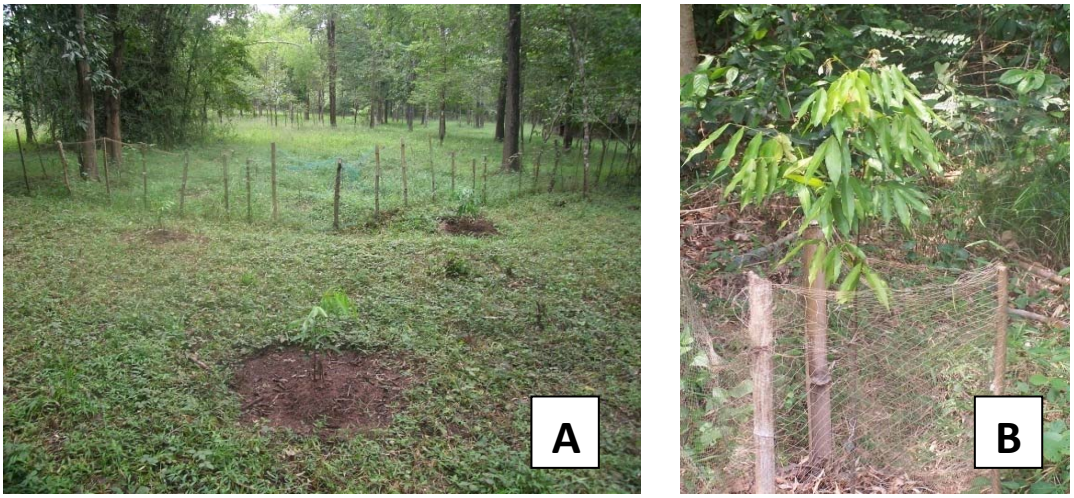

Figure 53. Restoration of *Humboldtia bourdillonii* at KFRI-Subcenter, Nilambur. Fig. 53 A. Planting site. Fig. 53 B. Planted seedling. Fig. 53 *ca.* A grown up seedling. Fig. 53 D. Seedling protected with fishnet fence.

Field Research Centre-KFRI, Velupadam: The seedling survival was 64%. The average seedling height was 72 cm and maximum 214 cm (Fig. 54). Browsing by spotted deer was found major threat to the seedlings.

