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**EVALUATION OF NEELAYAMARI (*Indigofera tinctoria* L.)
FOR YIELD AND GLYCOSIDE CONTENT UNDER
OPEN AND SHADED CONDITIONS**

SARADA.S.

**Thesis submitted in partial fulfilment of the requirement
for the degree of**

Doctor of Philosophy in Horticulture

**Faculty of Agriculture
Kerala Agricultural University, Thrissur**

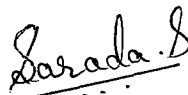
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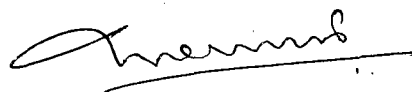
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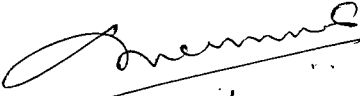
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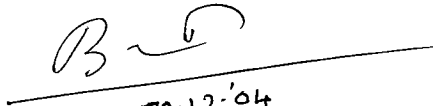
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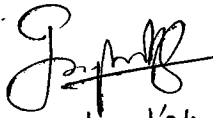

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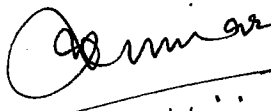
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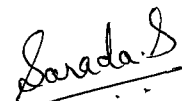
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LIST OF ABBREVIATIONS

%	–	per cent
@	–	at the rate of
°C	–	degree celsius
°E	–	degree East
°N	–	degree North
BCR	–	benefit cost ratio
CD	–	critical difference
CGR	–	crop growth rate
cm	–	centimeter
DAS	–	days after sowing
DMP	–	dry matter production
<i>et al.</i>	–	and others
Fig.	–	figure
FYM	–	farmyard manure
g	–	gram
GA	–	genetic advance
GCV	–	genotypic coefficient of variation
ha	–	hectare
HI	–	harvest index
i.e.	–	that is
K	–	potassium
kg	–	kilogram
LAD	–	leaf area duration
LAI	–	leaf area index
N	–	nitrogen
NAR	–	net assimilation rate
NS	–	not significant
P	–	phosphorus
PCV	–	phenotypic coefficient of variation
RBD	–	randomised block design
RGR	–	relative growth rate
Rs.	–	rupees
SE	–	standard error
<i>viz.</i>	–	namely
MSL	–	mean sea level

Introduction

1. INTRODUCTION

“The world is going herbal” is not just a phrase but a phenomenon, which is storming the globe with scientific rationale and leads that are fast emerging to support better health and life through plants and plant derived products. There is a distinct revival of global interest in botanical natural products such as colours, drugs, flavours and foodstuffs. Since ancient times, mankind depended mainly upon the plant kingdom to meet their needs. At one time, it was thought that the chemically synthesized materials would completely replace the materials of natural origin, but later it was realized that by no means a true copy of a natural substance could be made by synthetic means. Further more, the adverse side effects and high cost of production of synthetic materials also necessitated preferential use of natural materials for various purposes. India stands 10th among the richest countries for plant genetic resources in the world. By taking advantage of the rising green consumerism, India can acquire enviable position in the world. Hence the study on neelayamari (*Indigofera tinctoria*), a natural source of indigo dye, which is one of the oldest natural dyestuffs known to mankind.

Neelayamari (*Indigofera tinctoria* L.), commonly known as Indian indigo, is a medicinally as well as commercially useful leguminous plant. It was cultivated in India, China and other countries of the east as a source of indigo- a commercially valuable dye. The extract of leaves is reported to have remarkable effect on hair growth and in preventing juvenile greying of hair. In Ayurveda, neelayamari has been reported to be one of the major ingredients used in the treatment of hydrophobia, epilepsy, nervous disorders, bronchitis and to apply as an ointment for sores, old ulcers and haemorrhoids. Indigo plant is also used as a green manure. catch crop and transition crop. Owing to its permanent fast colour, indigo has long been used for dyeing and printing cotton, rayon and for dyeing wool. Various

pigments needed for paints, lacquers, rubber and printing ink were prepared from indigo.

In the forests of Kerala, there are about 550 non-wood forest produces yielding species. Among these, about 150 species are collected for the manufacturing of medicines, dyes, cosmetics *etc.* on a commercial basis (Sasidharan, 2000). In Kerala, indigo plant is cultivated by several pharmaceutical entrepreneurs both in public and private sector. The National Bank for Agriculture and Rural Development has identified neelayamari to be a possible crop for commercial cultivation in Kerala. In view of the medicinal and commercial importance of the species, *Indigofera tinctoria* was selected for the present study.

Lack of authentic varieties in medicinal plants is a major drawback, which hinders quality standardization of the pharmaceutical preparations made out of them. Screening of existing germplasm, evolving superior genotypes and releasing authentic varieties of medicinal plants would certainly help in maintaining the uniformity of raw materials used in the pharmaceutical industry which would ultimately lead to sustained superior quality of the products.

With these considerations, the following were set out as major objectives: -

- (1) Evaluation of the genetic stock of *Indigofera tinctoria* for yield of medicinally valuable plant parts (leaf and root) and pharmaceutically useful chemical constituents (glycosides)
- (2) Performance evaluation of selected accessions as pure crop in open and as intercrop in shaded situation in a coconut garden
- (3) Exploring the feasibility of cultivating *Indigofera tinctoria* commercially as a pure crop and as an intercrop in coconut garden

Review of Literature

2. REVIEW OF LITERATURE

Indigo, the blue dye, is one of the oldest dyestuffs known to the mankind. Mummy clothes, 5000 years old, have been found that were dyed with indigo. Indigo, has been obtained from a variety of plant sources such as *Indigofera tinctoria* found in Africa, Asia, East Indies and South America, *Polygonum tinctorium* found in Far East countries, China and Korea and *Isatis tinctoria* found in Europe.

Indigofera tinctoria L. commonly known as neelayamari in Malayalam, Indian indigo in English and neelini / neelika / renjini in Sanskrit is a medicinally as well as commercially useful leguminous plant. India appears to be the birthplace of this plant. *Indigofera* got its name from indigo (because it was believed to come from India) and from the Latin word "ferre" which means, "to bear". Hence *Indigofera* means "indigo bearing". The species name *tinctoria* refers to *tinctorius*, meaning "of dyes" or "belonging to dyes"(Marafioti, 1970; Simon *et al.*, 1984).

In India, the plant was found both in cultivated as well as in wild conditions in Bengal, Konkan, Karnataka and along the West Coast. It was cultivated on a large scale in many parts of North India for extracting indigo dye from its leaves, before the invention of synthetic dyes (Iyer and Kolammal, 1960).

In this chapter, literature on the history, origin, classification, growth habit, crop production and management, nodulation, economic importance and chemical constituents of *Indigofera tinctoria* is reviewed.

2.1 HISTORY OF INDIGO

India's expertise in preparing and using vegetable dyes dates back to thousands of years. Marco Polo, who passed through India in the 13th century, was the first to report on the preparations of indigo dye in India (Balasubramanian, 2002).

Evans (1990) reported that the use of indigo as a dye dates back to at least 4000 years, with evidence that the Egyptians used it to dye mummy clothes. The people of Central Africa and the Peruvians are thought to have used the dye since prehistoric times. It was also used extensively in India and the Far East.

Zohary (1998) established that *Indigofera tinctoria* grown in India and Pakistan since the end of the 3rd millennium B.C. was introduced into the Mediterranean basin only in early Islamic times (8th-11th centuries A.D.).

Leix (1938) recorded that during the middle ages, indigo moved like other valuable articles of trade through the established routes. Baghdad was a trade centre along the route from India to Europe, and the indigo that moved from Baghdad to the west was originally from plants grown in India.

Randhawa (1982) recorded that the indigo plant was grown in Oudh, Allahabad, Agra, Lahore, Multan, Malwa, Delhi and Ajmer in the sixteenth century. The best indigo, however, was grown at Bayana, in the Bharatpur district of Rajasthan, and the second best was that at Sarkhej in Gujarat. The dye extracted from the leaves was used mainly for dyeing clothes and for making paints.

