REPRODUCTIVE BIOLOGY OF *Bauhinia vahlii* Wight. & Arn.

Thesis

by

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FRONTIS PIECE

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CERTIFICATE-I

This is to certify that the thesis entitled, **"Reproductive Biology of *Bauhinia vahlii* Wight and Arn",** submitted in partial fulfillment of the requirement for the award of degree of **MASTER OF SCIENCE (FORESTRY) FOREST GENETIC RESOURCES** to Dr. Y.S. Parmar University of Horticulture and Forestry, Nauni, Solan (H.P.), is a record of bonafide research work carried out by **Ms. Bibi Nagaar (F-2015-23-M)** daughter of Mr. Qazi Abdul Qadoos under my guidance and supervision. No part of this thesis has been submitted for any other degree or diploma.

The assistance and help received during the course of investigations have been fully acknowledged.

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CERTIFICATE-II

This is to certify that the thesis titled, **"Reproductive biology of *Bauhinia vahlii* Wight and Arn"**, submitted by **Bibi Nagaar (F-15-23-M)** daughter of **Qazi Abdul Qadoos** to Dr. Yashwant Singh Parmar University of Horticulture & Forestry, (Nauni), Solan (H.P.)- 173 230 India in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE (FORESTRY)** in the discipline of **FOREST GENETIC RESOURCES** has been approved by the Student's Advisory Committee after an oral examination in collaboration with the External Examiner.

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Date
Place Nauni, Solan **(Bibi Nagaar)**

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LIST OF ABBREVIATION

%	:	Per cent
cm	:	Centimeter
CV	:	Coefficient of Variation
mm	:	Millimeter
m	:	Meter
No.	:	Number
P	:	Phenotypic
S	:	Site
σ	:	Standard Deviation
T	:	Tree
r	:	Replication
t	:	Treatment
g	:	Genotypic
p	:	Phenotypic
e	:	Environment
∑	:	Summation
e.g	:	for example
et al.	:	*et al* (Co- workers)
viz.	:	that is to say

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Chapter-1

INTRODUCTION

Bauhinia vahlii Wight. and Arn. is one of the giant woody climber belonging to family Caesalpiniaceae. The species is distributed in the sub-Himalayan region up to 1200 m above mean sea level and also in Assam, Central India, Bihar, Eastern and Western Ghats.

The species flowers in April – June and fruits ripen in the month of March-April next year. Flowers are white turning buff with age; hypanthium is 5-8 mm long, fertile stamen three along with two staminodes, petals five in number, ovary is densely tomentose, style hairy, flowers are white on long slender pedicels in terminal corymbose racemes. Pod dehiscence, seed dispersal and natural regeneration begins with the premonsoon rains. The seeds borne in a flat rusty coloured woody pod. It can grow on variety of soil, but it is important that soil should be well drained. Regeneration is poor in natural forest because, there is significant damage due to harvesting of leaves by the local people, for commercial purposes which not only damage the plants but also reduces its population. (Chauhan and Saklani 2013).

Reproductive biology is the study of flowers with its associated functions such as opening of flower buds (anthesis), anthers dehiscence, pollen viability, pollination pattern and stigma receptivity. Flowering phenology refers to the seasonal timing of flowering which has significance for both ecological and evolutionary reasons, (Kalloo, 1991). The reproductive pattern is one of the key factor leading to the abundance, distribution, and genetic diversity of organisms. In flowering plants, a highly diverse array of floral traits and reproductive systems have evolved, varying from obligated cross fertilization to obligated or promoted self-fertilization, with each strategy presenting selective advantages and disadvantages (Takebayashi & Morrell, 2001). Floral morphology form the basis of taxonomical phylogeny, pollination ecology and out crossing mechanism. Further the architecture of inflorescence, phenomenon of anthesis and floral orientation suffice the necessities of plant pollinator interface; especially in used pollinated species. Thus it is important in pollination studies to quantify a variety of morphological measurements of plant, inflorescences, or flowers (Fenster, 1991)

The female floral component i.e., gynoecium (stigma, style and ovary) is the most important part of pollination studies, mainly the viability of the stigma to support germination and subsequent growth. Also, pollen viability is one of the measures of male fertility (Kearns and Inouye, 1993), however the time and method of pollen collection and storage affect the viability (Stanley and Linskens 1974).

Studies on reproductive biology and breeding systems of trees are very important as the basic tool to carry out tree breeding programs. The breeding system through the mechanism of reproduction, the method of pollination and the degree of compatibility regulates the amount of recombination. The efficiency of reproductive system depends upon our understanding of factors affecting reproductive biology. To carry out any tree improvement program it is desirable to collect all the information regarding the variability which is existing in the species with respect to tree fruit, flower, seed and even chromosomal level.(Chauhan 2004)

Knowledge of the breeding system is also imperative for the conservation of threatened and endangered tree species. Study of the natural population structure, floral biology and the vector responsible for pollination and seed dispersal is needed in order to develop appropriate management strategies for both in-situ and ex-situ conservation (Sedgley and Griffen 1989).

Breeding systems include all aspects of sex expression in plants that affect the relative genetic contributions to the next generation of individuals within a species (Wyatt 1983). The study of breeding systems which leads towards the genetic structure of populations is also important to express the study of evolution and population genetics which is of considerable applied importance in agriculture (Richards 1997) as a tool to regulate and channalize the components of fecundity for selection purposes in cultivated plants (Frankel & Galun 1977) though in case of natural forests knowledge of breeding systems is an essential background for evaluating the factors responsible for the seed production and to understand the gene flow within and between populations. Different breeding systems have different implications for the rate of outcrossing as well as for pollination mechanisms and pollinator behavior (Dafni 1992).

Bauhinia vahlii is an indigenous, multipurpose species. In Kumaun Himalaya, it is most suitable for plantation programmes in mined, industrial waste areas and degraded forest

lands as it increases the soil fertility. The leaves provide an excellent source of fodder in the Central sub-Himalaya region and are also used as a material for making a variety of wrappers (Upreti and Dhar 1996). In the states of Madhya Pradesh, Orissa and Andhra Pradesh it can grow up to 10-30 m long. The woody stem can get as thick as 20 cm. The spreading stout branches are covered with rusty fine hair. Young branches, tendrils, petioles, underside of leaves especially along the nerves and inflorescences clothed with dense ferruginous tomentum. Leaves are variable in size, often up to 37cm in length as broad as long or broader, deeply cordate, 11-15 nerved, cleft through about 1/3 of the length, sub-coriaceous, dark green and glabrescent above. (Das *et.al.* 2013).

The plant leaves contain flavonoid, betulinic acid triterpene, campesterol, steroid catechin, gallic acid methyl ester, benzenoid, mopanol and 4-O-methyl ester. (Narayan *et.al* 2012). Various parts of this species have medicinal uses namely ; leaves as demulcent, edible seed as tonic and aphrodisiac, gum for medicines, bark yield tannin and leaves used as fodder, plate making (donas and pattal) and thatching (Agarwal 2003).

Leaf, petiole and stem of the species contain endophytic fungi which are mostly important in the biodiversity since they have an effect on structure and defence mechanism of plants and are also used to study the host-parasite relationship in natural ecosystem (Bagchi and Banerjee 2014).

Natural regeneration of a species is an ecological process of self organization , it is an open end process that expand future option for use of available forest land, to enrichment through planting other species of local importance to promote genetic and functional diversity. *Bauhinia vahlii* also perpetuates through natural regeneration, which is better on the forest sites having partial shade of the trees, but the regeneration is comparatively poor in drier areas. Naturally regenerating forests contribute more towards landscape heterogeneity, connectivity, biodiversity conservation and resilience (Chazdon 2014).

Therefore keeping in view the floral peculiarities and other multipurpose uses of the species, the present investigation have been framed with the following objective**s:**

1. To study morphometric characters of the species.
2. To study floral biology and breeding system.
3. To study seed characteristics and natural regeneration.

Chapter-2

REVIEW OF LITERATURE

Study of reproductive biology of *Bauhinia vahlii* is an important arena in the field of species improvement programme in general and *Bauhinias* breeding in particular. The species is open pollinated by insects hence the floral architect and the reproductive biology involved makes the studies more exhaustive. Therefore use relevant literature on *Bauhinia* spp. and other have been categorised as per the study outlines as below.

2.1 MORPHOMETRIC STUDIES

Khan D and Zaki M J (2015) studied leaf area, leaf architecture in *Bauhinia racemosa* Lamk. The graphically measured one-sided leaf area (LAM) of 50 individual leaves of *Bauhinia racemosa* Lamk. varied from 0.55 to 18.53cm^2. The Leaf length LL was determined as LL = LM + La + Lb where La was the apical leaf extension length and Lb the basal leaf extension length.

2.2 FLORAL BIOLOGY

Torres *et.al.* (2008) studied aspects of floral biology of buzz pollinated *Senna cana*. The flowers are perfect, without nectar, odorless and pollen is the main reward. Anthesis occurs around 7am. The stigma is receptive in the bud at pre-anthesis. They collected seven bee species visiting *Senna cana* flowers i.e. *Centris fuscata , Centris spp, Bombus brevivillus, Bombus morio ,Xylocopa cearensis, Xylocopa spp*. and *Euglossa spp* and another unidentified *hymenopterans* including ants and wasps. The morning (8AM to 12AM) was the day period with more visits (76.7%) and also with more flowers visited (86.8%).

Wani and Chauhan (2008) studied floral biology and stigma receptivity of *Bauhinia variegata* and reported that species started flowering during March –April the duration of flowering ranging from 26-49 days. Mean time taken to flower bud development was found to be between 20.88 hours to 24.92 days, maximum anthesis and anther dehiscence was observed between 7:30 to 8:30 am respectively.

Bergallo (1990) studied floral biology and breeding systems of *Bauhinia bongardii.* The floral biology showed features associated with chiropterophily (bat pollination). The

flowers were white and produce nectar, anthesis occurred during night between 16:00 and 18: 30h.

Lopes *et .al.* (1996) studied floral biology of *Swartzia pickelii* and its pollination by *Eulaema* spp. (Apidae-Euglossini). The species is mclittophilous and self-incompatible and its most important pollinators were three bee species of *Eulaema: E bomhiformis niveo/asciata, E. meriana flavescens* and *E. cingulata*.

2.3 BREEDING SYSTEM

Aguiara *et.al.* (2016) studied floral and reproductive biology of *Cenostigma macrophyllum.* The flowers are hermaphrodite, nectar produced and stored in hypanthium. The floral anthesis lasted two days, with fully distended petals at 6 AM, stigma become receptive at 7 AM and dehiscence-starting 9 AM, opening in groups throughout the day. The pollinators began visit around 6 AM until 5 PM flowers the first day of anthesis foraging in search of nectar. The effective pollinators were bees.

Smitha and Thondaiman (2016) studied reproductive biology and breeding system of *Saraca asoca* (Roxb.) the species bears fragrant flowers in paniculate corymbose inflorescence from December end to May, with peak flowering during February–March. The time of anthesis in this species is noticed in the early morning from 3.00 to 5.30 am, which coincided with anther dehiscence, stigma receptivity and insect activity. Pollen viability was maximum within 2 hour of anthesis, which decreased thereafter and no pollens were viable after 6 hours. The stigma was receptive at the time of anthesis and continued for 24 hour. The observations of the floral biology and breeding system indicated the cross pollination behavior

Costa *et.al.* (2014) Reproductive biology of *Vatairea macrocarpa* its flowering is biannual and occurs between the month of June to August. The flowers are andromonoecious anthesis is durinal and floral nectors are the main feature. *Centris* and *Xylocopa* are effective pollinators. The species is self-incompatible it produced no fruits experiments manual selfing and apomixes.

Neto (2013) studied floral biology and breeding system of *Bauhinia forficata* and reported that anthesis started at dusk release pollens and pollen production occurred at 8.00

and 10.00 pm respectively, flowers were protoandrous and stigma became receptive around 11.00pm .

Raju (2012) studied reproductive ecology of *Terminalia pallida* Brandis and found that the flowers are bisexual and obligately outcrossed and this is enforced by self-incompatibility The flowers offer both nectar and pollen for the foragers. The plant is entomophilous, and cross-pollination is effected mainly by large bees, wasps and butterflies. The natural fruit set was around 6% as against the 62% realized in manual xenogamous pollinations.

In a study on reproductive ecology of *Shorea roxburghii* G. Don (*Dipterocarpaceae),* an endangered semi-evergreen tree species of peninsular India, flowering observed was not annual, but when it does flower, in March, it shows massive blooming. Massive blooming, drooping inflorescence with pendulous flowers, ample pollen production, gradual pollen release as a function of anther appendage and aerodynamic pollen grains - all suggest anemophily. The characteristics of nectar secretion, hexose-rich sugars and amino acids in nectar are additional adaptations for entomophily. The plant is functionally self-incompatible, obligately outcrossing and ambophilous. The natural fruit set does not exceed 15% despite the plant being ambophilous. Raju *et.al.* (2011)

Almeida *et.al.* (2011) studied floral morphology, pollen transfer dynamics and breeding system of *Chamaecrista ramosa*. The polymorphism maximises cross-pollination and reduces self-pollination, Pollen deposition and capture occurred in specific sites of the floral visitor body, showing the functionality of enantiostyly. The floral architecture, associated with the floral visitor behaviour, resulted in indirect pollen deposition on the floral visitor body. the species is self-compatible, although -no fruit set was observed through spontaneous self-pollination.

Correia *et.al.* (2011) studied reproductive biology of four aggressive invasive Australian *Acacia* spp. in Portugal. *Acacia* spp. showed different investments in the production of reproductive units and in natural reproductive success, with *A. dealbata*, the most aggressive species, having the highest investment and reproductive success. Acacia *melanoxylon* showed a different reproductive strategy, andromonoecy, contrasting with the other hermaphroditic species. *Acacia* spp. were shown to be predominantly self-incompatible,

but a low level of spontaneous selfing enabled the production of viable offspring. *Acacia dealbata* and *A. longifolia* suffered pollen limitation.

Vikas and Tandon (2011) studied reproductive biology of *Azadirachta indica* (*Meliaceae*), a medicinal tree species from arid zones. Phenological observations established that trees within a population flowered synchronously, but populations varied in the time of onset of flowering; a few trees flowered twice a year.. The occurrence of natural pollination both by wind and insects indicates ambophily. Insect pollinators were predominantly represented by Hymenoptera and Lepidoptera; *Apis* spp. were the most effective pollinators.