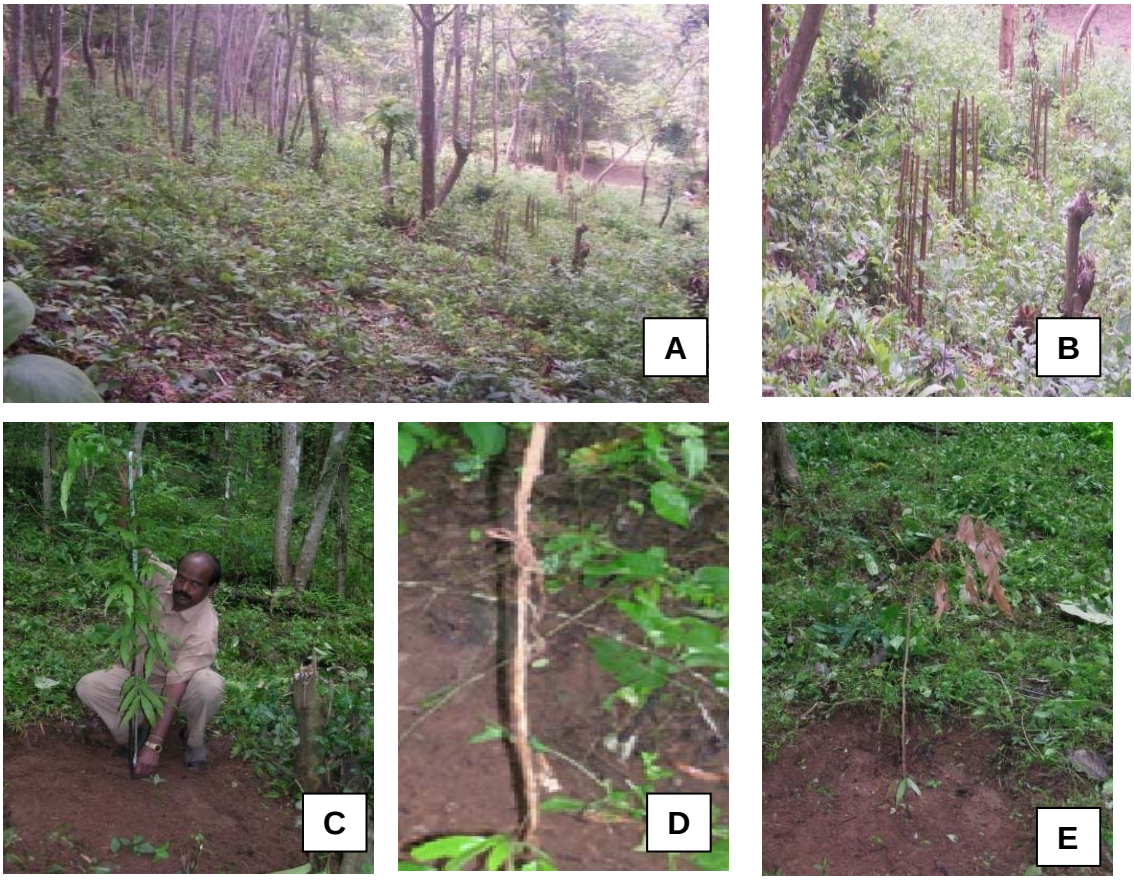

Figure 54. Restoration of *Humboldtia bourdillonii* at KFRI-Field Research Centre, Velupadam. Fig. 54 A & B. Views of planting site. Fig. 54 *ca.* Monitoring the sapling growth. Fig. 54 D. Seedling protection through wood

pegging. Fig. 54 E. Impact of deer browsing - Debarked wilted sapling.

Survival, mortality and growth of augment planted stocks

The details of seedlings of H. bourdillonii augment planted in *in-situ* and *ex-situ* sites are given in Table 23.

As with *Dipterocarpus bourdillonii*, the observed survival rates of planted seedlings of *H. bourdillonii* is also higher in *ex-situ* sites, in comparison to *in-situ* sites; 64-100% survival was obtained in the protective environment of the ex-situ sites. The low rates of survival in *in-situ* sites (20-50%) were due to many disturbances, and absence of protection.

Table 23. Survival, growth and factors affecting mortality of the restoration plantings of *H. bourdillonii.*

Site no.	*Planting sites*	*Seedlings planted*	*Survival after 2 yrs (%)**	*Mean height (cm)*	*Max. height (cm)*	*Factors affecting mortality*
	In-Situ sites					
3.	Kulamavu MPCA	200	41	37	110	Flooding
5.	Azhutha kadavu	50	20	39	79	Grazing, Wild pig, Flooding
6.	Ponnampara	50	50	34	75	Weeds, Elephants, Gaur
	Ex-Situ sites					
9.	KFRI-Arboretum	50	100	75	120	
10.	KFRI-Subcenter	50	98	55	140	Wild pig
11.	KFRI-FRC	50	64	72	214	Spotted deer

* Rounded off to whole digits.

6. General discussions and conclusions

Rare, endangered and threatened (RET) species constitute the weaker sections of the biota. They become rare either due to reproductive abnormalities, pest infestation, or due to antropogenic factors such as over-exploitation, land use change, changing management practices, *etc.,*. Several species do not produce viable seeds; *Cyanometra travancorica* and *Dipterocarpus bourdillonii* are typical examples. Species such as *Vateria macrocarpa* are restricted to very narrow geographic areas. There are instances where the species is known by 10-15 individuals (*Syzygium palghatense)*. In many instances, pollination is hindered due to absence of enough pollinators.

It is not unusual that some of the reproductive barriers which the RET species present are actually forced on the species. Reproductive biological studies of *Dipterocarpus bourdillonii* clearly show deficient pollinator populations. Degenerating embryos in the over 90% fruits/seeds and the non-germinating apomictic embryos of *D. bourdillonii* speak of abnormalities in the breeding system. There is every reason to believe that this situation has been brought about by inbreeding.