Although the Portugese had established trade links in indigo with India in the 16th century, the dye was effectively kept out of much of Europe until the end of the century by the activities of the woad (*Isatis tinctoria*) growers and distributors. However, by the middle of the 17th century, a considerable trade in indigo had been built up between Europe and India (Evans, 1990).

Kochhar (1981) opined that in the early years of the British occupation of India, indigo was a very important item of trade of the East India Company. By the middle of the seventeenth century indigo became the most popular dye in Europe, replacing woad, which produced a dye, which was more difficult to prepare and inferior to indigo.

But towards the end of the 18th century, cotton and sugar became more profitable crops for the colonists and the centre of indigo cultivation shifted back to India (David and Rembert, 1979).

Randhawa (1983) reported that by 1830 there were 300 or 400 indigo factories in Bengal, chiefly in Jessor, Krishnagar and Tirhoot and in the Champaran district of Bihar.

Indigo dye was first produced artificially by F. W. A. von Baeyer in 1880 (Leake, 1975). By 1897 it was manufactured and marketed in large scale by the Badische Aniline Company of Germany. At that time, India was growing 574,000 ha of indigo which, by 1911, had fallen to 86,600 ha (Leake, 1975). During world wars I and II, due to the shortage of the synthetic dye stuff, indigo cultivation received a brief stimulus.

2.2 ORIGIN AND CLASSIFICATION OF *INDIGOFERA*

The family Fabaceae (Leguminosae) is one of the largest families of angiosperms. It comprises of about 600 genera and 12,000 to 18,000 species distributed all over the world. In India, they occupy the third position in the list of ten dominant families (Bairiganjan *et. al*, 1985). Senn (1938) reported that the basic chromosome number of the genus *Indigofera* is $n = 8$.

Hooker (1876) recorded 40 species and 3 varieties of *Indigofera* in India. Revisionary study of the genus revealed the occurrence of 60 species and 8 varieties. Of these, 13 species and 2 varieties are found to be endemic.

According to Arora and Chandel (1972) the centre of diversity of *Indigofera* is mainly concentrated in Eastern and Western Himalayas and Eastern and Western Ghats. Out of 44 species distributed in India, 10 species occur throughout plains, ascending Himalayas and hills of India.

According to Benson (1957) the systematic position of *Indigofera tinctoria* is as follows:- Division : Spermatophyta, Class : Angiospermae, Subclass: Dicotyledonae, Group: Calyciflorae, Order: Rosales. Family:

Leguminosae / Fabaceae, Sub family : Papilionoideae, Genera: *Indigofera*,
Species: *tinctoria*.

Of the various species of *Indigofera*, the chief ones exploited for the dye in India are *I. arrecta*, *I. suffruticosa*, *I. sumatrana* and *I. tinctoria*. Of these, *I. arrecta* and *I. tinctoria* were the most important ones (C S I R, 1959).

The cultivated species of *Indigofera*, their occurrence, habit and floral characteristics are presented in Table 1.

Leake (1975) reported that there are about 350 species of *Indigofera* scattered throughout the tropical and warmer, sub-tropical countries of all continents. There are more than 40 species of *Indigofera* indigenous to India, largely localised. Originally in each locality, the dye was obtained from the species indigenous to that locality. With the arrival of the Europeans and consequent development of export trade, transference of the indigenous species from one locality to another, and importation of alien species had taken place.

According to Allen and Allen (1981), the genus *Indigofera* consists of 800 species and many of the species are annual or perennial herbs but some are shrubs, whilst others grow to small trees. The genus is widespread in tropical and subtropical areas; particularly the Sino- Indian region. They are also found in the tropics of both America and Africa. Indeed, the genus *Indigofera* is one of the largest and most widely distributed one in South Africa. Tropical Africa, with largest number of species and with greatest development of the genus, is considered to be the centre of distribution (Sanjappa, 1984).

According to Simon *et al.* (1984), indigo refers to several species of *Indigofera*. Of primary importance are French indigo/ Indian indigo (*Indigofera tinctoria* L.) and Guatemalan indigo (*Indigofera suffruticosa* Mill, which was formerly classified as *I. anil* L.). The French and Guatemalan indigo differ in size and shape of the leaflets and pods. According to Evans (1990), over the years some six species of *Indigofera* have been cultivated

Table 1. Cultivated species of genus *Indigofera* (C S I R, 1959)

Species	Common name	Area of Cultivation	Leaf characters	Flower characters	Pod characters	Uses
<i>Indigofera arrecta</i> Hochst.	Natal indigo/Java indigo/Bengal indigo	Bihar, U. P.	10-13 cm long, leaflets 7-8 pairs with one odd terminal leaflet	Pinkish red, in axillary racemes	Straight and reflexed	as a cover crop in coffee, tea and rubber plantations. as a green manure crop for rice. root used as abortifacient (Nyazema, 1987)
<i>I. suffruticosa</i> Mill.	West Indian indigo/Anil indigo	Indonesia, Malaya, Ceylon, Africa	5-8 cm long, leaflets 5-15, oblong, glabrous above and pubescent below	Pale orange in colour, in short racemes of 15-20	Curved, 0.3-0.6 in. long, 2-4 seeded, thickened at sutures	as a cover and green manure crop in coffee and tea plantations. the plant is febrifugal, vulnerary, purgative, antispasmodic, diuretic and stomachic
<i>I. sumatrana</i> Gaerth.	-	Andhra Pradesh, Madras	leaflets 9-15, obovate to nearly elliptic	Racemes 3-6 in. long	Obtusely curved, 2-4 cm long	as a green manure crop, preceeding cotton, maize or sugarcane.
<i>I. tinctoria</i> Linn.	Common indigo/Indian indigo	India, China and other countries of the east	1-3 in. long, 9-13 leaflets	Pink, in nearly sessile spicate raceme, 2-4 in. long	Glabrescent, slightly curved or straight, 3/4-1 in. long	as a cover or green manure crop in coffee plantations and rice fields plant extract used in epilepsy and nervous disorders, bronchitis as an ointment for sores, old ulcers and haemorrhoids.

as major sources of the dye. *Indigofera suffruticosa* from tropical America was widely grown in both the New and Old World, whilst *I. tinctoria* was the main source of indigo in the Far East. *I. sumatrana* was introduced by the Dutch to India from Malaysia because it produced a higher yield of indigo (this species is now considered to be synonymous with *I. tinctoria*). *I. sumatrana* was also taken from the Old world to the West Indies to establish the crop there. In addition, *I. articulata* and *I. arrecta* from tropical Africa and *I. longeracemosa* from Madagascar have also been introduced to that country for cultivation.

According to Rajwar (1983), in India, *Indigofera tinctoria* was seen in Kerala, Tamilnad and Himalayan regions. Vijayakumar and Ramayya (1986) have reported 28 species of *Indigofera* in South India. Some species of the genus are rather scarce. Several species of *Indigofera* became enlisted as rare or endangered. *Indigofera tinctoria* is now a threatened species in India (Hajra *et al.*, 1995).

Ramamurthy and Pullaiah (1998) recorded that *Indigofera* is represented by 25 species in Eastern Ghats. This is the second largest genus after the *Crotalaria*, which is represented by 55 species in Eastern Ghats.

Subudhi and Mitra (1999) observed that of the seven agro-climatic regions in India, Deccan is having highest number of *Indigofera* species, *ie.* 26, followed by 22 and 18 in Malabar (northern Kerala) and Gangetic plain respectively. Eastern Himalayas is having lowest number of species, *ie.* 6. The tropical species are mostly confined to peninsular India, which is a large tract with great variation in climate and physiography. Most of the *Indigofera* occur chiefly as undergrowth in deciduous/evergreen forests. Some of these are confined to grasslands.

2.3 GROWTH HABIT OF *I. TINCTORIA*

Ramamurthy and Pullaiah (1998) observed that the general features of the genus *Indigofera* are prostrate, suberect or twining herbs, leaves simple, trifoliolate and imparipinnate. Flowers axillary raceme,

panicles or heads. Keels with downward spur on each side. Stamens diadelphous (9 +1). pod usually oblong, linear, erect or deflexed, straight, curved, torulose, usually dehiscent.