Vasudeva and Sareen (2011) studied reproductive biology of *Dalbergia sissoo* Roxb the species flowers in the month of March to June and the peak flowering period was for a week. The anthesis, dehiscence of anthers, stigma receptivity and pollinator activity showed synchronised diurnal rhythm. Flowers were pollinated by honey bees, beetles, butterflies and thrips.

Lau and Saunders (2009) studied floral biology structure and breeding system of three climbing *Bauhinia* species namely *Bauhinia championii*, *Bauhinia corymbosa*, *Bauhinia glauca* in Hong Kong Southern China field observation revealed that all species are predominantly pollinated by bees (particularly *Apis mellifera*) and butterflies (*Graphium* a*nd Papilio* species). In controlled pollination fruit and seed set were generally highest followed by artificial outcrossing. *Bauhinia championii* was self compatible and *Bauhinia corymbosa* and *Bauhinia glauca* were self incompatibile. All species showed high level of heterozygosity.

Munin *et.al.* (2008) studied sphingophily and breeding system in *Bauhinia curvula* a shrub that bloom during six to seven month. The species was self incompatible and depends on pollinators and did not bear fruit after spontaneous self pollination. *Agrius cingulatus* was the only flower visitor.

Nautiyal *et.al.* (2009) *Aconitum balfourii* (Benth) was studied for reproductive biology. Protandry type of dichogamy was observed and is viewed as an anti-selfing mechanism. In general, higher pollen germination was achieved comparatively at low concentrations of GA_3, IAA and IB A (1 ppm). Tube elongation was maximum upto 65 pm in IAA 1 ppm and 63 pm in IAA (5 ppm) and sucrose 5%. Apomixis as well as autogamous self pollination was not observed in the species. However, fruit set differed significantly between

the hand-selfed and hand-crossed treatments. The abundance and efficiency of pollinators also affect mating patterns.

Ogilvie *et.al. (*2009) studied the pollination biology of the common shrub *Pultenaea villosa* Wild was examined in a subtropical dry sclerophyll forest in eastern Australia. The shrub's flowers are typical zygomorphic pea flowers with hidden floral rewards and reproductive structures. A range of insects visited the flowers, although bees are predicted to be the principle pollinators. Nectar and pollen are offered as rewards and were actively collected by bees. The breeding system experiment revealed that the species requires out crossing for high levels of seed set and that the overlap of stigma receptivity and pollen dehiscence within the flower suggests the potential for self-incompatibility.

Sunnichan *et.al.* (2009) studied reproductive biology of *Boswellia serrata*, The inflorescence is a terminal raceme and produces up to 90 bisexual, actinomorphic flowers. About 85% of the fresh pollen grains are viable. Flowers offer nectar and pollen as rewards to floral visitors. The giant Asian honey bee (*Apis dorsata*) and *A. cerana* var. *indica* (Indian honey bee) are the effective pollinators. The species is self-incompatible Cross-pollinated flowers allowed normal pollen germination and pollen tube growth, and resulted in fruit- and seed-set. Under open pollination fruit-set was only about 10%. Although manual cross-pollinations increased fruit set, it was only up to about 20%. Low fruit set appears to be the result of inadequate cross-pollination and other constraints, presumably limitation of available nutrients.

Wheeler *et.al.* (2009) studied reproductive biology of the endangered wildflower *Senna hebecarpa* pollination and fecundity. Study revealed that Wild Senna is not agamospermous; pollinator-excluded flowers produced no fruits . Flowering and fruiting was highly synchronous. Fruits set by 16 August and ripened through October. Self pollination produced a moderate reduction in fruit set.

Chauhan *et.al.* (2008) studied reproductive biology *Terminalia arjuna* It, flowers during April-July. The flowers are hermaphrodite, actinomorphic. Foragers include honeybees, butterflies, wasps, flies, ants and sunbirds. The fruiting behaviour indicates that this species shows facultative xenogamy, but mostly eliminates growing fruits from self-pollinated flowers. The facultative breeding system is considered to be adaptive for *T. arjuna*

for colonization as it facilitates fruit-set through self-pollination. Natural fruit-set is 48%. The winged and woody fruits are dispersed by wind and birds.

Diallo *et.al.* (2008) studied breeding system and pollination biology of the semi domesticated fruit tree, *Tamarindus indica.* Results revealed that *T. indica* is a species in which cross-pollination and self-pollination coexist together but the species is a cross-pollinated preferentially which is only partially self-incompatible.

Munin *et.al.* (2008) studied sphingophily and breeding system in *Bauhinia curvula* a shrub that bloom during six to seven month . The species was self incompatible and depends on pollinators and did not bear fruit after spontaneous self pollination. *Agrius cingulatus* was the only flower visitor.

Etcheverry and Aleman (2005) studied reproductive phenology, floral biology, degree of self-incompatibility, and floral visitors of *Erythrina falcata* were studied in an Argentinean population. The species fowers from August to October. Its phylogenetic position, floral morphology, and nectar characteristics suggest a hummingbird–passerine mixed pollination system. Bees were observed as occasional pollinators. Nectar production begins at anther dehiscence and coincides with maximum stigmatic receptivity. Controlled pollinations showed that this species is self-incompatible, although a few fruits develop from selfing. Only 1 percent of the flowers set seeds under natural conditions.

Nogueira and Arruda (2005) studied reproductive phenology, pollination and reproductive system of *Sophora tomentosa* L. on coastal sand dunes of Joaquina beach, Florianópolis, southern Brazil. The shrub flowers from the month of October to June. The anthesis takes place during the day, and does not have a defined schedule for the opening of the flower. Each inflorescence opens from 2 to 5 new flowers a day, lasting 4 or 5 days.The species presents a rate of open pollination of 78%, cross pollination of 70%, spontaneous self-pollination of 48%, and agamospermy of 18%. Pseudocentron sp. (*Megachilidae*) are an efficient pollinator.

Frawedozo (2004) studied reproductive phenology and dispersal patterns after natural regeneration in a limestone mining spoil banks of many species including *Bauhinia forficata* monthly observation of flowering and fruiting patterns of the community were done comparing the herbaceous shrub and woody species. The study showed that natural

introduction of the animals at the unreclaimed area increased the chance for long time of preserving the plant species since they are pollinated and dispersed their seeds.

Carvalho and Oliveira (2003) studied reproductive biology and pollination of *Senna sylvestris* (Vell) in the Panga Ecological Station, Uberlandia and in the Macaubas district of Patrocinio, Minas Gerais state, Central Brazil. The species is a shrub or small tree up to 5m which flowers from January to February. Flowers are hermaphrodite, zigomorphous,. The stigma is wet, non-papillous and crateriform, surrounded by hairs,self-sterile and non-apomictic. The main visitors and pollinators were large bees *Xylocopa brasilianorum*, *Oxaea flavescens*, *Bombus morio*, and species of *Centris*.

Liu and Koptur (2003) studied examined the breeding system and pollination of *Chamaecrista keyensis* Pennell. The species flowers during the peak flowering season (June to July). *Xylocopa micans* and *Melissodes* spp. Are effective pollinators the species is self-compatible ,61% of the self-pollinated flowers set fruit, no different from cross-pollinated flowers. Seed set of the control flowers was between that of selfed and crossed (higher than the former and lower than the latter), suggesting that flowers were pollinated by a mixture of self and outcross pollens under natural conditions. *Chamaecrista keyensis* thus has a mixed mating system.

Chavan *et.al.* (1999) studied on flowering phenology, floral biology and phenotypic variability for floral traits in *Tamarindus indica* L. Flowering was seen between last week of march to fortnight of June. Anther dehiscence was between 9-11am .Majority of trees were found to bloom one floral bud on every alternate day while 33 per cent trees has a regular blooming of at least one floral bud per day and only two trees were found to bloom more than one floral bud per day.

Rameriz *et.al.* (1984) studied floral biology and breeding system of *Bauhinia benthamiana* during two consecutive flowering cycles (Dec – Feb) anthesis occurred between 17:00 and 19:30 . Two phyllostomatid best species namely *Glossopgaga soricina* and *Phyllostimus discolor* visited flowers frequently. The species was self incompatible and functionally andromonoecious.

2.4 POLLINATION

Talwar S and Bhatnagar A K (2014) studied pollination biology of *Terminalia chebula* Retz. in Delhi and Western Ghats .The tree flowers from March to May in Dapoli

and from May to July .Flowers are bisexual, pale yellow, actinomorphic, epigynous and arranged in dense spikes. Floral nectaries are present at the base of style. Anthesis begins at night with the style emerging out before the stamens. A flower has two phases: (1) female phase when stigma is receptive; (2) bisexual phase,when stigma remains receptive and anthers mature and dehisce. Such a type of mechanism is known as incomplete protogyny. Anther dehiscence is asynchronous, facilitating cross pollination. The trees is entomophilous with a wide variety of insects visiting the flowers. The major pollinators of *Terminalia chebula* are *Apis dorsata, Apis cerana-indica, Polistes hebraeus, Vespa orientalis* and *Eristalinus* spp.

Borges (2009) studied phenology pollination, and breeding system *Caesalpinia echinata* flowering occurred mainly in the dry season and the peak of seed dispersal was at the beginning of the wet season. Anthesis is diurnal, lasting for day. The flowers are zygomorphic, yellow, sweet-scented, and the effective pollinators were mainly medium-sized to large bees of the genera *Centris* pollen viability was high Natural fruit set was low.

Tandon and Shivanna (2003) studied reproductive biology of *Butea monosperma*. flowers are typically papilionaceous; the stigma is wet papillate and the style is hollow. The flowers show characteristics of bird pollination being large. *B. monosperma* shows a weak form of self-incompatibility. Fruit set following manual self-pollination (5´25 %) was comparable with open-pollination (approx. 5 %) but was significantly lower than manual cross-pollination (22´51 %). This indicates that there is a high degree of geitonogamous pollination in this species.

Lewis and Gibbs (1999) studied the pollination biology, breeding system and fruiting success of *Caesalpinia calycina* and *C. pluviosa* var. *sanfranciscana* were studied in caatinga vegetation in Bahia. New Brazil. The principal pollinators for both species were carpenter bees. *Caesalpinia calycina* is andromonoecious but in C. *pluviosa* all flowers are hermaphrodite. In C. *calycina* all sclfcd flowers were abscised within 72 h despite rapid self-pollen tube growth to the ovary and ovule penetration. Prevention of selling therefore seems to be controlled by a post-zygotic mechanism. Both species had very low fruit-set and it is suggested that this is at least in pan due to geitonogamous pollinations with ovule penetration by self pollen tubes.

Hokche and Ramirez (1990) studied pollination ecology of seven *Bauhinia* species . *Bauhinia aculeate, Bauhinia moltinervia, Bauhinia pauletia. Bauhinia. ungulate. Bauhinia. glabra* . *Bauhinia* . *guionensis and Bauhinia rutilans* out of the above mentioned species first four were categorizes as pauletia which include trees. The species of *Tyloteacea* have comparatively small diurnal flowers and visited by a great variety of bees , wasps , butterflies and humming birds. The species of pauletia were mainly nocturnal.

2.5 SEED STUDIES AND NATURAL REGENERATION

Smitha and Das (2016) studied effect of seed moisture content, temperature and storage period on seed germination of *Saraca asoca* **.**In this study a correlation between seed moisture content and seed germination was established. It was observed that the rate of moisture loss in seeds is inversely proportional to the seed germination. Highest germination percentage (83.7%) was recorded at open nursery at 4 weeks after harvest followed by 25°C and 30°C at 3 weeks after harvest.

Dutta and Devi (2015) studied phenology, population structure and regeneration status of six tropical tree species namely *Bauhinia variegata, Carya arborea, Dillenia pentagyna, Sterculia colorata, Sterculia villosa* and *Terminalia bellerica*. Phenophases like leaf fall, leaf initation flowering and fruiting were recorded monthly for one year. Phenophases depend on environmental / meterological conditions of the study area and were species specific. *Bauhinia variegata and Terminalia bellerica* showed fruiting during cool and winter period. All six species indicated good regeneration or fair regeneration.

Lal *et.al.* (2015) studied regeneration status and floristic composition of natural and plantation forest ecosystems of barnawapara wildlife sanctuary, Chhattisgarh . A total of 33 species i.e *Bauhania racemosa , Bombax ceiba ,Cleistanthus collinus ,Careya arboriea ,Cassia fistula, Lagerstroemia parviflora* etc , and resulted that regeneration status in all the study sites were dissimilar. Closed natural forest (960 stems/ ha of seedlings and 220 stems/ha of saplings) displayed the better regeneration followed by open natural forest (350 stems/ ha of seedlings and 90 stems/ha of saplings) and teak plantations (340 stems/ ha of seedlings and 40 stems/ha of saplings) respectively. However under teak plantation *Lagerstroemia parvilora* has shown better regeneration.

Azad (2013) studied effects of variation in seed sources and pre-sowing treatments on seed germination of *Tamarindus indica*. Seeds were subjected to four pre-sowing treatments,

i.e., control, immersion in cold water (4°C for 24 h), immersion in hot water (80°C for 10 min) and scarification with sand paper. Seed germination was carried out in poly-bags with a mixture of topsoil and cow dung in the ratio of 3:1. The results revealed that the greatest success in germination (82.33%) was found in scarification with sand paper, followed with 81.67% in the cold water treatment.

Guevarra and Florece (2013) studied factors constraining the natural regeneration of alibangbang (*Bauhinia malabarica* Roxb.) in Carranglan Watershed, Nueva Ecija, Philippines field. The larva of Caryedon serratus (Olivier) infested the pods and seeds of *Bauhinia malabarica* with a mean infestation rate of 78.32 % and 73.23 %, respectively. On viability test, only 47.50 % germination was obtained in seeds soaked in tap water for 24 hours and seeds alternately soaked in tap water and hot water for 30 seconds had the highest germination energy (40.75 %). Therefore, serious infestation on *Bauhinia malabarica* seeds and low viability limits the regeneration of the species in the watershed.

Miranda *et.al.* (2013) studied germination of *Prosopis juliflora* (Sw.) D.C. seeds at different osmotic potentials and temperatures .To study the effects of polyethylene glycol (PEG) and NaCl stress and temperature on germination, two separate experiments were carried out at the Plant Ecophysiology Laboratory of the Federal University of Pernambuco in 2011. The overall germinability decreased significantly with increases in both PEG, however, the effects were more accentuated with PEG than NaCl. In contrast, NaCl-treated seeds usually lost their ability to germinate; this fact was possibly linked to the accumulation of Na^+ and Cl^- in the cells, which contributed to a loss of membrane function that led to the death of the embryos.