D. bourdillonii characteristically inhabits low level evergreen forests. The species being ripicole is characteristically confined to river courses where in the past it existed in continuum, roughly coinciding with the course of the river and its branching pattern. In the past, the low level evergreen forest existed much more extensively. However, much of these forests in the midlands of Kerala have been lost forever due to expansion of settlements and agriculture during the last three centuries. Along with the loss of the low level evergreen forest patches in the midlands, populations of *D. bourdillonii* contained in these forests would have been wiped out. As a consequence, the forests clothing the narrow tributaries higher up lost their connectivity resulting in fragmentation. Apparently the isolated populations of *D. bourdillonii* there perhaps have such a history. Unlike in the lower midlands, where the river is wide, the tributaries higher up are narrow and contained but limited individuals of *D. bourdillonii*.

As mature trees of *D. bourdillonii* are large having well over 1m diameter and with a clear bole of 25-30m, the trees were target to logging. This impoverished the populations further leaving 1-5 or often 1-2 trees in a population. Apparently this forced the species to inbreeding resulting in reproductive incompatibilities and embryonic mortality. Thus, prima facie, though it would look as if the loss of fecundity in *D. bourdillonii* stems from reproductive barrier, it is not difficult to see that this was forced on the species by human intervention.

In the case of *Humboldtia bourdillonii* again the total viable seeds derived from open-pollinated flowers are only 17%. Low pollen transfer through wind is quite inefficient and does not entertain profuse cross pollinations. Self incompatibility contributes to low fecundity and this is further aggravated by heavy forage of the seeds by weevils. *H. bourdillonii* inhabits pristine evergreen forests highly inaccessible due to the steep slopes and where logging or other human interventions have not disturbed the forest earlier. Thus, there is no reason to attribute the causes of low fecundity of the species to human intervention.

Transplantation experiments on *D. bourdillonii* and *H. bourdillonii* have shown that they are amenable for species recovery through assisted, augment planting. Though ecology and biology of the species are important in species recovery programmes (Bramwell, 1991; Vallee *et al.*, 2004), protection of the augment planted sites is more important, particularly where anthropogenic factors interfere with the survival of the planted stocks. An analysis of the survival rates of the augment planted stocks of *D. bourdillonii* and *H. bourdillonii* across *in-situ* and *ex-situ* sites shows that lack of protection is the main constraint in species recovery programmes.

* * * * *

A sizable entrenchment of global and regional fiscal resources for biodiversity conservation will be understandable only in terms of returns to mankind. Apart from the projected use value of the species, they also govern the functioning of ecosystems and the living environment. This tells us why biodiversity conservation should become so important a matter. Biodiversity is a future resource pool, the full potential of which has not been unfolded. Apart from the genetic materials, the real value of biodiversity is as a non-stop production system that continues to produce biomolecules unendingly.

Biodiversity conservation perhaps emerged first *ca.* 4000-5000 years ago in homesteads which later gave birth to agriculture and seed banking. With man's activities getting more and more anchored on land, game sanctuaries ensured availability of the game and thereby promoted conservation, though unwittingly. It took several decades to evolve the concept of 'reserved forests' with the shortfall of timber for ship building. The reserved forests became the precursors of the modern protected areas, *viz*, sanctuaries, national parks and biosphere reserves.

Today, there is a fairly large network of protected areas (Groombridge, 1992) which however does not encompass the entire plant diversity; at best these protected areas contain 50-60% of the total plant diversity of the respective regions. Limited by small areas, botanic gardens,

arboreta, medicinal plant gardens and germplasm conservatories, their content is often more skewed towards economically important species. 'Complete' conservation embracing all species of a given area seldom exists in *in-situ* or *ex-situ* conservatories. However, there is no dispute in that at least 'near-complete' conservatories are to be targeted.

* * * * *

Western Ghats (W. Ghats) are one of the two mega biodiversity centers in India, holding about 5,000 species of flowering plants. More than 70% of the plants (Mani, 1974; Sasidharan, 2004) known from the whole of W. Ghats are recorded from southern W. Ghats itself. Kerala State comprehending a major length of southern W. Ghats deserves special consideration for establishment of one or more larger 'near-complete' plant conservatories, where the state's native biodiversity can be conserved. These conservatories should also be able to conserve the 1,637 plants endemic to the region, which are distributed far and wide in inaccessible parts of the forests. Such establishments could also be utilized for long term researches, environmental education, staff trainings, and for imparting exposure to the public on our native biodiversity. Such centers will also enable to focus research on RET plants, their biology, conservation, and use values.