I. tinctoria Linn. is an erect, suffruticose, pubescent shrub 1.2 to 1.8 m high, with firm woody terete branches, light greenish brown to somewhat silvery grey in colour bearing alternate pinnate compound leaves 2.5-7.5 cm long, petioles 1.2-2.5 cm long, stipules small and subulate. Leaflets 7-13, opposite, membranous, bluish green, oblong or oblanceolate, rounded, glabrous above and thinly clothed with oppressed hairs beneath. Leaves turn greyish black on drying. Flowers numerous, in nearly sessile lax spicate racemes 5 -10 cm long. Calyx teeth triangular, acute. The corolla is vexillary pink, consisting of a rounded emarginate standard petal, brownish and pubescent at the back and two wing petals adherent to the two keel petals which are greenish in colour and furnished with a spur on each side often bending back elastically. The stamens are diadelphous, nine and one, anthers uniform. Ovary is sessile, eight to ten or more ovuled with a short incurved style ending in a capitate stigma. Pods 2-3.2 cm long, linear, straight or slightly curved, thickened at the sutures, glabrous, pale greenish grey when young and dark brown on ripening with 8-12 seeds (C S I R, 1959; Iyer and Kolammal, 1960; Kirtikar and Basu, 1935; Kochhar, 1981; Singh and Khan, 1990; Anita, 1998).

Jawahar (1996) while listing *Indigofera tinctoria* L. under the medicinal plants of the Kani tribes reported that the plant is usually grown by the tribes and scantily seen in the wild state near the tribal settlements.

2.4 CROP PRODUCTION AND MANAGEMENT

Sasidharan (2000) observed that most of the raw drugs used in the preparation of ayurvedic medicines are collected from the wild, particularly from forests. He listed about 150 species as non-wood forest produces, collected for manufacturing of medicines, dyes, cosmetics etc. on a commercial basis, which included *I. tinctoria*.

Kurian *et al.* (2000) identified twelve species of medicinal plants for commercial cultivation in Kerala, which included *I. tinctoria*.

Nair (2000) also included *I. tinctoria* in the list of medicinal plant species to be domesticated for homestead farming in Kerala.

2.4.1 Soil and Climate

As a cultivated crop, *I. tinctoria* prefers a mean precipitation of 146 mm, annual mean temperature of 22.5⁰ C and a soil having mean pH of 6.5 (Dukes, 1982).

Simon *et al.* (1984) opined that *I. tinctoria* tolerates 16 to 27⁰C with an annual precipitation of 700 to 4200 mm and a soil pH of 5 to 7.3. It can withstand temperature up to 40⁰ C provided adequate soil moisture could be maintained (Thomas *et al.*, 2000).

Bairiganjan *et al.* (1985) reported that *I. tinctoria* is a bushy shrub, commonly cultivated in rainy season.

Randhawa (1983) observed that the best soils for growing indigo were those subject to inundation from the Ganges. Rasulova *et al.* (1986) recommended *I. tinctoria* is to be grown in the valleys / lowlands.

Kerala Agricultural University (K A U, 1993) established that the best time for sowing *I. tinctoria* is in the NE monsoon, *i.e.*, September-October.

According to Thomas *et al.* (2000), Indian indigo requires good sunlight and grows well both in plains and hilly areas. Sandy loam soil is the best, while clayey soil where water logging is likely, is unsuitable. However, as a green manure crop it can be grown in clayey soil for incorporation into the soil (K A U, 1993). It can also be cultivated in coastal sandy soils.

2.4.2 Propagation

I. tinctoria is usually propagated by seeds. Seeds are very small and the seed rate is 3 kg ha⁻¹.

Leake (1975) reported that *Indigofera* species needed seed treatment since it had a seed with a very hard coat and a low germination percentage.

Abdi and Barker (1987), in a trial to enhance germination percentage of seeds of *Acacia horrida*, *A. nubica* and *Indigofera tinctoria* found that soaking in hot water significantly improved both per cent germination and mean germination time in all species. In *I. tinctoria*, rubbing with sandpaper also increased germination percentage significantly.

Ahmed *et al.* (1990) prescribed acid scarification for breaking seed dormancy in *I. tinctoria* and *I. linnaei*. Kumari and Francis (1998) used concentrated sulphuric acid for 4 minutes and reported effective in breaking dormancy and enhancing germinability in *I. tinctoria* seeds. Gupta (2003) reported that concentrated sulphuric acid scarification in *I. tinctoria* resulted in 80-90% germination. In scarification using sand, seeds are mixed with sand and ground gently to break the seedcoat. After pretreatment, seeds are broadcasted after mixing with sand 2 or 3 times its volume to ensure uniform coverage. The seedbeds should be covered with straw and irrigated. Seeds germinate within 15 days and the seedlings are ready for transplanting after one month (Thomas *et al.*, 2000; Sy *et al.*, 2001; KAU, 1993).

Vegetative propagation of *I. tinctoria* was attempted by Philip *et al.* (1991) along with some other medicinal plants grown in Kerala. Terminal shoot tips of 4 to 6 cm length with leaves, semi-hardwood cuttings of 5, 10, 15 and 20 cm length without leaves and hardwood cuttings of 5, 10, 15 and 20 cm length without leaves were tried in *I. tinctoria*. The stem cuttings failed to root even with the applications of rooting hormones *viz.* IAA, IBA and NAA at 10, 20 and 30 ppm respectively.

2.4.3 Cultural Practices

In Bihar, cultivators of *Indigofera* start preparation of the seedbed shortly after the cessation of rains in September-October, when the soil moisture is sufficient to ensure germination (Leake, 1975). However,

Randhawa (1982) suggested to sow *Indigofera* in June so that it can be cut at the end of September or early in October, when it is fully ripe. The plants are cut at a handbreadth from the ground, and next year the ratoon crop grows from the stumps. Thomas *et al.* (2000) found that the best time for sowing *I. tinctoria* in Kerala, is September-October.

Moody *et al.* (1986) reported that if indigo is broadcasted, little weeding is required. However, if the crop is drilled, light shallow cultivation with a native plough or tractor-mounted cultivator should commence 2 to 3 weeks after establishment. Two or three inter row cultivation may be needed to achieve good weed control. Cattle manure should be applied at the rate of 10 t/ha as basal dressing and incorporated into soil along with last ploughing. Weeding has to be done two times, three weeks after sowing and six weeks after sowing. Plants will start flowering 2-3 months after sowing.

2.4.4. Diseases

Varma (1991) reported incidence of various fungal diseases caused by *Alternaria*, *Cercospora*, *Colletotrichum* etc. in *I. tinctoria* in different parts of Kerala.

Report on other diseases of *I. tinctoria* is rather scarce.

2.4.5 Pests

Adams (1967) reported *Aphis craccivora* and *A. gossypii* in *Indigofera sp.* Aphid species collected over a three year period from host plants in Malawi included *Aphis craccivora* from *Indigofera arrecta*, *I. emarginella*, *I. hirsuta* and *I. spicata*.

Govindan (1975) found the weevil *Alcidodes (Alcides) bubo* (F.) infesting Sumatran indigo (*I. sumatrana*) at Bangalore. The eggs were laid on the growing shoots, and the larvae bored into the stem, causing oval galls.

Rao and Thirumalachar (1977) recorded *I. hirsuta* for the first time as host for the groundnut leaf miner.

Haseeb *et al.* (1985) reported *I. tinctoria* as a new host of the root-knot nematode *Meloidogyne javanica*.

In Kerala, extensive wilting and drying of *I. tinctoria* has been reported due to infestation by the psyllid *Arytaina punctipennis*. This is a new record of the pest from southern India. The pest infests the top shoot of *I. tinctoria* causing curling up and drooping of leaves and shoots, leading to wilting of plants (KAU, 1993). Spraying with quinalphos (Ekalux 20 AF) at 2 ml l⁻¹ gave good initial control of the pest, but recurrence was observed after three weeks. No pest attack was observed following treatment with monocrotophos (Nuvacron 36 EC) at 1.5 ml l⁻¹ (Skaria *et al.*, 1996).