Azad *et.al.* (2012) studied seed germination of *Albizia procera* (Roxb.) Benth. in Bangladesh: a basis for seed source variation and pre-sowing treatment effect.Seeds were treated with four pre-sowing treatments, i.e., control, immersion in cold water (4°C for 24 h) and immersion in hot water (80°C for 10 min and 100°C for 1 min). The The results revealed that pre-sowing treatments affected the rate of germination of seeds, which significantly increased the germination percentages of seeds in hot water(82.075) treatments compared with those in control (60.60%) and the cold water treatment (4°C for 24 h, 63.53%).

Pawar *et.al.* (2012) studied regeneration status in relation to anthropogenic disturbance in tropical deciduous forest of Chhattisgarh. Regeneration status of forest was

determined based on population size of seedlings and saplings. A total of 27 species of 19 families were encountered. In the entire three sites, least disturbed site was good regenerating because of high density of seedlings and saplings in forest site, however moderately disturbed site show comparatively better regeneration and highly disturbed site did not show good regeneration.

Azad *et.al.* (2011) studied effect of different pre-sowing treatments on seed germination percentage and growth performance of *Acacia auriculiformis*. Seeds treated with five pre-sowing treatments (control, immersion in cold water, immersion in hot water, scarification with sand paper and immersion in concentrated H2SO4). The highest germination success rate was found 83% in hot water treatment followed by 78% in scarification with sand paper, and 75% with immersion in H2SO4 Hot water treatment was recommended on seed germination of the species.

Mwase and Mvula (2011) studied effect of seed size and pre-treatment methods of *Bauhinia thonningii* Schum. on germination and seedling growth However, the seeds are dormant and the tree species remain undomesticated. Seeds grouped into two categories: small (1 to 5 mm) and large (>5 to 10 mm) were subjected to five main pre-sowing seed treatment methods namely; soaking in cold water for 12 h, soaking in hot water for 10 min, nicking, soaking in potassium nitrate (0.2%) for 10 min and soaking in concentrated hydrochloric acid (0.3 M) for 5 min, and a control where seeds were sown without any treatment. The results showed that the combination of nicking and large seeds produced the highest (100%) germination and highest height and diameter growth.

Noumi *et.al.* (2010) studied *Acacia tortilis* in the North African arid zone: the obstacles to natural regeneration they estimated rate of infestation of seeds by *Bruchidius raddianae* and *B. aurivillii* and tested the autotoxicity effect of leaf extracts (different concentrations) of *A. tortilis* in the laboratory at 30 °C (optimum germination temperature). Results show that the inhibitory effect on germination increase with the increased leaf extract concentration. At 100% concentration, a significant reduction of germination rate was found compared to other concentrations. On the other hand, the study also revealed a high infestation rate of seeds by *B. raddianae* and *B. aurivillii* which reduces the possibility in inhibits natural regeneration in *A. tortilis*

Azad (2010) studied effect of pre-sowing treatments and seed germination in *Albizia richardiana* and *Lagerstroemia speciosa*. Results revealed that the germination rates of seeds in different pre-sowing treatments were significantly increased compared to those in cold-water treatment in both species. The highest germination rate was found to be 96% in hot-water treatment followed by 87%, 83% and 49% in treatments with scarification, H_2SO_4 and control in *A. richardiana*, respectively. However, the highest germination rate (79%) was found in H_2SO_4 treatment followed by 64%, 62% and 25% in treatments with hot water, scarification and control in *L. speciosa*, respectively.

Seedling growth strategies in *Bauhinia* Species comparing lianas and trees were collected by Cai *et.al.* (2007). Lianas are expected to differ from trees in their growth strategies. As a result these two groups of woody species will have different spatial distributions, lianas are more common in high light environments. This study determines the differences in growth patterns, biomass allocation and leaf traits in five closely related liana and tree species of the genus Bauhinia.

In a study conducted by Matin *et.al.* (2006) on seed germination, seedling growth and rooting of branch cutting of *Dalbergia sissoo* Roxb. The seeds with coats showed higher success (68%) but seed coat delayed germination by limiting water absorption in dormant seeds. Higher growth was found in seeds with coat both in terms of height and diameter of seedlings. In case of rooting of branch cuttings, use of 0.8% IBA showed the highest rooting success (90%) with more root number (30) than control where rooting success and root number were 60% and 15, respectively. Difference was not found among the average root length in control and treated cuttings.

Alamgir and Hossain (2005) studied effect of pre-sowing treatments on germination and initial seedling development of *Albizia saman* in the nursery. Seeds were treated with five pre-sowing treatments to study the effect of pre-sowing treatments on germination and initial seedling development in the nursery. Results revealed that Nail clipping in one side of the seed (at the distal end of the seed) (T4) provides the highest (50%) seed germination. The second highest germination (42%) was obtained for the seeds treated with immersion in cold water for 24 h . Germination was completely inhibited when the seeds immersed in boiled water for 30s followed in cold water soaking for 24 h.

Prasad and Nautiyal (2003) studied effect of orientation of seed placement in soil on seedling emergence in two *Bauhinia* species *Bauhinia vahlii* Wight and *Bauhinia racemosa* Lam . They recorded earliest and highest seedling emergence for both the species, emergence was poor in *Bauhinia vahlii* seeds when sown in flat position and lowest when seed was sown in horizontal position while in case of *Bauhinia racemosa* emergence was poorest for seeds when sown in an inverted and horizontal position and was lowest when seeds were sown in flat position . Seedling survival, mean germination time, root and shoot growth followed the same trend.

Raja *et.al.* (2004) studied effect of palm age on seed germination and seedling vigour in *Areca catechu* L. The germination test was conducted in sand medium with 50 seeds for each replication at 25 and 95 per cent relative humidity. At the end of 90 days, the number of normal seedlings were counted and the germination per cent was calculated. The seeds from 60 years old palm were also showed the significant differences, and was followed by the seeds from 45 years old palm.

Mathew and Shivakumar (2002) studied accelerated seed germination in *Bauhinia purpurea* by pre sowing growth regulator treatment and reported that when seeds were treated with gibberlic acid at 500ppm for 120 minutes the germination percentage increased from 54.7 to 96% at the same concentration for 180 minutes has reduced the time required for 50 percent seed germination from 13 – 3 days.

Chapter-3

MATERIALS AND METHODS

The present investigations entitled " Reproductive biology of *Bauhinia vahlii* Wight and Arn'' was carried out in the Department of Tree Improvement And Genetic Resources, Dr. Y. S Parmar University Of Horticulture and Forestry, Solan Himachal Pradesh during 2016-2017. The study has been conducted at two sites namely Jabli (Shimla- Chandigarh) national highway and Jorhji (Solan –Nahan) state highway. The area falls in mid hill zone of Himachal pradesh and has subtropical climate. The maximum temperature rises up to 40^0C in summer and minimum temperature as low as 0^0C during winter. The rainy season start at the end of June month up to July- August .The landscape lushes green and fresh. A survey was conducted to identify the superior trees in natural population of *Bauhinia vahlii* and five trees were selected and marked with paint in both sites. In order to study the reproductive and morphometric traits, fifteen samples of flowers and leaves were taken from each tree and studied under three replicates.

Since this species is locally used as fodder and also for making Donas, plates, pattal etc which affected the natural regeneration, to mitigate the poor regeneration study was taken up by marking five major plot of size 20×25m; in each plot five quadrates of size 2m×2m were laid down and data was recorded during May, 2017. The observations were taken on number of new recruits, unestablished and established regeneration. (Chacko 1965).

Table 1. Natural regeneration

Site	Altitude	Latitude	Longitude
Jabli (Solan) S_1	1483metres AMSL	$31^0 18' 22'' N$	$77^0 9' 8'' E$
Jorhji (Sirmour) S_2	1502 metres AMSL	$30^0 50' 4'' N$	$77^0 3' 28'' E$

The present investigations have been categorised under following technical headings

3.1 MORPHOMETRIC CHARACTERS

Morphometric refers to the measurement of form including shape, structure and other aspects of external appearance. In taxonomy and other field of natural science morphometric contribute to increase scientific regour in the description of important aspects of the phenotypic diamensions of diversity.

3.1.1 Leaf characteristics

3.1.1.1 Leaf length(cm) Leaf length was measured with scale from tip of the apex toward base.

3.1.1.2 Leaf width(cm) Leaf width was measured with scale at point where the length of central vein was half.

3.1.1.3 Leaf area(cm^2) Leaf area was measured with Leaf Area Meter (LICOR-model 3100A) and recorded data was deducted from constant value 199.9.

3.1.14 Petiole length(cm) it was taken with scale from the point where it was attached to the main branch up to the position of leaf lamina.

3.1.2 Floral characteristics.

All parts of flowers were measured with scale

3.1.3 Pod characteristics

3.1.3.1 Pod length (cm) Pod length was measured with scale from the point were it attached to the pedicel.

3.1.3.2 Pod width (cm) Pod width was measured with scale across the widest expansion

3.2 FLORAL BIOLOGY

Floral biology is important to understand about the flower architecture which in turn regulate the effective pollination mechanism. Different parts of the flower have been examined carefully and their size, colour and system of arrangement was studied. The observations were recorded on different whorls of the flower viz calyx, corolla, androecium and gynoecium.

3.2.1 Calyx and Corolla

Observations were taken by examining sepals and petals, sepal and petal were separated and measurement was taken with the help of scale. In this way fifteen flowers were examined individually to find the morphological variation. The comparison of calyx and corolla of all the samples was made in order to find the prominent floral description.

3.2.2 Androecium and gynoecium

Reproductive parts were examined to understand the potential breeding behavior of the species. Pistil and stamens parts were separated with sharp blade and measured with scale. Querries were also taken with respect to type, arrangement, position, attachment and whorls cycly .

3.2.3 Anthesis

In order to study the time of bud opening, five inflorescence per tree from all the trees covering both sites were tagged and number of buds opened in different hours (6am-3pm) at each site were counted. Flowers were tagged with thread on the inflorescence in order to avoid recounting. The mode and manner of flower opening was observed from the moment petals begin to open till the flower open. Early morning onwards number of fully opened flowers were noted at an hour interval. The bud stages were compared for their appearance, size texture and compactness in order to decide the stages.

3.2.4 Anther dehiscence

The mode of anther dehiscence was carefully observed and recorded for each flower tagged, in this way five inflorescence were observed individually to determine the time of anther dehiscence in each flower. Buds at all the identified stages were physically examined under dissection microscope for anther dehiscence and adherence of pollen to anther surface. The stage at which such adherence of pollen was observed taken as anther dehiscence stage.

3.3 BREEDING SYSTEM

In Tree Breeding to encompass study of breeding system is prerequisite principle to the trait specific genotype improvement of the forest tree.

3.3.1 Pollen collection

Direct pollen collection: Inflorescence for pollen dehiscence were collected at anthesis stage and placed on paper sheets kept under partial shade under the laboratory conditions. The anthers dehisced thereby liberating the pollens on the paper sheets which were kept under partial shade and then brushed using sterile camel’s hair brush. All the pollen samples were collected in isolation to prevent contamination from any other source. The pollens thus dehisced were stored in glass vial to check viability.

3.3.2 Pollen germination and pollen viability (%)

Pollen germination and viability was studied using in-vitro assay and staining method and expressed as viability percentage and germination percentage respectively

Pollen viability (%)

The pollen viability of the freshly collected pollens was studied in 2 percent acetocarmine solution. Pollen were dusted on a clean homocytometer with hair brush to which a drop of acetocarmine solution was added that was kept as such for 10-15 minutes to allow the pollen to absorb the stain. Several fields of the pollen mass were examined under microscope. Deeply stained normal looking pollen grain were counted as viable pollen, shriveled and weakly stained were recorded as non viable ones. The pollen viability was calculated as the percent measure of viable pollens to the total pollens observed.

Pollen germination (%)

In -vitro pollen germination of individual tree flowers was conducted using various sucrose concentration (5%,15% and 20%) to detect the optimum level of sucrose required for pollen germination. Pollen germination was determined by hanging drop method in which sucrose solutions were prepared by dissolving required weight in beaker to which 100ml distilled water was added. To accomplish the pollen grain germination a drop of sucrose solution was put in cavity slide and pollen grain were dusted and evenly mixed with sucrose by means of sterilized needle. The cover slip was waxed to check the evaporation of the solution. The slides thus prepared for various concentration were placed in petri-dishes containing moist filter paper and data on germination recorded after 24 hours.

3.3.4 Stigma receptivity (%)

Study of stigma receptivity is an important criterion to get successful pollinations. Stigma receptivity refers to the viability of the stigma to support germination and pollen tube growth of viable compatible pollen. Data of stigmatic surface was taken by the visual observation method in which change in the appearance of stigma was observed from 24 hour before opening of bud till it withered completely. The green shiny stigma was considered receptive while dull green stigma considered as non receptive.

3.3.5 Pollination and Pollen vectors

The activities of flower visitors was carefully observed during 2nd and 4th week of May in the flowering period, insects were collected in bottle and identified in consultation with the experts of entomology.

3.4 CONTROLLED POLLINATION

To examime the predominant breeding behaviour of the species different method of pollination was performed and data was recorded on fruit set.

3.4.1 Controlled Pollination

On all trees already opened flowers and immature buds were removed and only suitable buds were kept. Emasculation was done carefully in order to avoid damage to stigma. Freshly collected pollens from a desired pollen parent were dusted on receptive stigma using camel's hair brush. In this way fifteen flower on each tree in both the sites were pollinated and covered with pollination bags, data on fruit set was recorded after fifteen days.

3.4.2 Selfing

In this experiment ten inflorescence per tree on all the ten trees in both site were allowed to self pollinate by putting a pollination bag on initial stage of floral bud development. Fruit set in these inflorescences were recorded.

3.4.3 Controlled crossing between selected trees at both sites

For study of controlled pollination pollens were collected from selected trees in both sites and kept in glass vial. Pollens collected from $site_1$ (Jabli) were used as male in $site_2$ (Jorhji) and pollen collected from $site_2$ (Jorhji) were used as male in $site_1$ (Jabli). Pollination was done during morning hour 7am – 11am when maximum number of flowers started opening. Hand pollination was done by dusting freshly collected pollen grain by using camel hair brush. Data on fruit set was recorded after 15 days.

Fecundity was calculated from the mother trees by obtaining the ratio of seed set to fruit set. Fruit set is the percentage of flowers that produce fruit and seed set is the percentage of ovules in pollinated flowers that develop into seeds.

3.5 SEED CHARACTERISTICS, GERMINATION AND NATURAL REGENERATION

Seed characteristics were studied by collecting the naturally set pods from the selected trees of the sites during March. 2017.