7. References

Aizen MA and Feinsinger P. 1995. Forest fragmentation, pollination, and plant reproduction in a chaco dry forest, Argentina. *Ecology* 75: 330-351.

Anonymous. 2000. *The 2000 IUCN Red list of threatened species*. IUCN, Gland.

Augustine J. 2002. Some threatened plants collected from Sabarimala and surrounding evergreen forests, Kerala. *Indian J. For.* 25 (3): 338-340.

Balasubramanyan K, Swarupanandan K and Sasidharan N. 1985. *Field key to the identification of indigenous arborescent species of Kerala Forests.* KFRI Research Report no. 33. KFRI, Peechi.

Bawa KS. 1990. Plant-Pollinator interaction in tropical rain forests. *Ann. Rev. Ecol. and Systemat.* 21: 399-422.

Bawa KS. 1998. Conservation of Genetic Resources in the Dipterocarpaceae. *In:* Appanah S and Turnbull JM. (Eds.). *A review of Dipterocarps: Taxonomy, Ecology and Silviculture.* Centre for International Forestry Research (CIFOR), Indonesia, pp. 45-55.

Bawa KS, Bullock SH, Perry DR, Coville RE and Grayum MH. 1985. Reproductive biology of tropical lowland rain forest trees: II. Pollination systems. *Am. J. Bot.* 72: 346-356.

Bell G. 1985. On the function of flowers. *Proc. Roy. Soc. London, Biol. Sci.* 224: 223–265.

Bramwell D. 1991. Botanic Gardens in conservation: Tree introduction into the wild. *In:* Heywood VH and Jackson PSW (eds.). *Tropical Botanic Gardens, their role in conservation and development.* Academic Press, London.

Chan HT. 1981. Reproductive biology of some Malaysian dipterocarps. III. Breeding systems. *Malaysian Forester* 44: 28-36.

Chan HT. 1977. *Reproductive biology of some Malaysian dipterocarps. Ph. D. Dissertation.* University of Aberdeen, Aberdeen, Scotland, UK.

Charpentier A. 2002. Consequences of clonal growth for plant mating. *Evolutionary Ecology 15: 521–530.*

Charpentier A, Grillas P and Thompson DJ. 2000. The effects of population size limitation on fecundity in mosaic populations of the clonal macrophyte *Scirpus maritimus* (Cyperaceae). *Am. J. Bot.* 87: 502–507.

Daljeet-Singh K. 1974. Seed pests of some dipterocarps. *Malaysian Forester* 37: 24−36.

Dayanandan S, Attygalla DNC, Abeygunasekera AWWL, Gunatilleke IAU N and Gunatilleke CVS. 1990. Phenology and floral morphology in relation to pollination of some Sri Lankan dipterocarps. *In:* Bawa K S, Hadley M. (Eds.). *Reproductive ecology of tropical forest plants.* UNESCO, Paris, pp.103–133.

Dev SA, Shenoy M and Borges RM. 2010. Genetic and clonal diversity of the endemic ant-plant *Humboldtia brunonis* (Fabaceae) in the Western Ghats of India. *J. Biosci.* 35(2): 267-279.

Gamble JS. 1915-1936. *Flora of the Presidency of Madras.* Adlard & Sons, London.

Ganeshaiah KN. 2005. Recovery of endangered and threatened species: developing a national priority list of plants and insects. *Current Science* 89(4): 599-600.

Ghazoul J. 1997. The pollination and breeding system of *Dipterocarpus obtusifolius* (Dipterocarpaceae) in dry deciduous forests of Thailand. *J. Nat. Hist.* 31: 901–916.

Ghazoul J, Liston KA and Boyle TJB. 1998. Disturbance-induced density-dependent seed set in *Shorea siamensis* (Dipterocarpaceae), a tropical forest tree. *J. Ecol.* 86: 462–473.

Ghazoul J and Satake A. 2009. Nonviable seed set enhances plant fitness: The sacrificial sibling hypothesis. *Ecology* 90: 369—377.