Johnson and Anton (1999) recorded that the female holotype of *Bruchus indigoferae* was presumed to have been collected in India from the seeds of *I. tinctoria*. Kagawa *et al.* (2000) reported presence of *Aphis indigoferae* based on specimens from *I. pseudotinctoria*.

2.4.6 Harvesting and Yield

Nair *et al.* (1991) reported that *I. tinctoria* can yield upto 210 kg leaves from 400 m² of land from six harvests.

According to Nair (1992), if cultivated in a scientific way the harvesting of matured leaves of *I. tinctoria* could be done once in two months. By severe pruning at the end of every year the economic life of the plant could be prolonged for a few years.

Thomas *et al.* (2000) suggested harvesting of *I. tinctoria* to be done by cutting the plants at flowering time, at a height of about 10 cm from ground level. Subsequent harvests can be made at 1.5-2 months interval. Four to five cuttings can be taken in a year depending on the growth. A few plants per plot are left without cutting, for seed production. Ripe pods are to be harvested in the early morning to prevent loss of seeds by shattering during harvest.

Green matter production of *I. tinctoria* is 8-10 t ha⁻¹ (KAU, 1993). Better yield is obtained from *I. tinctoria* when two irrigations are given during the whole crop period.

2.4.7 Soilless Culture

Khazhakyan *et al.* (1986) while studying effect of day length on growth and productivity of *I. tinctoria* and *I. articulata* in soilless culture, observed that both species grew better and yielded more in long days (18 h) than in short days (8 h). *I. tinctoria* gave five times higher leaf yields than in short days.

I. tinctoria and *I. articulata* were also successfully grown on gravel or gravel : volcanic slag (70 : 30%). Volcanic slag alone was a less suitable medium. The species were grown as annual crops and the first harvest had maximum indigo content. *I. tinctoria* had a 21 per cent higher content of the dye than *I. articulata*. Leaf productivity and dye output was three or more times greater with soil less cultivation than with soil. The best substrate was a mixture of gravel and volcanic slag (3:1). For indigo, the optimum N P K contents of the nutrient solutions were 18.9: 3.1: 11.9 mmol l⁻¹ (Mairapetyan *et al.*, 1986; 1988).

Khazhakyan and Egibyan (1987) reported that when the above species were sprayed with GA₃ at various concentrations (100-200 mg l⁻¹) in natural day length, GA₃ at high rates inhibited flowering, whereas at low rates in long (18 h) days it did not affect plant development.

2.5 NODULATION AND NITROGEN FIXATION

Like other members of the Leguminosae, the species *Indigofera* have the capacity, in association with *Rhizobium*, to fix nitrogen. *I. arrecta* produces a deep root system with a plentiful number of nodules (Allen and Allen, 1981). The amount of nitrogen fixed by *I. arrecta* ranged from 208.64 to 342.92 mg N nodule⁻¹ hour⁻¹ (Prana *et al.*, 1984).

Sen (1993) observed that legumes, besides their wide variety of uses, also contribute considerable amount of nitrogen through their root nodules and indirectly help in maintaining the soil fertility. The symbiotic association between *Rhizobium* strain and a legume is connected with biological nitrogen fixation. In *I. tinctoria* nodules were found to be abundant, diffused and globose in structure.

Sunitha (1996) reported that the ability for nodulation in herbaceous leguminous medicinal plants varied with plant species. This may be due to factors like inherent ability of genotype, genotype-strain interaction and the lack of sufficient population of desirable, infective native strain of rhizobia in the soil. In *I. tinctoria*, the number of root nodules and their fresh weight were significantly higher under open condition than under shade.

A major benefit of legume green manures is the contribution of N to the soil *via* nitrogen fixation. Thonnisen *et al.* (2000a) while assessing the feasibility of meeting N needs of tomato with legume green manure found that *I. tinctoria* could derive 71.8 per cent of the N accumulated in its plant biomass from biological nitrogen fixation.

2.6 ECONOMIC IMPORTANCE OF *INDIGOFERA SP.*

2.6.1 As a Dye

Francis (1984) reported that most of the *Indigofera* species (about 40 species) yield dyes, and are useful for hair and cloth dyeing.

Kochhar (1981) reported the method of extracting dye from *Indigofera* species. Freshly cut plants, after crushing, were steeped in water for 10-15 hours in specially designed indigo-vats, during which the glycoside was converted into glucose and indoxyl by the activity of indimulsin, naturally present in the plant. The yellow supernatant liquid was passed through beating or oxidising vats equipped with paddle wheels, which facilitated aeration of the solution. The liquid was continuously agitated to bring about oxidation and the operation was stopped, by heating

as soon as the blue colour developed. The blue dye formed a fine bluish mud at the bottom of the vat. The spent liquor was then drawn off. The bluish mass was treated with boiling water (sometimes lime water) to aid further granulation. The filtered sludge, after pressing, was cut into small cubes of roughly 7 cm size, which after air drying was graded into various types.

Nayar *et al.* (1999) reported a simplified method in which fresh leaves of *I. tinctoria* are expressed with water to make juice. The juice is sieved through a piece of cotton and kept for a long time for the sediment to form. Supernatant liquid is decanted and sediment is dried to a powder that serves as the blue colour.

2.6.2 As Medicine

Kirtikar and Basu (1935) reported that the plant extract of *Indigofera tinctoria* is given in epilepsy and nervous disorders. It is also used in bronchitis and as an ointment in sores, old ulcers and haemorrhoids.

According to *Bhavaprakasa*, nili (*I. tinctoria*) is purgative in action, bitter, hot, improves hair and is useful in urinary diseases, giddiness, abdominal enlargement, enlargement of spleen and alcoholic intoxication. In Ayurvedic practice, nili is mainly used as an antitoxic and promoter of hair growth (Iyer and Kolammal, 1960).

Anand *et al.* (1979) noticed that the alcoholic extract of the aerial parts of *Indigofera tinctoria* exhibited marked antihepatotoxic effect against carbon tetrachloride induced liver damage in rabbits, rats and mice. The extract also increased bile flow and liver weight in rats suggesting stimulation of microsomal enzymes of liver.

Xiujuan *et al.* (1981) found that at therapeutic dosage synthetic indirubin, the component responsible for the anticancer activity of *I. tinctoria*, yielded marked inhibition of Lewis lung carcinoma and Walker carcinosarcoma 256 in mice.

Flaumenhaft (1982) recorded that southeast Asians and Chinese value *I. tinctoria* for treating diseases of the liver and dysentery.

Murthy (1984) observed that in Charaka Samhita, Susrutha Samhita and Ashtangahridaya the use of *I. tinctoria* in the preparations of medicine for greying of hair and constipation has been mentioned. Neeli is one of the main ingredients of Neelidaladi thailam and Neelibhringadi Kesathailam, which are popular hair oils.

Simon *et al.* (1984) reported that the Chinese use *I. tinctoria* to clean the liver, detoxify blood, reduce inflammation, alleviate pain and reduce fever.

According to Chang *et al.* (1988) indirubin and natural indigo are effective for the treatment of chronic granulocytic leukemia.

Various pharmacological studies carried out on the leaf and stem extracts of *I. tinctoria* on central nervous system revealed that they had significant depressant action and a beneficial anticonvulsant action (Rahamathullah *et al.*, 1990).

Liu and He (1991) observed that in China a complex indigo powder (mixture of leaves of *I. tinctoria* or *I. suffruticosa*) is used to treat scrofula -tuberculosis of the cervical lymph nodes.

Nair *et al.* (1991) reported that Neelibhringadi oil is used not only for inducing abundant growth of hair, but also for maintaining its natural black colour. Ahmad *et al.* (1993) reported on the contraceptive potential of five Unani drugs which included *I. tinctoria*. Anticancer drugs have been reported from *I. tinctoria* (Han, 1995).

Masilamani (1995) reported that *Indigofera tinctoria*, is one of the most relied upon antidotes to snakebite among Gounda tribals of Tirupathur.

Thomas *et al.* (2000) recorded that Nili is purgative in action, bitter, hot, cures giddiness, abdominal enlargement, gout and intestinal obstruction.