3.5.1 Seed weight

Individual tree seeds were extracted, cleaned and kept in polythene bags with respective tags. Hundred seed weight was taken individually for each tree using weighing balance (ISTA 1966).

3.5.2 Seed germination

Seed germination study was carried out in polyhouse. Seeds from each tree were collected and sown in polythene bags using RBD design with three replicates. The germination percentage was calculated as given below**.**

$$\text{Seed germination percentage (\%)} = \frac{\text{Total number of seed germinated}}{\text{Total number of seed sown}} \times 100$$

3.5.3 Natural regeneration

To study natural regeneration survey was carried out in February and May 2017 on both sites. The sufficiency of regeneration was judged on the basis of number of established plants in a unit area. According to Chacko (1965), desired number of established plant is 2500 per hectare and the quadrate is considered fully stocked when it contained at least one established plant. In both sites five major sample plot of size 25×20m and within each major sample plot five quadrates of size 2m×2m were laid down. The survey was conducted on recruits, unestablished and established regeneration. The data thus collected was analyzed using the formula of Chacko (1965) as follows:

Recruits(ri)/ha $= 2500 \sum_{i=1}^{n} \text{ri} / \text{m}$

Unestablished regeneration(ui) /ha $= 2500 \sum_{i=1}^{n} \text{ui} / \text{m}$

Established regeneration (ei)/ha $= 2500 \sum_{i=1}^{n} \text{ei} / \text{m}$

Where

n = Number of sampling units

m= Total number of recording units in survey

r i = Total number of recruits in each sampling unit

ui = Total number of unestablished plants in each sampling unit.

ei = Total number of established plants in each sampling unit.

Data on morphological, breeding system and seed characteristics of *Bauhinia vahlii* were analysed using 'Randomised Block Design (RBD) .

Statistical analysis for each parameter were carried out on mean values and the Analysis of variance (ANOVA) was set up (Cochran and Cox , 1957) as depicted in the table.

ANOVA table for Randomised Block Design (RBD)

Source of variation	Degree of freedom	Mean Sum of Square	Variance ratio
Replication	(r-1)	MSR	MSS
Treatment	(t-1)	MST	MSE
Error	(r-1)(t-1)	MSE	

r = number of replications,

t = number of treatments

MSR = Mean sum of square due to replications

MST = Mean sum of square due to treatment

MSE = Mean sum of square due to error

Standard error of difference of mean of treatments were calculated as (SE_d)

$SE_d = \sqrt{2MSE/r}$

The critical difference (CD) was calculated as

$CD = SE_d \times t_{0.05}$

Where

$t_{0.05}$ = table value at error degree of freedom and 5% level of significance

Environmental variance ($\sigma^2 e$) = MS

Genotypic variance ($\sigma^2 g$) = $(\frac{MST-MSE}{r})$

Phenotypic variance ($\sigma^2 p$) = $\sigma^2 g + \sigma^2 e$

Coefficient of variability were worked out by formula suggested by Burton and De Vne (1953)

Phenotypic coefficient of variability (%) = $\frac{\sigma p}{\bar{x}} \times 100$

Genotypic coefficient of variability(%) = $\frac{\sigma g}{\bar{x}} \times 100$

Environmental coefficient of variability(%) = $\frac{\sigma e}{\bar{x}} \times 100$

Genetic advance = H×σp×K

Where H= $\frac{\sigma 2g}{\sigma 2p}$

The value of K= 2.06 (Allard ,1960)

Genetic gain (%) = $\frac{\text{genetic advance}}{\bar{x}} \times 100$

Chapter-4

RESULTS AND DISCUSSION

The present investigations entitled **"Reproductive biology of *Bauhinia vahlii* Wight. and Arn."** was carried out in the Department of Tree Improvement and Genetic Resources, College of Forestry, during 2016-2017, to study variation in morphometric traits, floral biology, breeding system, seed characteristics and natural regeneration of this species. The results obtained from the study have been presented under the following heads:

4.1 Morphometric traits
4.2 Floral biology
4.3 Breeding system
4.4 Controlled pollination
4.5 Seed studies and natural regeneration
4.6 Genetic estimates

4.1 MORPHOMETRIC STUDIES

The observations on morphological characters viz. leaf characteristics, floral biology and pod characteristics have been recorded for each site and presented as below.

4.1.1 Leaf morphmetric characters

These characters were studied to observe variation in leaf parameters viz. leaf length, leaf width, leaf area and petiole length. The results obtained were found significant with respect to leaf length, leaf width, leaf area and petiole length.

4.1.1.1 Leaf length (cm)

Individual tree leaf samples were measured for obtaining mean leaf length. Statistical analysis depicted that in S_1 (Jabli) the mean leaf length was 20.15 cm which was higher than S_2 (Jorhji) where it was 18.22 cm (Table.1). The variation in leaf length from individual trees of both sites was found to be significant within site$_1$ and the maximum length was registered by S_1T_1 (24.21 cm) which was at par with S_1T_4 (22.5cm), where as in S_2 (Jorhji) there was no significant variation between the trees , however the maximum leaf length was recorded in S_2T_4 (19.44 cm) and minimum in S_2T_3 (16.92 cm).

4.1.1.2 Leaf width (cm)

The mean leaf width recorded in S_1 (Jabli) was 21.69 cm and in S_2 (Jorjhji) it was 19.20 cm. The study site S_1 (Jabli) registered maximum mean leaf width and the variation was significant between the two sites. The leaf width of trees within S_1 (Jabli) was found maximum in S_1T_1 (25.80 cm) and minimum in S_1T_5 (17.93 cm). The S_1T_1 was at par with S_1T_4. In case of S_2 the maximum leaf width was recorded in S_2T_4 (20.19) and minimum in S_2T_3 (17.79 cm) (Table1).The individual tree selection can be made in each site. The promising tree in S_1 are S_1T_1 and S_1T_4.

Table 1: Variation in leaf characteristics

Tree Number	Leaf length(cm)		Leaf width (cm)	
	S_1(Jabli)	**S_2(Jorhji)**	**S_1(Jabli)**	**S_2(Jorhji)**
T_1	24.21	17.81	25.80	18.80
T_2	18.00	19.43	19.22	20.02
T_3	19.37	16.92	21.37	17.79
T_4	22.51	19.44	24.15	20.19
T_5	16.67	17.49	17.93	19.18
Mean	**20.15**	**18.22**	**21.69**	**19.20**

$CD_{0.05}$

Site	1.07	1.2
Tree within site $_1$	2.40	2.86
Tree within site $_2$	NS	NS

4.1.1.3 Leaf area (cm^2)

Significant variation was found between sites with respect to leaf area. In S_1 (Jabli) mean leaf area was (349.10 cm^2) and in S_2 (Jorhji) it was(237.94 cm^2). In S_1 the maximum leaf area was recorded in S_1T_1 (542.13 cm^2) and minimum in S_1T_5 (220.92 cm^2). In S_2 the maximum leaf area was found in S_2T_4 (328.90 cm^2) and minimum in S_2T_5 (208.89 cm^2) . For both sites tree to tree variation with respect to leaf area was found to be significant (Table.2).

4.1.1.4 Petiole length (cm)

In S_1 (Jabli) the mean petiole length was 9.46 cm and in S_2 (Johrji) it was 7.33 cm. The maximum and minimum petiole length in S_1 was found in S_1T_1 (12.69 cm) and S_1T_5 (7.46 cm) respectively, where as in S_2 the maximum and minimum was found in S_2T_4 (8.71cm) and in S_2T_5 (6.55 cm). The petiole length was found to vary significantly between

Plate 1. Leaf length and width in *Bauhinia vahlii*

Plate 2. Pod length and width in *Bauhinia vahlii*

sites and also for the trees within site$_1$, the results have been found to be nonsignificant in site$_2$ for this character.(Table.2).

The trees viz. S_1T_1,S_1T_4 and S_2T_4 found superior for leaf length, leaf width and leaf area which increases the utilization scope of the species in donas, pattal and plates making. In this species the leaf size varies from tree to tree and also within the same tree. Present findings are consistent with report of Khan and Zaki (2015) in *Bauhinia racemosa* as they have explained that leaf area within individual tree varies from 0.5 to 18cm^2. The leaf area is directly related to light interception, photosynthesis, transpiration and carbon gain and storage. It is considered to be the most important single determinant of plant productivity (Kathirvelan and Kalaiselvan, 2007).

Table 2: Variation in leaf characteristics

	Leaf area (cm^2)		**Petiole length(cm)**	
Tree Number	**S_1 (Jabli)**	**S_2 (Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**
T_1	542.13	217.99	12.69	7.17
T_2	226.80	215.80	7.91	7.07
T_3	328.32	216.55	9.00	7.17
T_4	427.35	328.99	10.22	8.71
T_5	220.92	208.89	7.46	6.55
Mean	**349.10**	**237.64**	**9.46**	**7.33**

$CD_{0.05}$

Site	18.22	0.75
Tree within site $_1$	40.74	1.75
Tree within site $_2$	40.74	NS

4.1.2 Floral characteristics

Floral morphology. The data was taken with respect to flower size, ovary length, style length, filament length, petal length, hypanthium length, pedicel length and mean number of flowers per inflorescence.

4.1.2.1 Ovary length (mm)

The results were found significant between sites as well as between trees within both sites (Table.3). In case of S_1 (Jabli) the mean ovary length was (6.68 mm) and in S_2(Jorhji) it was 6.75 mm. In S_1 the maximum value was found in S_1T_1 (8mm) and minimum in S_1T_2, $S_1T_3$and S_1T_4 (6mm), where as in S_2 the maximum and minimum was obtained for S_1T_1 (7mm) and S_2T_2 (5mm) respectively.

Table 3: Variation in floral characteristics

Ovary length (mm)			Style length (cm)	
Tree Number	**S_1(Jabli)**	**S_2(Jorhji)**	**S_1 (Jabli)**	**S_2(Jorhji)**
T_1	8.33	7.93	1.95	1.77
T_2	6.00	5.47	1.73	1.71
T_3	6.47	7.47	1.69	1.79
T_4	6.33	7.67	1.72	1.90
T_5	6.27	5.23	1.83	1.84
Mean	**6.68**	**6.75**	**1.78**	**1.80**

$CD_{0.05}$		
Site	0.12	NS
Tree within site $_1$	0.27	NS
Tree within site $_2$	0.27	0.70

4.1.2.2 Style length (cm)

The result for the style length were found to be non significant between the sites, however the variation between trees for the character was significant in Site$_2$. In S_1 (Jabli) the mean style length was 1.78 cm and in S_2 (Jorhji) it was 1.80 cm. Within S_1 the maximum and minimum values registered by S_1T_1 (1.95 cm) and S_1T_3 (1.69 cm). In S_2 the maximum value was found in S_2T_4 (1.90 cm) and minimum in S_2T_2 (1.71 cm) (Table.3).

4.1.2.3 Filament length (cm)

The mean filament length for S_1 (Jabli) and S_2 (Jorhji) was 2.33 cm and 2.18cm respectively. In S_1 the maximum filament length was found in S_1T_2 (2.65 cm) and minimum in S_1T_1 (1.83cm). In S_2 the maximum and minimum was registered by S_2T_4 (2.47cm) and in S_2T_1 (4.55cm). Statistical analysis revealed significant variation between sites as well as between trees of both sites (Table.4).

4.1.2.4 Hypanthium length (mm)

The results were found to be non significant between sites and also between trees within site$_1$, however the variation was significant between trees of site$_2$.The mean hypanthium length in S_1 (Jabli) was 4.79 mm and in S_2 (Johrji) it was 5.09 mm. Within S_1 the maximum hypanthium length was found in S_1T_5 (5.00 mm) and minimum in S_1T_2

Plate 3. Stages of floral bud development in *Bauhinia vahlii*

(4.53mm). However in S_2 the maximum and minimum was recorded for S_2T_5 (5.08 mm) and S_2T_2 (4.63 mm) respectively (Table.4).

Table 4: Variation in floral characteristics

	Filament length (cm)		Hypanthium length (mm)	
Tree Number	**S_1(Jabli)**	**S_2(Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**
T_1	1.83	2.35	4.53	4.73
T_2	2.65	1.60	4.83	4.63
T_3	2.39	2.22	4.83	4.93
T_4	2.55	2.47	4.73	5.30
T_5	2.21	2.25	5.00	5.80
Mean	**2.33**	**2.18**	**4.79**	**5.08**

$CD_{0.05}$

Site	0.12	NS
Tree within site $_1$	0.27	NS
Tree within site$_2$	0.27	0.70

4.1.2.5 Number of flowers per inflorescence

The variation for this parameter is found to be significant for sites and also for the trees within site$_2$. The mean number of flowers per inflorescence in S_1 (Jabli) and S_2 (Johrji) were 41.11 and 45.91 respectively. In S_1 the maximum and minimum values with respect to number of flowers per inflorescence was found in S_1T_5 (44.13) and S_1T_4 (38.20) respectively. Similarly S_2T_4 (50.47) and S_2T_5 (37.60) registered maximum and minimum values in S_2 (Jorhji) and the trees namely S_1T_1, S_2T_2 and S_2T_3 was found at par with S_2T_4 (Table.5).

4.1.2.6 Pedicel length (cm)

In S_1 (Jabli) the mean pedicel length was 3.16 cm and in S_2 (Jorhji) it was 3.02cm. Among the trees within S_1,the maximum pedicel length was found in S_1T_2 and S_1T_4 (3.27 cm) and minimum in S_1T_1 (3.03 cm), where as in S_2 the maximum and minimum values was recorded in S_2 T_3 (3.23 cm) and S_2T_5 (2.93 cm). Variation in pedicel length was found to be non-significant between sites as well as between trees of both sites (Table.5).

Table 5 : Variation in floral characteristics

No. of flowers per inflorescence			Pedicel length(cm)	
Tree Number	**S_1 (Jabli)**	**S_2 (Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**
T_1	40.67	49.93	3.03	2.95
T_2	38.87	44.97	3.27	2.99
T_3	43.67	46.60	3.17	3.23
T_4	38.20	50.47	3.27	3.01
T_5	44.13	37.60	3.06	2.93
Mean	**41.11**	**45.91**	**3.16**	**3.02**

$CD_{0.05}$		
Site	2.60	NS
Tree within site $_1$	NS	NS
Tree within site $_2$	5.81	NS

4.1.2.7 Larger petal length (cm)

In this species, the petals are found to vary in their size. Three petals were observed to be larger while the other two were smaller. For the larger type of peals, mean petal length for S_1 (Jabli) and S_2 (Jorhji) was 2.15cm and 2.20cm respectively. In S_1 the maximum petal length was found in S_1T_5 (2.47 cm) which differ significantly from minimum value found in S_1T_1 (2.00 cm). The results for larger petal length were found to vary significantly for the sites as well as trees within the sites (Table.6).