Groombridge B. (ed.). 1992. *Global biodiversity: status of the Earth's living resources.* Chapmann & Hall, London.

Ishtiyak PZ and Puni L. 2008. New record of seed pest, *Cryptorhynchus indicus* Motschulsky (Coleoptera: Curculionidae) in *Humboldtia vahliana* Wight. *Indian Forester* 134 (6): 849-854.

Jose PA, Pandurangan AG and Hussain A. 2011. *Ex-situ* conservation through macro propagation of *Humboldtia bourdillonii* Prain- A critically endangered tree of Southern Western Ghats, India. *Ann. For.*, 19(2) : 179-184.

Kaur A, Jong K, Sands VE and Soepadmo E. 1986. Cytoembryology of some Malaysian dipterocarps, with some evidence of apomixis. *Bot. J. Linn. Soc.* 92: 78-88.

Kenta T, Shimizu KK, Nakagawa M, Okada K, Hamid AA and Nakashizuka T. 2002. Multiple factors contribute to outcrossing in a tropical emergent *Dipterocarp tempehes*, including a new pollen-tube guidance mechanism for self-incompatibility. *Am. J. Bot.* 89(1): 60–66.

Krezdorn AH. 1986. *Citrus flowering, fruit set and development.* Citrus Research and Education Center, Florida, p 14.

Lee SL, Wickneswari R, Mahani MC and Zakri AH. 2000. Mating system parameters in a tropical tree species, *Shorea leprosula* Miq. (Dipterocarpaceae), from Malaysian lowland dipterocarp forest. *Biotropica* 32 (4): 693-702.

Lillesoe JPB. 1996. *Dipterocarps. A review of literature of relevance to tree seed source programmes with emphasis on the species in Malesia, ed. 2.* Ministry of Forestry, PT. Ardes Perdana, Indonesia.

Mani MS. (ed.). 1974. *Ecology and biogeography in India.* Dr. W. Junk Publishers, The Hague.

Momose K, Nagamitsu T and Inoue T. 1994. Reproductive ecology of an emergent tree, *Dryobalanops lanceolata*, Dipterocarpaceae, in a non-general flowering period in Sarawak. *In:* Inoue, T., Hamid, A. A. (Eds.). *Plant reproductive systems and animal seasonal dynamics: Long term study of dipterocarp forests in Sarawak.* Kyoto University, Japan, pp. 158-172.

Momose K, Yumoto T, Nagamitsu T, Kato M, Nagamasu H, Sakai S, Harrison RD, Itioka T, Hamid AA and Inoue T. 1998. Pollination

biology in a lowland dipterocarp forest in Sarawak, Malaysia I. Characteristics of the plant-pollinator community in a lowland dipterocarp forest. *Am. J. Bot.* 85: 1477–1501.

Muller-Dombois D. and Ellenberg H. 1974. *Aims and methods of vegetation ecology.* John Wiley & Sons, New York.

Murawski DA, Gunatilleke IAUN and Bawa KS. 1994. The effect of selective logging on inbreeding in *Shorea megistophylla* (Dipterocarpaceae) from Sri Lanka. *Conserv. Biol.* 8: 997–1002.

Nagamitsu T, Ichikawa S, Ozawa M, Shimamura R, Kachi N, Tsumura Y and Muhammad N. 2001. Microsatellite analysis of the breeding system and seed dispersal in *Shorea leprosula* (Dipterocarpaceae). *Internat. J. Plant Sci.* 162(1): 155–159.

Nair KSS. 1982. Seasonal incidence, host range and control of the teak sapling borer, *Sahyadrassus malabaricus* (Moore). Kerala Forest Research Institute, Peechi, Thrissur. pp36.

Nakagawa M, Takeuchi Y, Kenta T and Nakashizuka T. 2005. Predispersal seed predation by insects vs. vertebrates in six dipterocarp species in Sarawak, Malaysia. *Biotropica* 37(3): 389-396.

Narayanan IV and Swarupanandan K. 1996. Regeneration status of moist deciduous forests in South West India. *New Forests* 11: 185-205.

Nason JD, Aldrich R and Hamrick JL. 1997. Dispersal and the dynamics of genetic structure in fragmented tropical tree populations. *In:* Laurance WF and Berregaard RO Jr. (Eds.). *Tropical Forest Remnants: Ecology, Management and conservation of fragmented communities.* Univ. Chicago Press, Chicago, pp. 304-320.