Kavimani *et al.* (2001) found that the methanolic extract of *I. tinctoria* exhibited antitumour activity in Dalton's ascitic lymphoma (DAL) bearing mice. Red blood cell count, haemoglobin content and white blood cell count were more or less normal after extract treatment of the tumour bearing mice.

Singh *et al.* (2001b) reported that indigotin, a bioactive fraction obtained by fractionation of a petroleum ether extract of the aerial parts of *I. tinctoria* showed significant dose-related preventive and restorative effect of hepatoprotective activity against carbontetrachloride induced liver injury in rats and mice.

Sreepriya *et al.* (2001) and Malarvannan and Devaki (2003) investigated the effects of pre-treatment with the alcoholic extract of *I. tinctoria* on liver antioxidant defense system during acute hepatitis on the activities of enzymic antioxidants in the liver of rats. Results indicate that pretreatment with *I. tinctoria* extract in rats is very effective in reducing oxidative stress suggesting an antioxidant effect.

The therapeutic uses of *I. tinctoria* L. are illustrated in Table 2.

2.6.3 As Green manure, Catch crop and Transition crop

I. arrecta has been tried as a cover crop in coffee, tea and rubber plantations. It is also utilized as a green manure crop for rice, specially recommended in rotations before cotton. *I. articulata*, *I. enneaphylla*, *I. hirsuta*, *I. suffruticosa* and *I. sumatrana* has also been recommended as green manure (CSIR, 1959).

Leake (1975) observed that when the residues of the indigo fermentation vats- 'seet' is spread onto the land and worked into the soil, the health and vigor of the subsequent crop was amazingly high.

Bhandari *et al.* (1977) observed that *I. cordifolia* improves soil fertility and can be used as a fertilizer for bajra.

Green- manuring improves the yield and partially substitutes the nitrogen requirement of a crop (Singh, 1984). Sharma *et al.* (1985) observed

Table 2. Therapeutic uses of *Indigofera tinctoria* L.

Part	Therapeutic uses	References
Whole Plant	Laxative, expectorant, alexipharmic, anthelmintic, promote the growth of hair, used in abdominal complaints, heart diseases, insanity, cure "vata", tumours, fever, leucoderma, enlarged spleen, cephalagia, injuries.	Kirtikar and Basu (1935)
	Cures chronic bronchitis and asthma, piles, leucoderma bites of insects and reptiles, burns, scalds, ulcers and skin diseases, acts as a diuretic and cathartic	Nadkarni (1976)
	Used in hepatitis, epilepsy and nervous disorders and ointments and in hemorrhoids.	Singh and Khan (1990); Saraswathy <i>et al.</i> (1998)
	Methanolic extract exhibited inhibitory activity on HIV-1 and HIV-2	Kavimani <i>et al.</i> (2000)
Leaf	A cure for hydrophobia, urinary complaints, against inflammation and itching.	Kirtikar and Basu (1935)
	Prophylactic against hydrophobia	Rao and Rao (1987)
	For treating leukaemia	Han (1988)
	For the treatment of liver disorders	Singh <i>et al.</i> (1998)
	Against snakebite, jaundice and spleen swelling	Thomas and Britto (1999)
	Forms major ingredient of 'Nilibhringadi' a reputed drug for promotion of hair growth	KAU (1993)
	Possess antitoxic property, antifebrile and antidote	Stuart (1911)
For treating skin diseases such as wound, injury, abscess, ringworm and eczema	Jawahar (1996)	
Root	Useful in difficult micturition, snakebite, caries of the teeth, consumption and used in hepatitis	Kirtikar and Basu (1935)
	Powdered, made into a paste with water and applied to worm infested wounds.	Jain and Dam (1979)
	Root paste-antidote for spider poisoning	Sudhadevi (1992)
	To treat enlargement of scrotum, relieves gastralgia	Unnikrishnan (2001)
Indigo dye	Given in liver complaints and nervous diseases, a soothing balm for burns and scalds, dust applied to ulcers and boils.	Dastur (1977)

that legumes benefit the succeeding crop through symbiotic nitrogen fixation and mobilization of lesser available forms of plant nutrients, improvement of soil structure and decrease in leaching loss of nutrients. According to Meelu and Morris (1986) *I. tinctoria* L. and *Sesbania* species are the most acceptable green manure crops to Indian farmers.

Morton (1989) recorded that creeping indigo (*I. spicata*), a minor source of indigo dye, has been much planted for cover, green manure and erosion control in coffee, tea and rubber plantations in Florida.

Dennis *et al.* (1990) observed that among the species that appear to have significant potential as green manure in the Philippines rice-production system, *I. tinctoria* is the most common.

Indigofera species are used as a cover crop to protect the soil from erosion or improve the quality of the soil. *I. arrecta* gives a dense shade, which smothers weeds. It has been suggested as an intercrop to provide shade and protection for young cocoa trees and for oil palms. *Indigofera* species have also been used as a green manure to enrich impoverished tropical soils (Evans, 1990).

In field studies in Brazil when *I. tinctoria* cover crop residues was incorporated into soil the average fertilizer- N substitution value was 26 kg (Smyth *et al.*, 1991).

Meelu *et al.* (1992) assessed *I. tinctoria* as green manure for wet season rice in the Philippines and was found to increase the rice grain yield. According to Sen (1993), *I. tinctoria* is used as cover or green manure crop in coffee plantation and rice fields in India.

In a study conducted by Michel (1996), 60 day-old- *Indigofera* plants conformed to high quality litter characteristics releasing N quickly to the soil. *Indigofera* green manure contained between 5 and 40 kg N ha⁻¹.

In Bangladesh, Uddin *et al.* (1996) found that *I. tinctoria* contributed 47 kg N ha⁻¹ to the sugarcane field, when incorporated as green manure into soil. Organic C, total N, available P and S and exchangeable K status of the soil were also slightly increased. Similar observations were

also made by Alam *et al.* (1997). He reported a total biomass production of 2.62 tonnes ha⁻¹. However in southern Taiwan, incorporating legume biomass into the soil resulted in immobilization of available soil N. A positive influence of the previous year's biomass of legume clippings on vegetable yields was observed in the long term. Crop N status was better in live- mulch plots (Kleinhenz *et al.*, 1997).

Anita (1998) reported that the residual parts of *I. tinctoria* plants, left after the extraction of the dye, are rich in available nitrogen and are used as manure for cereals, oilseeds, sugarcane and tobacco.

Shrestha and Ladha (1998) reported that on an average indigo (*I. tinctoria*) captured 194 kg N ha⁻¹. A suggestion for developing indigo plus maize N-catch crop rotations is made to decrease NO₃ leaching and maximize N use efficiency in the rice-sweet pepper cropping system.

In a long-term field experiment conducted in Philippines to determine the effects of indigo (*I. tinctoria*) on the productivity of rainfed lowland rice-based cropping systems it was found that indigo green manure has a positive effect on rice yields (Agustin *et al.*, 1999a). According to Agustin *et al.* (1999b) indigo is used to replace and supplement chemical fertilizers and to improve soil fertility.

Kondo *et al.* (2000) found that below 20 and down to 70 cm depth, indigo (*I. tinctoria*) showed an almost uniform root length density, reflecting the high capacity for trapping leaching NO₃ from the deep soil layer as N catch crops.

Shrestha and Ladha (2000) reported that significant amounts of NH₄-N accumulated in soil at 15 days after incorporation of residues of indigo (*I. tinctoria*) alone (12 kg ha⁻¹). It maintained low NO₃ throughout the soil profile.

Thonnissen *et al.* (2000b) reported that *I. tinctoria* yielded 0.9 Mg biomass ha⁻¹ and 5 to 40 kg N ha⁻¹. Faster decomposition of *Indigofera* in the dry season was caused by its smaller and tender leaves, lower C/N- ratio (10.6) and higher initial N content (4.2). The relatively high polyphenol

(3.7%) and tannin (1.6%) content of *Indigofera* may have retarded decomposition in the wet season.