4.1.2.8 Smaller petal length (cm).

The mean smaller petal length in S_1 (Jabli) was 1.68 cm which differ significantly from the mean petal length of S_2 (Johrji) where it was 1.74 cm. Between tree variation was found significant in S_1 (Jabli), where S_1T_2 and S_1T_4 registered maximum (1.87 cm) and minimum (1.45 cm) values respectively. In S_2 the maximum and minimum petal length was found in S_2T_3 (1.85) and in S_2T_1 (1.55 cm) respectively and these results were significant for the trees within both sites (Table.6).

The results obtained for the floral parameters revealed that the length of gynoecium is slightly larger than the length of androecium which find its importance in outcrossing mechanism and ovary length directly related to the number of ovules. Each flower bear pedicel and number of flowers varies from inflorescence to inflorescence. Similar results for floral parameters of this species has been reported by Chauhan *et.al* (2013).

Table 6: Variation in floral characteristics

	Larger petal length(cm)		Smaller petal length(cm)	
Tree Number	**S_1 (Jabli)**	**S_2 (Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**
T_1	2.16	2.00	1.64	1.55
T_2	2.24	2.12	1.87	1.65
T_3	1.98	2.33	1.61	1.85
T_4	1.90	2.24	1.45	1.80
T_5	2.47	2.33	1.81	1.82
Mean	**2.15**	**2.20**	**1.68**	**1.74**

$CD_{0.05}$

Site	0.03	0.03
Tree within site $_1$	0.08	0.07
Tree within site $_2$	0.08	0.07

4.1.3 Pod characteristics

4.1.3.1 Pod length (cm)

Data on mature pods were collected from each tree in both sites. In S_1 (Jabli) the mean pod length was 21.80 cm and in S_2 (Jorhji) it was 16.71 cm. In S_1 the maximum pod length was found in S_1T_1 (25.80 cm) and minimum in S_1T_5 (17.93 cm) ,where as in S_2 the maximum was in S_2T_2 (18.69 cm) and minimum in S_2T_1 (15.65 cm).(Table.7).The results were found to be significant between sites and also for the trees within site$_1$.

4.1.3.2 Pod width(cm)

In S_1(Jabli) the mean pod width was 5.25cm which was significantly higher than the mean pod width of S_2 (Jorhji) where it was 4.29cm. In S_1 the maximum pod width was found in S_1T_1(4.70cm) which differ significantly from the minimum value of S_1T_5 (4.50cm). In S_2 the maximum and minimum was recorded in S_2T_2 (4.93) and S_2T_1 (3.87cm) respectively (Table.7).

The fruit of this species is legume and production depends upon the number of flowers visited by pollinators. In present study the pod size varies from 15 to 22 cm in length and 3to 5cm in width. Similar result of this species was explained by Narayan *et.al*.(2013) that fruit is a flat woody pod with fine rusty hairs, 20-30cm long and 5to 7.5cm broad. In *Bauhinia purpurea* Orwa *et. al*.(2009) found that fruit is brown, elongated dehiscent pods, 15- 30 cm long and up to 1.5-2.5 cm wide.

Table 7: Variation in pod characteristics

	Pod length(cm)		Pod width(cm)	
Tree Number	**S_1 (Jabli)**	**S_2 (Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**
T_1	25.80	15.65	5.70	3.87
T_2	19.22	18.69	5.17	4.93
T_3	21.37	15.79	5.40	4.00
T_4	24.69	17.59	5.50	4.50
T_5	17.93	15.85	4.50	4.17
Mean	21.80	16.71	5.25	4.29

$CD_{0.05}$

Site	1.34	0.40
Tree within site $_1$	2.99	0.91
Tree within site $_2$	NS	NS

4.2 FLORAL BIOLOGY

The inflorescences of *Bauhinia vahlii* were found to be a terminal corymbose raceme. The mean number of flower per inflorescence ranged from 38.20 to 50.47. The flower maturity is characterized by peripheral initiation advancing towards the center.

4.2.1 Calyx and corolla

Biserate calyx and corolla was found in discrete whorl. The number of member in each whorl was found to be different, hence they are in an isomerous merosity. (Table.8)

4.2.2 Androecium

On careful examination of the androecium the stamens were found to have large filament borne inbetween petal or opposite to sepal. The number of stamens were three and free form each other. Anther attached dorsally and medially to the filament and can rotate in 45^0 (versatile) and dehisce longitudinally (Table.8).

4.2.3 Gynoecium

Gynoecium was unicarpellous as all sepal, petal,stamen and hypanthium were attached to the base of the ovary (ovary superior) and had marginal placentation. Style was reddish pink with green and discoid stigma (Table.8).

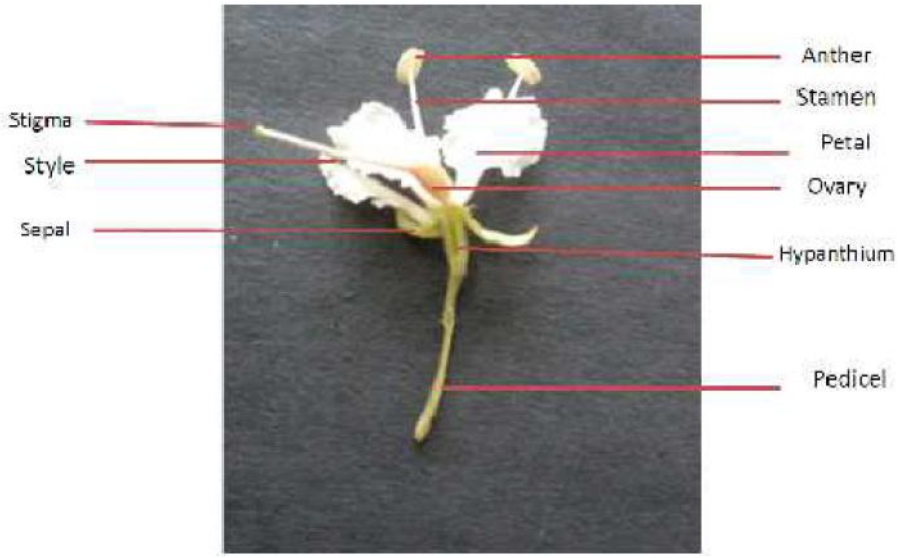


Plate 4. Flower parts of *Bauhinia vahlii*

Apostemonous

Versatile/Dorsifixed

Marginal plancentation

Ovary superior

Plate 5. Anderocium and gynoecium

Table 8: Floral biology

Flower	**Inflorescense type**	**Flower type**	**Symmetry**	**Development**	**Flower sex**
	Corymbose raceme	Pedicellate	Zygomorphic	Indeterminate	Bisexual
Perianth	**Perianth cycly** Biseriate	**Merosity** Anisomerous	**Perianth fusion** Aposepalous/apopetalous	-	-
Androecium	**Stamen type** Filamentous	**Stamen cycly** Antisepalous /alteripetalous	**Stamen fusion** Apostemonous	**Anther Attachment** versatile (dorsifixed)	-
Gynoecium	**Gynoecial fusion**	**Ovary attachment**	**Placentation**	**Style position**	-
	Unicarpellous	Ovary superior	Marginal	Discoid	

The study of floral morphology find its importance in fulfilling the objectives of tree breeding and also in connection with taxnomical perospective. The present study on floral morphology revealed that *Bauhinia vahlii* produced a terminal corymbose raceme, bearing a number of perfect flowers. The mean number varying from 38.20 up to 50.47.On the raceme the flower maturity begin from periphery followed by those which keep on appear subsequently and towards the center of the corymb. The flower showed protogynous dichogamy. In each flower the number of stamens was three along with two stamenodes. The calyx and corolla was biserate and number of member in each whorl was three and five respectively. The length of gynoecium was larger than anderoecium. The filaments was free and borne in between petal or opposite to sepal. Anther can rotate in acute angle which is the characteristic feature of the entomophilous species. The ovary was found to be superior and had marginal placentation. The colour of style was raddish pink connected with green discoid stigma. These results was found parallel with the results of Chauhan *et.al.* 2004 in *Bauhinia vahlii* and Darathy (1951) in Guava, Torres *et.al.* (2007) in *Senna cana* and Solomon *et.al.* in *Terminalia pallida.* Smitha and Thondaiman (2016) in *Saraca asoca* (Roxb.) the species bears fragrant flowers in paniculate corymbose inflorescence.

4.2.4 Time of anthesis

Anthesis is the opening of the floral bud and to find out the time of anthesis observations were taken in three different time interval at both the sites. Maximum percent anthesis took place between 6 to 9 am and minimum between 12 noon to 3 pm. In S_1 (Jabli) the mean percent anthesis recorded in $Time_1$,$Time_2$ and $Time_3$ was 62.66, 22.6 and 14.67 respectively, where as in S_2 the mean percent anthesis in three different timings was 59.99, 28.00 and 13.33 respectively.

S_1T_2 and S_1T_5 registered maximum (37.77) and minimum (28.89) mean percent anthesis in S_1 (Jabli), where as in S_2 (Jorhji) the maximum and minimum mean percent anthesis was found in S_2T_1 (40.00) and S_2T_5 (31.11) (Table.9). The finding on the time of anthesis conclude that the day temperature has significant role in governing the mechanism of anthesis. Mode of anthesis was observed from the moment petals begin to open till the flower attained complete open position. It was found that first a small slit appeared on the top of the bud, stigma started protruding out of the bud, petal gradually separated from each other after an hour flower become fully opened.

Anthesis is the opening of floral bud which is essential for progressive reproductive process. *Bahunia vahlii* was found to be among those species responding to summer dawn conditions favouring anthesis beginning with sunrise. During the course of study anthesis in this species found to be maximum between 6am to 9am.The results of this study are supplemented by the findings of Balalia and Chauhan (1994) in *Delonix regia* where they found that maximum anthesis occurred between 6am to 7am. Chauhan *et.al.* also found same result in *Dalbergia sissoo*. In *Senna cana* Torres *et.al.* (2008) has reported the time of anthesis as around 7am around. Wani and Chauhan (2008) in *Bauhinia variegata.* Smitha and Thondaiman (2016) noticed in *Saraca asoca* (Roxb.) that anthesis in this species occurred in the early morning from 3.00 to 5.30 am. Srivastava (1983) reported that in *Butea monosperma* anthesis occurs between 5-6am. Bhattacharjee (1985) in *Jacaranda* noted that 99 per cent of the flower opening occurs between 5-7am.

Table 9: Opening of the floral bud during different timings of the day(%)

Tree Number	S_1(Jabli)				S_2 (Jorhji)			
	$Time_1$ (6am-9am)	**$Time_2$ (9am-12noon)**	**$Time_3$ (12noon-3pm)**	**Mean**	**$Time_1$ (6am-9am)**	**$Time_2$ (9am-12noon)**	**$Time_3$ (12noon-3pm)**	**Mean**
T_1	59.99	20.00	13.33	31.11	73.33	33.33	13.33	40.00
T_2	66.66	33.33	13.33	37.77	53.33	26.66	13.33	31.11
T_3	73.33	13.33	13.33	33.33	59.99	26.66	13.33	33.33
T_4	60.00	26.66	20.00	35.55	53.33	26.66	20.00	33.33
T_5	53.33	20.00	13.33	28.89	59.99	26.66	6.67	31.11
Mean	62.66	22.66	14.67		59.99	28.00	13.33	

$CD_{0.05}$

Between time of the site	11.0	11.38
Between trees	NS	NS
Time x trees	NS	NS

Stage 1 Stage 2

Stage 3 Stage 4

Plate 6. Floral bud development in *Bauhinia vahlii*

Lab. conditions Field conditions

Plate 7. Anther dehiscence

4.2.5 Anther dehiscence

Anther dehiscence observed under dissection microscope, in field hand lens was used for observations. In this species number of stamen in each flower was three along with two stamenodes. Observation were take in three different time interval and it was found that anther dehisce longitudinally. Maximum percent dehiscence at both sites was found in $time_1$ (11am to 1 pm) and minimum in $time_3$ (1 pm to 3 pm). In site one (Jabli) the mean percent anther dehiscence in three different time interval was 44, 57.33 and 10.67. In S_2 (Johrji) percent anther dehiscence was 38.6, 54.66 and 17.33. The maximum mean percent anther dehiscence in S_1 was found in S_1T_3, S_1T_4 and S_1T_5 (40.00) and minimum in S_1T_1 and S_1T_2 (33.33). Similarly S_2T_1 (40.00) and S_2T_2 (33.33) registered maximum and minimum percent anther dehiscence in $site_2$. In both site anther dehiscence showed significant variations with respect to time (Table.10).

Table 10: Anther dehiscence during different timings of the day (%)

Tree Number	S_1(Jabli)				S_2(Jorhji)			
	$Time_1$ (9am-11am)	**$Time_2$ (11am-1pm)**	**$Time_3$ (1pm-3pm)**	**Mean**	**$Time_1$ (9am-11am)**	**$Time_2$ (11am-1pm)**	**$Time_3$ (1pm-3pm)**	**Mean**
T_1	40.00	53.33	6.67	33.33	40.00	59.99	20.00	40.00
T_2	33.33	53.33	13.33	33.33	40.00	46.66	13.33	33.33
T_3	46.66	60.00	13.33	40.00	40.00	53.33	20.00	37.77
T_4	46.66	59.99	13.33	40.00	33.33	59.99	13.33	35.55
T_5	53.33	59.99	6.67	40.00	40.00	53.33	20.00	37.77
Mean	44.00	57.33	10.67		38.66	54.66	17.33	

$CD_{0.05}$

Between time of the site	11.85	14.04
Between trees	NS	NS
Time x trees	NS	NS

Results were supplemented by Chauhan *et.al.* (1999) in *Tamarindus indica* where anther dehisce between 9.00am to 11.00am. Wani (2008) in *Bauhinia variegata* also observed anther dehiscence in the morning hour. Aguiara *et.al.* (2016) in *Cenostigma macrophyllum* reported that anther dehisce took place near around 9am. Anther dehisce between 5 to 6am in *Butea monosperma* (Srivastava 1983). Sareen and Vashist (1982) has reported the similar timing of anther dehiscence taking place between 10:20 to 11:30am in *Delonix regia* and the type of dehiscence as observed was through the longitudinal slits anther dehisced longitudinally between 10:30 am – 11:30am.

4.3 BREEDING SYSTEM

4.3.1 Pollen collection

Inflorescences were collected from each tree. In laboratory conditions, anthers were removed with forceps on butter paper and kept under partial shade to allow the anther dehiscence. Pollen grains were collected in glass vial and stored at low temperature to check the viability.