Pascal JP. 1988. *Wet evergreen forests of the Western Ghats.* Institut Francais, Pondicherry.

Ramesh BR and Pascal JP. 1991. Distribution of the endemic arborescent evergreen species of Western Ghats. Pages: 20-29. *In:* Karunakaran CK (Ed.). *Proc. Symposium on Rare endangered and endemic plants of the Western Ghats.* Kerala Forest Department Wildlife Wing, Thiruvananthapuram.

Ramesh BR and Pascal JP. 1997. *Atlas of endemics of the Western Ghats (India): distribution of tree species in the evergreen and semi-evergreen forests.* Institut Francais de Pondicherry.

Reisch C, Schurm S and Poschlod P. 2007. Spatial genetic structure and clonal diversity in an alpine population of *Salix herbacea*. *Annals of Botany* 10: 1–5.

Sanford MT. 1992. *Pollination of citrus by honey bees.* Institute of Food and Agricultural Sciences, Univ. Florida.

Sanjappa M. 1986. A revision of the genus *Humboldtia* Vahl (Leguminosae–Caesalpinioideae). *Blumea* 31: 329-339.

Sanjappa M. 1991. Endemic legumes of Western Ghats. Pages: 30-43. *In:* Karunakaran CK (Ed.). *Proceedings of the symposium on rare endangered and endemic plants of the Western Ghats.* Kerala Forest Department (Wildlife Wing), Thiruvananthapuram.

Sasidharan N. 1998a. *Studies on the flora of Periyar Tiger Reserve.* Kerala Forest Research Institute, Peechi.

Sasidharan N. 1998b. Rediscovery of four threatened and possibly extinct endemic tree legumes from Kerala. *Higher plants of Indian Subcontinent* 8: 205-213.

Sasidharan N. 2003. Red listed threatened tree species in Kerala: a review. Pages: 1-12. *In:* Kallarackal J, Swarupanandan K and Sharma JK. (eds.). *Conservation and research needs of the rare, endangered and threatened (RET) tree species in Kerala part of the Western Ghats.* Kerala Forest Research Institute, Peechi.

Sasidharan N. 2004. *Biodiversity documentation for Kerala. Part 6: Flowering plants.* Kerala Forest Research Institute, Peechi.

Sathish BN, Kushalappa CG, Srinath K, Madhu KS and Nethravathi N. 2006. An insight in to the seed dispersal in *Dipterocarpus indicus* Bedd. A globally endangered and endemic tree species of Western Ghats. *My Forest* 42(4): 373 -378.

Schneider H. 1968. The anatomy of citrus. *In:* Reuther W, Batchelor LD, Webber HJ. (Eds.). *The citrus industry, vol. 2.* Univ. California Press, Berkeley. pp. 1–85.

Seavey SR and Bawa KS. 1986. Late-acting self-incompatibility in Angiosperms. *Botanical Review* 52: 195-219.

Shenoy M, Borges RM. 2008. A novel mutualism between an ant-plant and its resident pollinator. *Naturwissenschaften* 95: 61–65.

Shivanna KR and Rangaswamy NS. 1992. *Pollen biology- a laboratory manual.* Narosa Publishing House, New Delhi, 119p.

Tamari C, Jacalne DV. 1984 Fruit dispersal of Dipterocarps. *Bull. For. and For. Prod. Res. Inst., Ibaraki,* 325: 127–140.

Toy RJ and Toy SJ. 1992. Oviposition preferences and egg survival in *Nanophes shoreae* (Coleoptera, Apionidae), a weevil fruit predator in South-east Asian rain forest. *J. Trop. Ecol.* 8: 195-203.

Vallee L, Hogbin T, Monks L, Makinson B, Matthes M and Rosetto M. 2004. *Guidelines for the translocation of threatened plants in Australia, ed. 2.* Australian Network for Plant Conservation, Canberra.

Webber AC. 2000. Floral biology and pollination of some neotropical Annonaceae. Proceedings of the Workshop on Annonaceae, 31 Oct- 3 Nov 2000, Utrechet, The Netherlands, pp.18-21.