Shrestha *et al.* (2002) studied the effects of transition crop indigo (*I. tinctoria*) grown during the dry-to-wet transition on carbon dynamics and its residue management. The decrease in labile C with cropping ranged from 6 to 21% and increase with residue incorporation ranged from 18 to 37% at different sites, but total C remained unchanged.

KAU (1993) recorded that the green matter production of *I. tinctoria* as green manure is 8-10 t ha⁻¹.

2.6.4 As a Means to Control Nematodes

Dominguez *et al.* (1986) studied reaction of 10 accessions of *I. hirsuta* to *Meloidogyne arenaria*, *M. incognita* and *M. javanica*. Significant variation in resistance was detected for all nematodes tested. FL 24, of local origin, was significantly more resistant than the other accessions.

Reddy *et al.* (1986) also reported lower population of *Meloidogyne incognita*, *Belanolaimus longicaudatus* and *Pratylenchus brachyurus* when *I. hirsuta* was rotated with the succeeding crops.

Baltensperger (1989) established that *I. hirsuta* germplasm, FL 24 (early-maturing) and FL 101 (late maturing) both are fairly resistant to root-knot nematodes, including *Meloidogyne arenaria*, *M. incognita* and *M. javanica*. Baltensperger *et al.* (1990) reported that *I. hirsuta* cultivar 'Flamingo' was also resistant to *Meloidogyne sp.* of nematode.

Sreeja and Charles (1998) reported that root extracts of *I. tinctoria* exhibited a high degree of nematostatic action against nematode adults and larvae.

The tissue amendments of several non-traditional legumes including *I. tinctoria*, *I. hirsuta*, *I. spicata* and *I. suffruticosa* are known to contain bioactive phytochemicals and may offer considerable promise as

soil amendments for control of plant-parasitic nematodes (Morris and Walker, 2002).

2.6.5 As an Antifeedant

CSIRO (1972) reported that no *I. spicata* plants, treated with a chemical mutagen, produced plants completely free of indospicine but a number of low-indospicine plants were detected.

Hegarty and Peterson (1973) observed that there are reservations about the use of *Indigofera* species as forage because it is known that there are deleterious substances present in a number of species. *I. spicata* in the diet causes the death of chicks and is hepatotoxic to cattle, rabbits and mice. Indospicine, a free non-protein amino acid analogue of arginine has been detected in the leaves and seeds of several *Indigofera* species including *I. spicata*. Smolenski *et al.* (1981) also recorded *I. spicata* to be toxic.

Williams (1981) observed that *I. echinata* and *I. pseudotinctoria* were moderately toxic when fed to young chicks. Concentrations of nitro compounds in these species were amongst the highest observed. *I. hirsuta* which has been introduced for commercial use in USA did not contain nitro compounds and is not toxic.

Krishna *et al.* (1986) concluded that leaves of *I. teysmanni* are not suitable as a sole feed for sheep.

I. spicata was demonstrated to be toxic to rabbits by Morton (1989).

2.6.6 Miscellaneous Uses

Kirtikar and Basu (1935) reported that in China the blue bag of indigo is used as a domestic remedy for the stings of bees and wasps.

Bean (1970) found that about a dozen species of *Indigofera*, including *I. amblyantha*, *I. potaninii* and *I. delisiana* are valued in Britain and Northern Europe as ornamental shrubs.

Jain and Borthakur (1980) observed that the leaves of *I. dosua* is used for tattooing and colouring garments.

Kochhar (1981) opined that owing to its permanent fast colour, indigo has long been used for dyeing and printing cotton, rayon and for dyeing wool. Various pigments needed for paints, lacquers, rubber and printing ink were prepared from indigo.

Secoy and Smith (1983) reported that the roots of *Indigofera* sp. are used against insects, especially lice.

The fine branches of *I schimperi* was used in making brooms. (Medley, 1993)

Anita (1998) recorded that indigo is used to a small extent by artists and in wallpaper decoration. It possesses excellent fastness to light and dyeings on wool are faster than those on cotton.

2.7 INTERCROPPING OF MEDICINAL PLANTS IN COCONUT SHADE

People who commercially cultivate medicinal plants in Kerala attempt to mitigate price risks by cultivating medicinal plants as intercrops in coconut gardens. This diversifies the cropping pattern and supplements the farm income (Suneetha and Chandrakanth, 2003).

In order to utilize the natural resources like light, soil nutrients and water efficiently the practice of inter/mixed cropping is followed. Inter or mixed cropping in coconut gardens is very popular in important coconut growing states in India. The transmission of light through the coconut canopy is one of the most important factors affecting the success of intercropping programmes (Kushwah *et al.*, 1973).

Nair *et al.* (1974) observed that with respect to light penetration to the ground, coconut has the advantage of having two periods, initially up to ten years after planting and again twenty years after planting upto senescence of the crop during which intercropping could be practiced.

In coconut plantations of more than 25 years old, 45-50 per cent of the sunlight is infiltrated on to grounds without interception by the coconut (Maheswarappa and Anithakumari, 2002).

Lalithabai and Nair (1982) opined that the factor that decides the compatibility of a crop combination in a cropping system such as coconut culture is the ability of associated crops to come up under shade. This ability varied between species of plants.

Nair *et al.* (1991b) reported the possibility of growing ten medicinal plants as intercrops in 8-20 year old coconut plantations. The potential plants recommended are *Catharanthus roseus*, *Coleus aromaticus*, *Coleus zeylanicus*, *Costus speciosus*, *Ocimum species*, *Plumbago rosea*, *Rauwolfia serpentina*, *R. tetraphylla*, *Curcuma amada* and *Kaempferia galanga*. The growth of these plants was not affected by the shade that prevailed in the coconut plantation.

Nair and Reghunath (2002) evaluated thirteen accessions of *Clitoria ternatea* L. for growth and yield as an intercrop in 18-year-old coconut garden and identified five superior genotypes. Six selected accessions of *Clitoria ternatea* L were evaluated by Resmi and Reghunath (2003a) for growth, nodulation and yield parameters of root *viz.*, root length, root girth, dry weight, nodule count and nodule dry weight under open condition as well as under shaded condition as intercrop in a coconut garden. Results indicated that the crop performance was significantly superior under open condition in all parameters studied, when compared to shade. Resmi and Reghunath (2003b) also reported higher values under open condition for shoot growth and yield of selected high yielding *C. ternatea* accessions.

An economic analysis of five medicinal crops grown in Kerala namely *Kaempferia galanga*, *Plumbago zeylanica*, *Aloe vera*, *Ocimum sanctum* and *Piper longum* indicated that it is a feasible venture giving positive net returns (Suneetha and Chandrakanth, 2003).

2.8 CHEMICAL CONSTITUENTS

2.8.1 In *Indigofera tinctoria*

According to Luckner (1990), secondary metabolites in higher plants are stored in plant cells as energy sources, plant hormones, toxic products for defense, detoxified products, metabolic wastes etc. These metabolites accumulate in several intracellular compartments such as vacuole, cell wall, cytosol and other organelles. A large number of metabolites are stored in the form of glycosides.

The actual colouring agent in indigo dye is indigotin, which as such is not present in the indigo plant. Instead the plant contains indican, which is a colourless glycoside (indoxyl- β -D-glycoside). The name indican was first coined in 1855 by Schunk to describe the indigo precursor. On hydrolysis indican yields glucose and indoxyl. Subsequent oxidation of indoxyl in air results in the precipitation of blue particles of indigotin (Epstein *et al.*, 1967; Finar, 1967; Evans, 1990).

In addition, indigo plants contain trans-indirubin (isoindigotin, red), certain impurities such as cis-indigo (blue), cis-indirubin (isoindirubin, red) indigo brown (isoindigo), indigo gluten, indigo yellow and traces of flavonoids. Altogether these make natural indigo a more pleasing tinge than synthetic indigo (Perkin, 1907).