The amount of pollen produced by any species is determined by complex factors, including age, nutrient status and environmental conditions during the time of pollen development (Stanley and Linskens,1974) . For effective use in tree breeding, large quantities of pollen produced by selected genotype are desirable. Most of the procedure involves isolating flowers, indicating dehiscence and collection of released pollen for storage and eventual applications in breeding. The present investigations were carried with the objectives of collecting large quantities of pollen using direct extraction method for future use in breeding program. The procedure adopted for collection is in agreement with the procedure advocated by Stanley and Linskens (1974) .

4.3.2 Pollen viability (%)

To make tree breeding programme pollen viability was checked at three different stages viz. before anthesis , at the time of anthesis and in fully opened flowers. In both sites maximum pollen viability was found in stage 1 (before anthesis) and minimum in stage 3 (fully opened flower). In S_1 (Jabli) the mean percent pollen viability in three different stages was 95.43, 90.55 and 83.87 respectively. In case of S_2 (Jorhji) the mean percent pollen viability in three different stages was 94.73, 89.51 and 86.17 respectively.

Table 11: Variation in pollen viability(%)

Tree Number	Before anthesis		At the time of anthesis		Fully opened flower	
	S_1 (Jabli)	**S_2 (Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**
T_1	94.60	94.13	92.40	94.03	83.03	83.70
T_2	97.43	93.53	90.50	91.90	73.33	86.40
T_3	93.87	95.43	90.90	88.60	85.70	92.60
T_4	96.07	95.73	91.77	86.23	82.33	84.57
T_5	95.17	94.80	87.20	86.78	94.93	83.57
Mean	95.43	94.73	90.55	89.51	83.87	86.17
$CD_{0.05}$						
Site	NS		NS		NS	
Tree within site $_1$	NS		NS		5.23	
Tree within site $_2$	NS		4.26		5.23	

The results for pollen viability before anthesis were non significant between the sites. However the percent viability was maximum in S_1T_2(97.43) which is higher than the viabilities recorded for the other two stages of pollen collection. At the time of anthesis in S_1 the maximum and minimum pollen viability was found in S_1T_1 (92.40) and S_1T_5 (87.20) and the difference was non significant between all the tress within site$_1$. In case of S_2 the difference in pollen viability were found to be significant at the time of anthesis and highest value was found in S_2T_2 (94.03) and minimum in S_2T_4 (86.23). In stage 3 namely fully opened flower; the results were non significant for the two sites but significant for the trees within each site. The maximum viability was recorded in T_5S_1(94.93) and minimum S_1T_5 (94.93) and S_1T_2 (73.33). Similarly in S_2 the maximum and minimum was recorded in S_2T_3 (92.60) and S_2T_5 (83.57) respectively(Table.10).

The results on pollen viability are compatible with the study of Gozleki (1997), Nath and Randhawa (1959), Josan *et.al.* and Sharma and Bist (2005) in Wild pomergernate. Srivastava and Kumar (1982) in *Maughania macrpphylla* where the pollen stored at room temperature remained viable for 72hr. Sunnichan *et.al.* (2009) *Boswellia serrata,* about 85% of the fresh pollen grains were viable.

4.3.3 Pollen germination (%)

Pollen germination was studied from the sample collected on individual tree basis for both sites in three different sucrose concentrations (5%, 15%and 20). Among different concentrations in both sites the maximum average germination percentage was observed in 15 % sucrose solution and minimum in 5 % sucrose solution. In site ×tree interaction maximum percentage observed was 80.3 in S_1T_2 and minimum was observed in S_1T_1(78.1%). In site× concentration interaction maximum average percentage observed was 84.3 with 15% sucrose solution at S_1(Jabli) and minimum was 73.6 percent at S_2 (Jorhji) with 5% sucrose solution. In tree × concentration interaction maximum percentage observed was 87.4 percent inS_2T_3 with 15% sucrose solution and minimum percentage was 71.7 percent in 5% sucrose solution in tree S_2T_1.

The present results are parallel to the findings of Shrivastava (1983) in *Butea monosperma,* Pal and Singh (1979) in *Solanum khasianum*, Malasi *et.al.* (1981) in *Berberis asiatica* and *Berberis lycium*, Nath and Randhwawa(1959) in *Punica granatum,* Bhattacharya and Mandal (2000) in *Bombax ceiba.*

Table 12: Pollen germination (%)

Tree Number	S_1 (Jabli)				S_2(Jorhji)			
	Pollen germination (%) in sucrose concentrations							
	5%	15%	20%	Mean	5%	15%	20%	Mean
T_1	76.3 (8.73)	81.4(9.02)	76.8(8.76)	78.1(8.84)	71.7(8.47)	86.7(9.31)	78.0(8.84)	78.8(8.87)
T_2	75.9(8.71)	85.3(9.24)	79.6(8.92)	80.3(8.96)	74.6(8.64)	86.6(9.31)	77.8(8.82)	79.7(8.92)
T_3	73.0(8.54)	87.4(9.35(	79.8(8.94)	80.1(8.94)	74.7(8.65)	90.6(9.52)	79.8(8.93)	81.7(9.03)
T_4	74.9(8.66)	82.9(9.11)	77.5(8.77)	78.5(8.85)	74.7(8.65)	85.7(9.26)	78.8(8.83)	79.7(8.83)
T_5	75.5(8.69)	84.3(9.18)	78.5(8.81)	79.4(8.91)	72.4(8.51)	87.0(9.33)	77.5(8.81)	79.0(8.88)
Mean	75.1(8.67)	84.3(9.18)	78.4(8.86)		73.6(8.58)	87.4(9.34)	78.4(8.85)	

$CD_{0.05}$

Between conc.	0.11	0.11
Between trees	NS	NS
Conc. x trees	NS	NS

4.3.4 Stigma receptivity

The change in the appearance of stigma colour was observed with hand lens from one day before anthesis up to next three days. It has been observed that stigma become receptive 2 hr after anthesis and remain receptive upto 48 hours. Dull green stigma was considered non-receptive and shiny green was receptive.

Stigma become receptive 2hr after anthesis. Similar results were observed by Randhawa and Das (1962) in *Grewia asiatica* where stigma was fully receptive on the day of anthesis. Srivastava *et.al.* (1987) in *Moghania chappar* and Pant *et.al.* (1997) in *Grewia optiva* where the receptivity of stigma range from 24hr before anthesis up to 12 hr after antheis. Chauhan *et.al.* (2004) in *Dalbergia sissoo* observed that stigma become receptive few hour before anthesis and remain receptive few hour after anthesis.

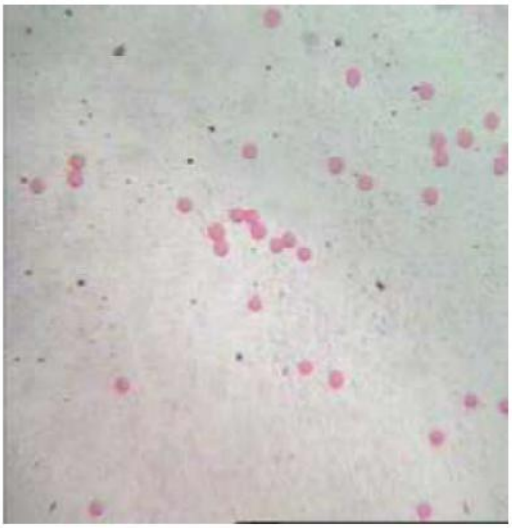
4.4.5 Pollination and pollen vectors

Those insects which used to visit the flowers collected and identified in consultation with the experts of the subject. The pollinators were identified as *Apis india*, *Apis dorsata* and Aphids.

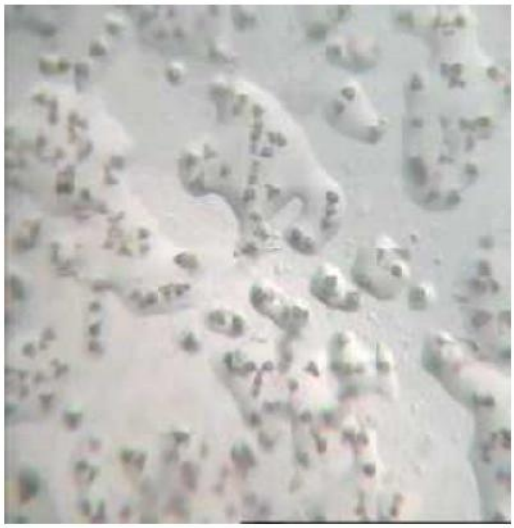
4.4 POLLINATION

4.4.1 Controlled pollination and fruit set.

In controlled pollination at each site fifteen buds where pollinated and bagged to avoid contamination and fruit set was recorded. In S_1 (Jabli) the maximum fruit set was found



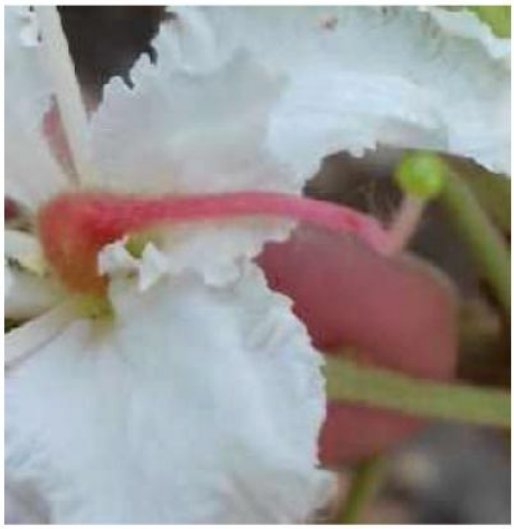
Viable pollens



Pollen germination

Plate 8. Pollen viability and pollen germination

1 day before anthesis



On the day of anthesis

1 day after anthesis

2 day after anthesis

Plate 9. Stigma receptivity

in S_1T_1 (40.00) and minimum in S_1T_4 (20.00), where as in S_2 (Jorhji) the maximum and minimum was found in S_2T_4 (46.67) and in S_2T_1 (26.67) respectively (Table.13).

Table 13: Controlled pollination and fruit set within two sites.

Tree Number	Site$_1$ (Jabli)			Site$_2$ (Jorhji)	
	No. of buds pollinated	No. of fruit set	Percent	No. of fruit set	Percent
T_1	15	6	40.00	4	26.67
T_2	15	4	26.67	6	40.00
T_3	15	4	26.67	5	33.33
T_4	15	3	20.00	7	46.67
T_5	15	5	33.33	6	40.00

4.4.2 Selfing

Self mechanism was studied by bagging the inflorescences at appropriate time on each tree and data on inflorescences were obtained on fruit setting by opening the bags , there was no fruit set in these inflorescences.

4.4.4 Controlled crossing between two sites

In controlled crossing attempted among different tress at both sites, in S_1 best cross combination were $S_1T_4 \times S_2T_3$ and $S_1T_5 \times S_2T_3$ where as in S_2 best cross combination were $S_2T_1 \times S_1T_3$, $S_2T_1 \times S_1T_5$, $S_2T_2 \times S_1T_1$, $S_2T_3 \times S_1T_1$, $S_2T_3 \times S_1T_2$. and $S_2T_3 \times S_1T_3$ (Table.14 & Table 15).

Table 14: Fruit set (%) under controlled crossing in Jabli (S_1)

♀ \ ♂	S_2T_1	S_2T_2	S_2T_3	S_2T_4	S_2T_5	Mean
S_1T_1	33.3(29.9)	50.0(44.9)	0.00	50.0(44.9)	50.0(44.9)	**36.6(44.98**
S_1T_2	33.3(29.9)	33.3(29.9)	0.00	50.0(44.98)	33.3(29.9)	**30.0(26.9)**
S_1T_3	33.3(29.9)	33.3(29.9)	0.00	33.3(29.9)	33.3(29.9)	**26.6(23.9)**
S_1T_4	0.00(0.00)	16.6(14.9)	66.6(59.9)	33.3(29.9)	16.6(14.9)	**26.6(23.9)**
S_1T_5	16.6(14.9)	33.3(29.9)	66.6(59.9)	50.0(44.6)	16.6(14.9)	**36.67(32.9)**
Mean	**23.3(20.9)**	**33.3(29.9)**	**26.6(23.9)**	**43.33(38.9)**	**30.0(26.9)**	

$CD_{0.05}$

S_1	NS
S_2	NS
$S_1 \times S_2$	30.30

Table 15: Fruit set (%) under controlled crossing in Jorhji (S_2)

♀ \ ♂	S_2T_1	S_2T_2	S_2T_3	S_2T_4	S_2T_5	Mean
S_2T_1	50.0(44.9)	16.6(14.9)	66.6(59.9)	16.6(14.9)	66.6(59.9)	**43.3(38.9)**
S_2T_2	66.6(59.9)	16.6(14.9)	16.6(14.9)	50.0(44.9)	33.3(29.9)	**36.6(32.9)**
S_2T_3	66.6(59.9)	66.6(59.9)	66.6(59.9)	0.00	16.6(14.9)	**43.3(38.9)**
S_2T_4	50.0(44.9)	16.6(14.9)	16.6(14.9)	33.3(29.9)	16.6(29.9)	**26.6(23.9)**
S_2T_5	0.00	0.00	0.00	33.3(29.9)	33.3(29.9)	**13.3(12.0)**
Mean	**46.6(41.9)**	**23.3(20.9)**	**33.3(29.9)**	**26.6(23.9)**	**33.3(29.9)**	

$CD_{0.05}$

S_1	NS
S_2	18.57
$S_1 \times S_2$	41.52

In *Bauhinia vahlii*, pollination studies revealed that hand cross pollination resulted into higher pod setting. The results are supplemented by Sarsaki and Gibbs (1997) who found that in *Dalbergia miscolobium* fruit set was higher under controlled cross pollination method. Gibbs and Sarsaki (1997) also found that cross pollination is efficient fruit set method for *Bauhinia corymbosa.* Bawa *et.al.* (1985) found that majority of tree species in the rain forest appear to be obligate outcrosser. Pant (1997) observed that *Grewia optiva* is predominantly, a cross pollinated species. Wani and Chauhan (2008) reported outcrossing of *Bauhinia variegate.*

In *Bauhinia vahlii* the present investigation concluded that it is entomophilous and depends upon external vectors for fertilization. Flowers were visited by bees and flies among which two bees namely *Apis dorsata* and *Apis indica* was the dominanat pollinator vistors. Similar results were recorded by Lau *et.al.* in *Bauhinia championii* and *Bauhinia gluca* and Hicks *et.al.* (1985) in *Mitchella repens.*

As the study concluded that no fruit set occurred in self pollination experiment. Keeping in view the architecture of the flowers, where the position of the style describe outcrossing mechanism supported by protogyny. The style in the flowers faces opposite direction in context to stamens and attained slightly longer length which is evoulationary process for out crossing. Our results were pillered by Lau *et.al.* (1967) in *Bauhinia championii* and Rameriz *et.al.* in *Bauhinia angulata* which is genetically self – incompatible and functionally andromonoecious. *Vatairea macrocarpa* is self-incompatible as it produced no fruits under manual selfing and apomixes (Costa *et.al*. 2014). In *Bauhinia curvula* Munin

Selection of bud | Opening of bud | Emasculation | Dusting

Bagging | Pod size after 7 days | Pod size after 15 days

Plate 10. Controlled crossing

Apis indica | *Apis dorsata* | Aphid

Plate 11. Pollinators

et.al. (2008) found that the species was self incompatible and depends on pollinators and did not bear fruit after spontaneous self pollination.

Under natural conditions fecundity is obtained for both sites from the mother trees which was found to be low. There are various reasons for low fecundity mainly due to insufficient pollens production, temperature variation less population of the pollen vectors etc which render number of flowers un-pollinated.

4. 5 SEED CHARACTERISTICS, GERMINATION AND NATURAL REGENERATION

4.5.1 Variation in seed weight (gram).

To determine variation in seed weight from each tree mature pods were collected for seed extraction. 100 seed weight was taken individually from each tree in which maximum seed weight was found in S_1T_1 (138.85g) and minimum in S_1T_4 (112.55 g). In S_2 the maximum and the minimum seed weight was found in S_2T_4 (140.54g) and in S_2T_1 (105.98 g). Seed weight was found to vary significantly from tree to tree in both sites (Table.16).

4.5.2 Seed germination (%)

Seeds were collected and sowing was done in poly bags under controlled condition. S_1T_5 (82%) and S_1T_2 (46 %) registered maximum and minimum germination percentage for S_1 (Jabli), where as in S_2 (Johrji) the maximum and the minimum germination percentage was found in S_2T_2(68%) and S_2T_4 (26%) (Table.16).

Table 16: Variation in seed traits

Tree Number	Seed weight (g)		Seed germination (%)	
	S_1 (Jabli)	**S_2 (Jorhji)**	**S_1 (Jabli)**	**S_2 (Jorhji)**
T_1	**138.85**	**105.98**	62(51.95)*	64(53.16)
T_2	123.59	133.04	46(42.69)	68(55.94)
T_3	132.98	118.97	56(48.46)	62(51.95)
T_4	**112.55**	**140.54**	54(47.31)	26(30.64)
T_5	131.27	138.13	82(64.93)	32(33.20)
Mean	**127.85**	**127.33**	**60(50.76)**	**50(45.22)**

*The value in the parenthesis are sign transformed values

$CD_{0.05}$

Site	NS
Tree within site $_1$	5.76
Tree within site $_2$	5.76

The results obtained are found parallel with Smitha and Das (2016) in seed germination of *Saraca asoca.* Azad (2013) seed germination of *Tamarindus indica*. Azad *et.al.* (2012) in seed germination of *Albizia procera*. Mwase and Mvula (2011) on seed germination and seedling growth of *Bauhinia thonningii.*

4.5.3 Natural regeneration

In natural regeneration survey was conducted for new recruits, unestablished and established regeneration the data collected from both sites is presented in Table. 17.

These results are supported by Lal *et.al.* (2015) in Barnawapara wildlife sanctuary, Chhattisgarh and concluded that closed natural forest displayed the better regeneration. Guevarra and Florece (2013) acquired that serious infestation on *Bauhinia malabarica* seeds and low viability limits the regeneration of the species in the watershed. Pawar *et.al.* (2012) found that in tropical deciduous forest of Chhattisgarh the least disturbed site was good regenerating because of high density of seedlings. In *Acacia tortalis* Noumi *et.al.* (2010) reported that infestation of seeds by the pests viz.*Bruchidius raddianae* and *B. aurivillii.* were the main obstacles to natural regeneration.

Table 17: Natural regeneration

Regeneration	**S_1(Jabli)**	**S_2(Jorhji)**
Recurits/ha	300	700
unestablished/ha	1000	1300
Established /ha	500	1700

4.6 GENETIC ESTIMATES

4.6.1 Genotypic coefficient of variability. (GCV%) Maximum GCV was obtained for leaf area (38.54%).

4.6.2 Phenotypic coefficient of variability (PCV %). Table 18 depicted similar pattern with maximum phenotypic coefficient of variability (39.38%) for leaf area .

4.6.3 Heritability. The maximum value of heritability (Broad sense) was found for leaf area (0.96)

4.6.4 Genetic advance. Maximum value of the genetic advance was obtained for leaf area (77.00) .

4.6.5 Genetic gain (%) . Maximum genetic gain was found for leaf area 38.9(%) percent

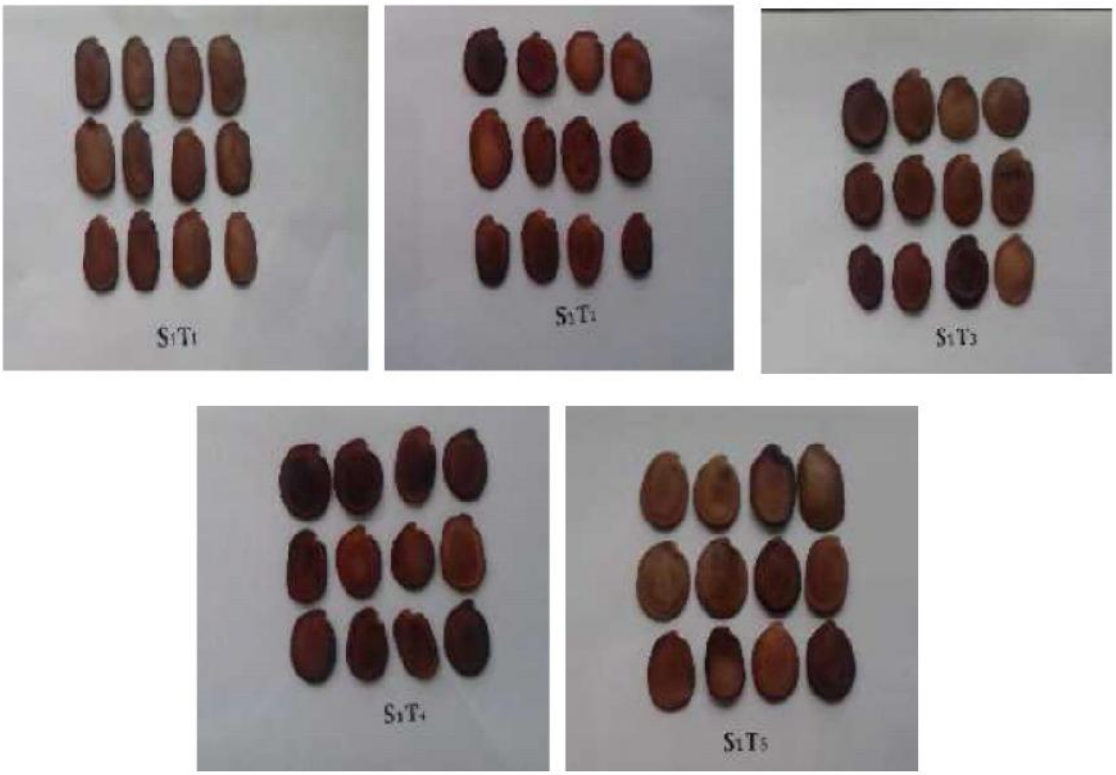


Plate 12. Seed colour in S_1 (Jabli)

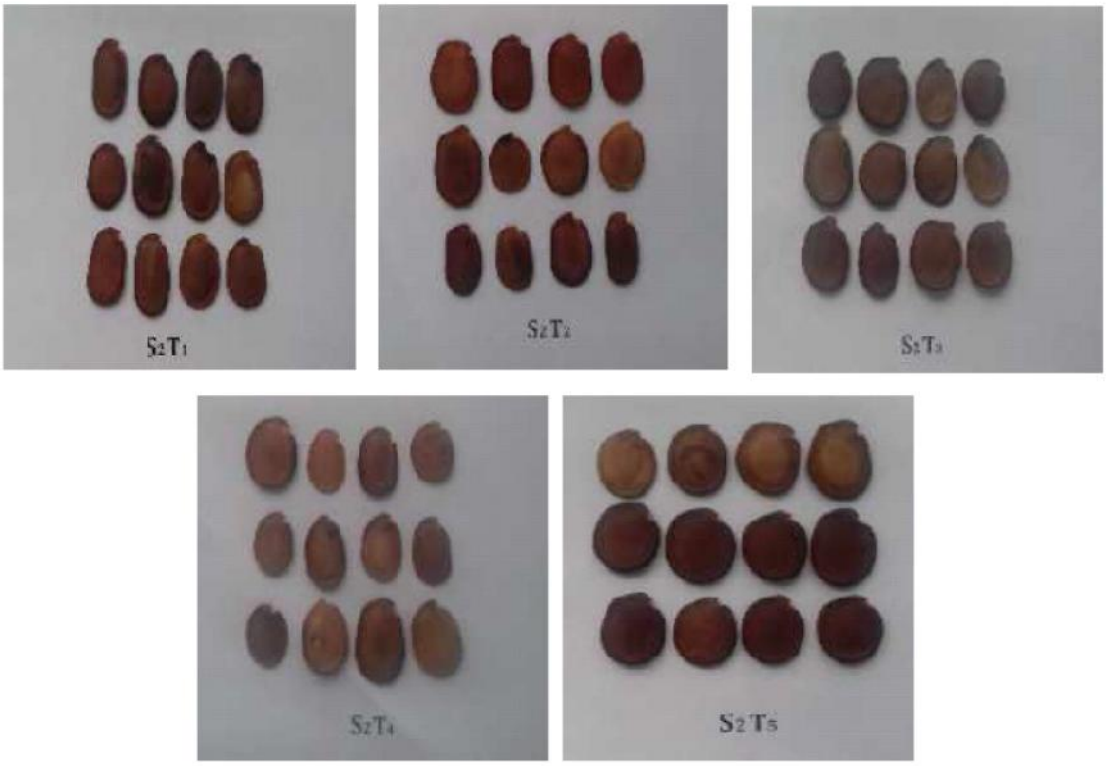


Plate 13 Seed colour in S_2 (Jorhji)

Plate 14. Seed germination and development

Table 18: Genetic estimates

Characters	GCV (%)	PCV (%)	H	GA	GG
Leaf area	38.54	39.38	0.96	77.00	38.9
Leaf length	12.06	14.10	0.73	4.07	21.23
Leaf width	12.04	14.49	0.69	4.21	20.60
Ovary length	15.28	17.03	0.80	1.90	28.26
No. of flowers	9.69	12.44	0.61	6.77	15.56
Filament	13.67	15.44	0.78	0.56	24.94
Style	3.53	7.14	0.24	0.06	3.59
Petal(L)	8.06	8.40	0.92	0.35	15.92
Petal(S)	8.33	8.68	0.92	0.28	16.46
Pod length	18.09	20.23	0.81	6.41	33.31
Hundred Seed weight	9.15	9.53	0.92	23.12	18.12

The observations on various traits along with their phenotypic variance, genotypic variance, heritability, genetic gain and genetic advance are presented in table no.18. It is apparent that the higher phenotypic coefficient of variance and genotypic coefficient of variance was found in all the traits. The highest PCV and GCV was found for leaf area(39.38, 38.54) followed by pod length (20.23, 18.04), ovary length (17.03, 15.28) and filament length (15.54, 13.67). The efficiency of selection on the magnitude of variability, present in the material depends on the extent to which the desirable characters are inherited. Higher PCV than the corresponding GCV for all the characters indicated that the environment interacts with these characters. Burton and De Vane (1953) suggested that the study of GCV together with heritability gives the best picture of progress to be achieved through selection.

The maximum heritability was observed for leaf area (0.96) followed by pod length (0.81), ovary length (0.8 0) and filament length (0.78). Therefore, it can be inferred that these traits were under the strong genetic control (Dean and Burdon, 1991). The maximum genetic gain was obtained for leaf area (38.9) followed by pod length (33.31), ovary length (28.26) and filament length (24.94). Johnson *et.al*. (1955) pointed out that heritability estimates along with genetic gain is more useful than heritability alone, because the heritability estimates indicates only the effectiveness of selection of genotype based on phenotypic performance, but fails to indicate real genetic progress. Therefore, it can be concluded that the selection is quite desirable for the improvement of these traits.

Chapter-5

SUMMARY AND CONCLUSION

5.1 INTRODUCTION

Bauhinia vahlii Wight. and Arn. is one of the giant woody climber belonging to family Caesalpiniaceae. The species is distributed in the sub-Himalayan region up to 1200 m above mean sea level and also in Assam, Central India, Bihar, Eastern and Western Ghats. Collection of leaves is done in Madhya Pradesh, Orissa and Andhra Pradesh for 'pattal' and 'dona' making. The species flowers in April – June and fruits ripen in the month of March-April next year. Pod dehiscence, seed dispersal and natural regeneration begins with the premonsoon rains. Fruit is flat rusty coloured woody pod. Regeneration is poor in natural forest because, there is significant damage due to harvesting of leaves by the local people, for commercial purposes which not only damage the plants but also reduces its population. The inflorescence is terminal corymbose raceme. Calyx and corolla are biserate, anisomerous, aposeplous. The number of stamens are three along with two staminodes, borne in between petal or opposite to sepal. Gynoecium is unicarpellous with superior ovary.

Various parts of this species have medicinal uses namely; leaves as demulcent, edible seed as tonic and aphrodisiac, gum for medicines, bark yield tannin and leaves used as fodder, plate making (donas and pattal) and thatching .

5.2 OBJECTIVES OF THE STUDY

1. To study morphometric characters of the species.
2. To study floral biology and breeding system.
3. To study seed characteristics and natural regeneration.

5.3 METHODOLOGY

Present investigations entitled 'Reproductive biology of *Bauhinia vahlii* Wight. and Arn.' was carried out in the Department of Tree Improvement and Genetic Resources, University of Horticulture and Forestry Nauni, Solan, Himachal Pradesh. Two sites were selected to study morphometric traits, floral biology, breeding system, pollination mechanism, seed characteristics and natural regeneration. Trees were selected at both sites, to

study floral and leaf morphometric characteristic fifteen samples were taken and studied under three replicates. Pods were collected during March-April 2017, and seeds extracted by drying the pods in partial shade. After cleaning the seeds 100 seed weight was taken individually for each tree as per ISTA(1966) regulations. To study natural regeneration 25 quadrates were laid down at both sites and survey was taken on new recruits, unestablished and established regeneration. From each tree 50 seeds were sown in polythene bags under polyhouse conditions and data was recorded on germination percentage.