The colouring matter of *Indigofera*, indigotin ($C_{16}H_{10}O_2N_2$) and indican ($C_{14}H_{17}O_6N$) are present in the lamina and to a small extent in the midribs or rachis. Finar (1967) reported the official name of indigo as indigotin. Indigotin, used to be obtained from plants of the *Indigofera* group, is now prepared synthetically. The indigotin content varies in different species of *Indigofera* and also according to the season and age of the plant (CSIR, 1959). These plants do not contain indigotin but indican, which on hydrolysis with hydrochloric acid or enzymes which occur in the crushed plant, is converted into indoxyl which on oxidation with atmospheric oxygen, gives rise to indigotin (Kochhar, 1981).

The structure of indican and reactions in the degradation of indican are illustrated in Fig. 1.

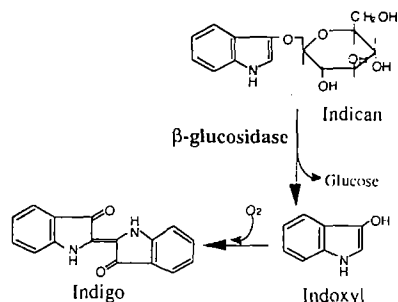


Fig. 1. Hydrolysis of indican by β -glucosidase

Zavatskaya and Mashanova (1978) studied the sugar synthesis in *I. tinctoria* leaves during plant development in relation to accumulation of the glycoside form of the dye. Sugar synthesis was maximal during full bloom when the formation of the dye of the leaves was also high. Organic acid content remained relatively stable during growth, the content of volatile acids reached a peak at fruit development and that of non-volatile acids was low.

Khazhakyán (1986) reported that *Indigofera tinctoria* contains more indigo than other members of the genus. Khazhakyán and Egibyan (1988) isolated indican from the leaves of hydroponically grown plants and the amount was determined by spectrophotometry. It varied from 5 to 15 mg g⁻¹ fresh leaf, depending on experimental conditions.

Ya and Hua (1989) determined the contents of indigo and indirubin in six specimens of natural indigo by first derivative spectrophotometry. The content of indigo was 2.21- 6.90 % and that of indirubin was 0.20- 0.47%.

Preliminary phytochemical studies conducted by Nair *et al.* (1991a) indicated the presence of carbohydrate in chloroform extract of leaves of *I. tinctoria*, which indicates presence of glycoside. It also showed steroids in chloroform, alcohol and water extractions. Presence of protein,

carbohydrate and steroids were also noted in alcohol and water extractions. Tests for saponins were positive only in water extract.

Thomas *et al.* (2000) recorded that indican, the precursor of the blue dyestuff obtained from the indigo plant- *I. tinctoria* is an yellow amorphous material with a nauseous bitter taste with an acid reaction, readily soluble in water, alcohol and ether.

Maugard *et al.* (2001) have shown that young leaves contain higher concentrations of indigo precursors than older ones. In young leaves plants protect indoxyl, a highly unstable intermediate, from spontaneous oxidation to indigo by glycosylation to yield indican. Indican in old leaves is oxidized spontaneously by air yielding indigo.

Kamal and Mangla (1987) found that the rotenoid contents in *I. tinctoria* were 0.51% in roots, 0.32% in stems, 0.60 % in leaves, 0.34% in fruits and 0.42% in seeds. The rotenoids had a LD₅₀ value of 116.8 ppm against *Mesocyclops leuckarti*, the carrier of Guinea worm larvae (*Dracunculus medinensis*). They reported that the rotenoid content of *I. tinctoria* plants decreased with increasing age, the maximum and minimum rotenoid concentration was found in the leaves and stem, respectively. Six rotenoids were isolated. The toxicity of the rotenoids was greater to larvae of *Anopheles stephensi* than adults of *Callosobruchus chinensis* (Kamal and Mangla, 1993).

Maier *et al.* (1990) and Minami *et al.* (2000) reported that besides *I. tinctoria*, *Polygonum tinctorium* also contain indican (indoxyl β -D-glucoside) (plant indican), which serves as the starting material for indigo blue production.

Minami *et al.* (2000) conducted experiments on the indigo plant- *Polygonum tinctorium* and reported that it has some glycosides as secondary metabolites. The major metabolite is a colourless glucoside called indican, indoxyl β -D-glucoside. Indican is stored mainly in the vacuoles of leaf cells, it was not detected in any other tissue. Indican was not detected until the appearance of leaves, and then the indican content

increased with the growth of the leaves. In the mature plants, younger leaves (especially first and second leaves) contained larger amounts of indican per gram of tissue than older ones.

2.8.2 Role of Environment in Secondary Metabolite Production in Medicinal Plants

Chopra *et al.* (1958) found that plants generally owe their virtues as medicinal agents to certain characteristic constituents like alkaloids, glycosides, saponins, flavonoids, tannins, volatile oil, steroids or terpenoids, resin and mucilage present in them. Plants synthesise these organic compounds during their metabolic process when they grow. The amount of active substances present in plants is dependent upon several factors such as the nature of the soil, the climate, the season, the stage of growth of a plant, the nature and intensity of light, cultivation practices etc. That is, the environmental conditions to which the plant is exposed influence the production of these secondary metabolites and ultimately the efficacy of the drug (Miniraj, 1997; Ramamurthi, 2002).

Several reports are there to support these observations. According to Gupta and Chadha (1995), the light intensity plays an important role in many crops. *Coptis japonica*, when grown as a four- year crop, in foothills of Korea in 70 per cent light interception, the berberine content is reported to be 9-10 per cent higher (Hans and Hu, 1987).

Varghese (1989) found that curcumin content of turmeric rhizome showed a progressive decrease with increase in shade. In mints, ample sunshine is necessary at maturity to enable plants synthesise higher content of oil and menthol. The crop is harvested on sunny days only because rainy season or cloudy days convert menthol into menthone, lowering the value of the produce.

Li *et al.* (1996) recorded that the total essential oil concentration in Sage (*Salvia officinalis*) was highest (0.38 % on fresh weight basis) in the plants grown at 45 per cent of full sunlight. In thyme (*Thymus vulgaris*), the

highest essential oil concentration (0.49 % on fresh weight basis), occurred in full sunlight. In *Solanum laciniatum*, solasodine production was found to be 10-12 times higher depending on light intensity (Jaggi and Kapoor, 1997).

Lambers *et al.* (1998) observed that the concentration of caffeine, in the shoot of tea (*Camellia sinensis*) is higher when the plants are grown at higher radiance rather than in shade.

According to Resmi (2001), there was no significant variation between shade and open conditions for the crude alkaloid content (per cent) in seeds of *Clitoria ternatea*.

Jessykutty (2003) recorded that shade under oil palm plantations significantly influenced the oleoresin content of kacholam (*Kaempferia galanga*) rhizomes and the highest content was recorded under open conditions.

Geetha (2004) found that proline content in the leaves of *Kaempferia galanga*, *Alpinia calcarata*, *Pogostemon patchouli* and *Piper longum* decreased significantly with increase in shade intensity and maximum value was registered under open condition.

Materials and Methods

3. MATERIALS AND METHODS

The present work on “Evaluation of Neelayamari (*Indigofera tinctoria* L.) for yield and glycoside content under open and shaded conditions” was conducted at the Department of Plantation Crops and Spices, College of Agriculture, Vellayani, Thiruvananthapuram during 2000-2003. The field experiments were conducted at the Instructional Farm, College of Agriculture, Vellayani. The performance evaluation of selected accessions was made for identifying the best accessions yielding high biomass and glycosides. The details of the materials used and the techniques adopted for the study are presented in this chapter.

The experiments were conducted in two phases. In the phase I experiment, collection of available geographical accessions/ landraces of *Indigofera tinctoria* was made from within and outside the state. Growth and yield analysis of 30 accessions of *Indigofera tinctoria* as pure crop in open and as an intercrop in shaded situation in a coconut garden was done. From these, best 10 accessions were selected based on shoot yield and glycoside content. In the phase II experiment, the selected 10 accessions were raised in replicated trial in open and under shade in coconut garden to identify the genotypes having high biomass yield and more glycoside content.

3.1 EXPERIMENTAL SITE

The experimental site at the College of Agriculture campus at Vellayani, Thiruvananthapuram, Kerala state, was situated at 8.5⁰ N latitude, 76.9⁰ E longitude at an altitude of 29 m above MSL. Experimental site had a lateritic red loam soil belonging to Vellayani series with a pH of 5.2. The area enjoyed a warm humid tropical climate. The meteorological data for the cropping periods are presented in Appendix-I.