5.4 MAJOR FINDINGS

- In morphometric traits maximum leaf length, leaf width and leaf area was recorded in S_1T_1 with the values of 24.21cm, 25.80cm and 542.13cm^2 respectively.
- The flowers of *Bauhinia vahlii* are bisexual and zygomorphic , having superior ovary. The inflorescence is terminal corymbose raceme.
- The mean number of flowers per inflorescence ranged from 38.20 up to 50.47. The inflorescence maturity begin from periphery followed by those flowers which keep on appearing and growing towards the center of the corymb.
- The flowers has protogynous conditions of appearance and maturity. The number of stamens are three along with the conspicuous presence of staminodes.
- The calyx and corolla were biserate as the number of member in each whorl were three and five respectively. The length of gynoecium was larger than androecium. The filaments were free and borne in between petal or opposite to sepal. Anther can rotate in acute angle. The ovary was found to be superior and had marginal placentation. The colour of style was reddish pink connected with green discoid stigma.
- The mode of anthesis was in longitudinal fashion, maximum anthesis was recorded as 62.66 and 59.99 percent during 6.00am-9.00am in S_1 (Jabli) and S_2 (Jorhji) respectively. Anther dehiscence takes place after 2 to 3hrs of bud opening
- Stigma become receptive 2 hr after anthesis and remain receptive up to 48 hr after anthesis.
- The mode of pollination was observed to be open and no fruit set occurred under self pollination experiment. Fecundity was found to vary from 13.54%-13.61%.
- The main pollinators were identified as *Apis dorsata*, *Apis indica* and Aphids.
- The maximum seed germination was found in S_2T_5 with value of 82%.
- Natural regeneration was ample initially but survival was poor because of moisture deficit conditions in S_1(Jabli) as compared to site$_2$(Jorhji)

- Genotypic coefficient of variability , phenotypic coefficient of variability, heritability, and genetic advance was found maximum for leaf area, pod length, ovary length and filament length**.**

5.5 CONCLUSIONS

On the basis of present research work on *Bauhinia vahlii* following conclusions can be drawn :-

1. The species flower in the month of 2^{nd} week of April – 3^{rd} week of June.
2. Trees namely T_1S_1,T_4S_2,T_1S_4 and T_2S_2 were found superior for leaf length, leaf width and leaf area . The leaves of this species has enormous economic importance.
3. Inflorescence was in terminal corymbose raceme, characterised by peripheral maturation.
4. Flowers were white, turning buff with age, bixexual, zygomorphic , biserate , ovary superior , marginal placentation chacterized by protogyny.
5. Maximum anthesis and anther dehiscence occurred in the morning hours.
6. Maximum pollen germination was recorded in 15% sucrose solution .
7. Stigma become receptive two hour after anthesis and the receptivity period is up to 48hour.
8. The species is insect pollinated, fecundity is low and there was no seed setting under selfing.
9. The main pollinators were identified as *Apis indica*, *Apis dorsata* and Aphids
10. In case of controlled crossing the promising combinations for fruit set were found to be $S_1T_4 \times S_2T_3$ and $S_1T_5 \times S_2T_3$ for which the fruit set has been obtained as 66.6%. In the other site (Jabli) similar results were obtained from $S_2T_1 \times S_1T_3$, $S_2T_1 \times S_1T_5$, $S_2T_2 \times S_1T_1$, $S_2T_3 \times S_1T_1$ and $S_2T_3 \times S_1T_3$.
11. The best performance of individual trees for percent seed germination under lab conditions was recorded from S_1T_5(82%) and S_2T_2(68%). Natural regeneration was ample initially but seedlings establishment is affected by the natural and anthropogenic disturbance.
12. High heritability was recorded for leaf area, pod length, ovary length and filament length hence it can be used for further planning of the improvement work of this species.

Hence it can be concluded that *Bauhinia vahlii* can be improved on the basis of morphometric traits of the individual tree. Also the pedicellate flower type, corymbose raceme inflorescence characterized by protogynous conditions which offer convenient outcrossing mechanism. The reproductive biology of this species offer a vast potentiality for future improvement inter-specific and intra-specific breeding programmes.

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Title of the thesis	**:**	**"Reproductive Biology of *Bauhinia vahlii* Wight. and Arn."**
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ABSTRACT

The present investigations entitled "Reproductive Biology of *Bauhinia vahlii* Wight. and Arn." was carried out in the Department of Tree improvement and Genetic Resources, Dr. Y.S.Parmar University of Horticulture and Forestry Nauni,Solan (HP) during 2016-2017 by studying selected tree climbers of *Bauhinia vahlii* from two populations namely S_1(Jabli) in district Solan and S_2 (Jorhji) in district Sirmour of Himachal Pradesh. The type of inflorescence is terminal corymbose raceme. The flowers are zygomorphic, bisexual, pedicellate with superior ovary. At the flowering stage calyx has three sepals one free and two partially fused which also extend the physical support to the larger petals. Corolla consists of five petals; three being larger and two smaller. Maximum anthesis was recorded as 62.66 per cent and 59.99 percent during 6.00 am to 9.00 am in S_1(Jabli) and S_2(Jorhji) respectively. Anther dehiscence take place 2 to 3hrs after anthesis. The initiation of stigma receptivity was observed at about 2hrs after anthesis and remain receptive up to 48hour. This species was found to be open pollinated, mostly pollinated by insect vectors. Controlled crosses were made between the trees of two sites and maximum mean pod setting was recorded in S_2 (Jorhji).The pollen viability was found to be maximum from the buds, before anthesis. Pollinators were identified to be *Apis indica, Apis dorsata* and aphids. The individual tree variability was recorded for hundred seed weight. The tree No. S_1T_1 registered maximummean seed weight(138.85g) for $Site_1$(Jabli) and in case of $Site_2$(Jorhji) maximum mean seed weight (140.54g) was recorded from tree No.S_2T_4 which was at par with S_2T_2 and S_2T_5.The maximum seed germination(82%) was obtained from S_1(Jabli), under glasshouse conditions. The Genetic estimates for morphometric and floral traits showed high genetic coefficient of variability and higher heritability for leaf area, pod length, ovary length, and filament length which indicates that the selection for these traits can bring about effective and desirable improvement in the species by using appropriate mating designs.

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APPENDIX

Appendix –I. Variation in leaf length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.959853	NS
Treatment	9	18.01642	S
Between sites	1	27.99468	S
Within S1	4	29.53511	S
Within S2	4	4.003173	NS
Error	18	1.972698	
Total	29		

NS = Non Significant
S = Significant

Appendix –II. Variation in leaf width

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	1.288973	NS
Treatment	9	20.884	S
Between sites	1	46.67521	S
Within S1	4	32.47156	S
Within S2	4	2.84864	NS
Error	18	2.726721	
Total	29		

NS = Non Significant
S = Significant

Appendix –III. Variation in leaf area

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	272.3464	NS
Treatment	9	38913.34	S
Between sites	1	93177.22	S
Within S1	4	56401.57	S
Within S2	4	7859.155	S
Error	18	564.2251	
Total	29		

NS = Non Significant
S = Significant

Appendix –IV. Variation in petiole length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.14428	NS
Treatment	9	10.49741	S
Between sites	1	33.83532	S
Within S1	4	13.19451	S
Within S2	4	1.965827	NS
Error	18	1.050739	
Total	29		

NS = Non Significant
S = Significant

Appendix –V. Variation in pod length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	1.986813	NS
Treatment	9	39.44712	S
Between sites	1	194.1581	S
Within S1	4	34.66369	S
Within S2	4	5.5528	NS
Error	18	3.054428	
Total	29		

NS = Non Significant
S = Significant

Appendix –VI. Variation in pod width

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.120333	NS
Treatment	9	1.299111	S
Between sites	1	6.912	S
Within S1	4	0.642667	NS
Within S2	4	0.552333	NS
Error	18	0.282556	
Total	29		

NS = Non Significant
S = Significant

Appendix –VII. Variation in ovary length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.362333	NS
Treatment	9	3.415741	S
Between sites	1	0.040333	NS
Within S1	4	2.649333	S
Within S2	4	5.026	S
Error	18	0.254185	
Total	29		

NS = Non Significant
S = Significant

Appendix –VIII. Variation in hypanthium length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.444333	NS
Treatment	9	0.413333	NS
Between sites	1	0.645333	NS
Within S1	4	0.087667	NS
Within S2	4	0.681	S
Error	18	0.169889	
Total	29		

NS = Non Significant
S = Significant

Appendix –IX. Variation in number of flowers per inflorescence

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	16.033	NS
Treatment	9	64.83559	S
Between sites	1	173.2803	S
Within S1	4	22.03067	NS
Within S2	4	80.52933	S
Error	18	11.50337	
Total	29		

NS = Non Significant
S = Significant

Appendix –X. Variation in filament length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.02412	NS
Treatment	9	0.310483	S
Between sites	1	0.167253	S
Within S1	4	0.3168	S
Within S2	4	0.339973	S
Error	18	0.026105	
Total	29		

NS = Non Significant
S = Significant

Appendix –XI. Variation in pedicel length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.00684	NS
Treatment	9	0.051622	NS
Between sites	1	0.147	NS
Within S1	4	0.036733	NS
Within S2	4	0.042667	NS
Error	18	0.092129	
Total	29		

NS = Non Significant

Appendix –XII. Variation in style length

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.032053	NS
Treatment	9	0.022437	NS
Between sites	1	0.002803	NS
Within S1	4	0.033307	NS
Within S2	4	0.016477	NS
Error	18	0.012372	
Total	29		

NS = Non Significant

Appendix –XIII. Variation in petal length (larger)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.000563	NS
Treatment	9	0.095058	S
Between sites	1	0.022413	S
Within S1	4	0.147383	S
Within S2	4	0.060893	S
Error	18	0.002686	
Total	29		

NS = Non Significant
S = Significant

Appendix –XIV. Variation in petal length (smaller)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.02677	S
Treatment	9	0.062295	S
Between sites	1	0.028213	S
Within S1	4	0.084093	S
Within S2	4	0.049017	S
Error	18	0.00174	
Total	29		

NS = Non Significant
S = Significant

Appendix –XV. Variation in pollen viability
Before anthesis

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	2.192333	NS
Treatment	9	4.046704	NS
Between sites	1	3.675	NS
Within S1	4	5.715667	NS
Within S2	4	2.470667	NS
Error	18	2.322704	
Total	29		

NS = Non Significant
S = Significant

Appendix –XVI . Variation in pollen viability at the time of anthesis

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	7.121053	NS
Treatment	9	21.38842	S
Between sites	1	8.17452	NS
Within S1	4	12.18767	NS
Within S2	4	33.89264	S
Error	18	6.17735	
Total	29		

NS = Non Significant
S = Significant

Appendix –XVII. Variation in pollen viability of fully opened flowers

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	6.246333	NS
Treatment	9	103.3002	S
Between sites	1	39.675	NS
Within S1	4	179.8717	S
Within S2	4	42.635	S
Error	18	9.313741	
Total	29		

Appendix –XVIII. Variation in seed weight
Seed weight

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	0.31948	NS
Treatment	9	420.5134	S
Between sites	1	1.986613	NS
Within S1	4	308.3365	S
Within S2	4	637.322	S
Error	18	11.28502	
Total	29		

NS = Non Significant
S = Significant

Appendix –XIX. Variation in anther dehiscence

Anther dehiscence S_1 (Jabli)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	4	88.88445	NS
Treatment	14	2183.723	S
Time	2	14441.76	S
Trees	4	199.9733	NS
Time*Trees	8	111.0889	NS
Error	56	438.021	
Total	74		

NS = Non Significant
S = Significant

Appendix –XX. Variation in anther dehiscence

Anther dehiscence S_2 (Jorhji)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	4	392.5141	NS
Treatment	14	1328.835	S
Time	2	8768.616	S
Trees	4	96.27704	NS
Time*Trees	8	85.16815	NS
Error	56	614.6919	
Total	74		

NS = Non Significant
S = Significant

Appendix –XXI. Variation in anthesis

Anthesis S_1 (Jabli)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	4	259.2074	NS
Treatment	14	2539.317	S
Time	2	16530.91	S
Trees	4	185.1815	NS
Time*Trees	8	218.4882	NS
Error	56	378.2669	
Total	74		

NS = Non Significant
S = Significant

Appendix –XXI I. Variation in anthesis

Anthesis S_2 (Jorhji)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	4	348.0852	NS
Treatment	14	2173.223	S
Time	2	14234.71	S
Trees	4	200.0222	NS
Time*Trees	8	144.4511	NS
Error	56	403.6415	
Total	74		

NS = Non Significant
S = Significant

Appendix –XXIII. Variation in pollen germination

Pollen germination S_1 (Jabli)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	4	0.084018	NS
Treatment	14	0.278504	S
Concentration	2	1.673472	S
Trees	4	0.044449	NS
Conc.*Trees	8	0.04679	NS
Error	56	0.044014	
Total	74		

NS = Non Significant
S = Significant

Appendix –XXIV. Variation in pollen germination

Pollen germination S_2 (Jorhji)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	4	0.066379	NS
Treatment	14	0.56347	S
Time	2	3.742628	S
Trees	4	0.060767	NS
Time*Trees	8	0.020031	NS
Error	56	0.039964	
Total	74		

Appendix –XXV. Variation in controlled crossing S_1 (Jabli)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	13233.33	S
Treatment	24	1133.33	S
S1	4	883.33	S
S2	4	383.33	NS
S1XS2	14	1580.95	S
Error	48	420.83	
Total	74		

NS = Non Significant
S = Significant

Appendix –XXVI. Variation in controlled crossing S_2 (Jorhji)

Source of variation	Degree of freedom	Mean sum of squares	Conclusion
Replication	2	11033.33	S
Treatment	24	1769.44	S
S1	4	1200.00	NS
S2	4	2450.00	S
S1XS2	14	1990.48	S
Error	48	790.28	
Total	74		

NS = Non Significant
S = Significant

BRIEF BIO-DATA

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Xth	2008	Govt. Taad Higher Secondary	JK-Board	63.00	First
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Year : 2015

Publication : NA

Research Paper in Peered Journal : NA

Others : NA

(Bibi Nagaar)