3.2 COLLECTION OF ACCESSIONS OF *INDIGOFERA TINCTORIA*

Seeds of thirty different accessions of *Indigofera tinctoria* were collected from inside and outside the state. The accessions were duly entered in the medicinal and aromatic plant accession register of the Department of Plantation Crops & Spices, College of Agriculture, Vellayani and accession numbers were allotted. The details of the accessions and their sources are presented in Table 3. Among the thirty accessions, twelve were collected from Thiruvananthapuram district, one from Kottayam, one from Ernakulam, three from Thrissur, two from Palakkad, one from Malappuram, one from Kozhikode, one from Cannanore, two from Idukki and three from Pathanamthitta districts. One accession was collected from Bangalore, Karnataka state, one from Lucknow, Uttar Pradesh state and one from Nigeria in Africa.

3.3 SEED CHARACTERIZATION AND GERMINATION STUDIES

Observations such as hundred seed weight and seed colour were made on each accession.

Three replicates of twenty seeds of each accession were surface sterilized using 0.1 per cent mercuric chloride for one minute and then washed thrice with distilled water. Sterilized seeds were placed on moistened filter paper in petridish. Seeds were considered to have germinated when the greenish structure of the radicle emerged out of the seed coat. Seed germination count was taken from the second day and the germination percentage of each accession was determined.

3.4 PHASE I EXPERIMENT- GROWTH AND YIELD ANALYSIS OF GENETIC STOCK OF *INDIGOFERA TINCTORIA*

3.4.1 Experimental Design and Layout

The accessions were raised in lines with two replications both in the open condition (Plate 1a.) and under shade in coconut garden consisting of palms of 35 years of age. Twenty- one plants of each accession was grown

Table 3. Particulars of *Indigofera tinctoria* accessions used in the experiment

Sl. No.	Accession No.	District /State/ Country	Source	Date of collection
1	IT-93	Thrissur	Biju Jacob, Muthipeedika house, Ollur P.O., Thrissur.	28/4/2001
2	IT-94	Palakkad	Mahila Society, Kanjikode, Palakkad	28/4/2001
3	IT-95	Pathanamthitta	R.S. Traders, Vennikulam, Thiruvalla	9/5/2001
4	IT-96	Thiruvananthapuram	Seed Bank, TBG&RI, Palode	22/5/2001
5	IT-97	Thiruvananthapuram	Medicinal Plant Garden, TBG&RI, Palode	22/5/2001
6	IT-98	Thiruvananthapuram	Ravi, Palappoor, Vellayani	23/6/2001
7	IT-99	Idukki	Thomas Mathew, Puthenchanta, Mundakkayam	13/7/2001
8	IT-100	Thiruvananthapuram	Instructional Farm, College of Agriculture, Vellayani	19/7/2001
9	IT-101	Thrissur	AICRP on Medicinal & Aromatic Plants, Vellanikkara	28/7/2001
10	IT-102	Idukki	Nagarjuna Herbal Concentrates Ltd., Kalayanthani P.O., Alakode, Thodupuzha	28/7/2001
11	IT-103	Thiruvananthapuram	Girish, Kallikkad P.O., Neyyardam	30/7/2001
12	IT-104	Kottayam	Mathewkutty Theruvapuzha, Secretary, Vrikshabandhu Social Forestry Club, Pala, Kottayam	4/8/2001
13	IT-105	Malappuram	Dr. Indira Balachandran, Aryavaidyasala, Kottakkal	7/8/2001
14	IT-106	Karnataka	University of Agricultural Sciences, Bangalore	8/8/2001
15	IT-107	Uttar Pradesh	National Botanical Research Institute, Lucknow	13/8/2001
16	IT-108	Thiruvananthapuram	Neyyattinkara Market	15/8/2001
17	IT-109	Pathanamthitta	Herbal Garden, State Farming Corporation, Pandalam	18/8/2001
18	IT-110	Thrissur	Department of Plantation crops, College of Horticulture, Vellanikkara, Thrissur	20/8/2001
19	IT-111	Ernakulam	Aromatic and Medicinal Plants Research Station, Odakkali	20/8/2001
20	IT-112	Nigeria	International Institute for Tropical Agriculture, Ibadan, Nigeria	20/8/2001

Table 3. Continued

21	IT-113	Thiruvananthapuram	Shanthigiri Ashram, Pothencode	22/8/2001
22	IT-114	Thiruvananthapuram	Dr. Hareendran Nair, Pankajakasthuri, Poovachal	28/8/2001
23	IT-115	Palakkad	Kanjikode Farm, Walayar	29/8/2001
24	IT-116	Kozhikode	Syamala Radhakrishnan, Chalappuram, Kozhikode	4/9/2001
25	IT-117	Thiruvananthapuram	Rema, 'Meleputhenveedu', Puliyarakonam, Thiruvananthapuram	8/9/2001
26	IT-118	Pathanamthitta	Meera, 'Mynakom', Adoor	25/9/2001
27	IT-119	Thiruvananthapuram	Rajesh, 'Melathilveedu', Ookkode, Vellayani, Thiruvananthapuram	29/9/2001
28	IT-120	Thiruvananthapuram	Kunjan Nadar, Secretary, Pidayani Haritha Sangam, Pappad, Kachani P.O.	4/10/2001
29	IT-121	Thiruvananthapuram	Bhamini, Pattavila melethattu puthen veedu, Athiyannoor, Thannimoodu P.O.	15/10/2001
30	IT-122	Cannanore	Elykutty, Naduvil Village, Thalipparambu Taluk, Cannanore	23/10/2001



a



b

Plate 1. General view of the crop in Phase I & II experiments in flowering stage under open condition
a. Phase I crop
b. Phase II crop

in a line with a spacing of 60 cm between lines and 45 cm within the line. The layout of the experiment is presented in Fig. 2 and details of individual plot in Fig. 3.

From each accession after leaving two border plants, 10 plants were retained for data collection at various growth stages namely pre-flowering, flowering and seed maturation stages. Nine plants were utilised for destructive sampling coinciding with these three stages. Three adjoining plants were sampled at each time.

3.4.2 Field Preparation, Sowing and Cultural Operations

Seeds of 30 accessions were first sown in seed pots containing sand and watered daily. After two weeks, the seedlings were transplanted to polythene covers of 250 gauge and 15 x 10 cm size, filled with soil, sand and cattle manure. After one month, the seedlings were transplanted to the main field. The crop received management practices as per Package of Practices recommendations of Kerala Agricultural University (KAU, 1993). The land was thoroughly prepared by digging and levelling. Trenches were taken at a spacing of 60 cm. Dried and powdered cowdung was incorporated at the rate of 1 kg m⁻². Fortyfive days old seedlings were transplanted into the trenches in the main field at a spacing of 45 cm between plants.

3.4.3 Growth Observations

The following biometric observations were recorded at three growth stages, viz. 1) pre-flowering stage (90 DAS) 2) flowering stage (150 DAS) and 3) seed maturation stage (240 DAS) from ten observational plants in each replication and the mean values were taken.

3.4.3.1 Plant Height (cm)

The height of the plant was measured from the base of the plant to the tip of the tallest branch.

Fig. 2 Layout of the phase 1 experiment

IT 113
IT 103
IT 97
IT 112
IT 115
IT 118
IT 114
IT 96
IT 102
IT 111
IT 121
IT 95
IT 120
IT 101
IT 122
IT 119
IT 104
IT 117
IT 105
IT 93
IT 100
IT 116
IT 106
IT 94
IT 110
IT 107
IT 98
IT 108
IT 99
IT 109

Replication 1

IT 95
IT 106
IT 113
IT 105
IT 98
IT 108
IT 96
IT 120
IT 97
IT 107
IT 102
IT 99
IT 116
IT 104
IT 119
IT 110
IT 93
IT 109
IT 94
IT 118
IT 103
IT 101
IT 112
IT 100
IT 117
IT 122
IT 114
IT 121
IT 111
IT 115

Replication 2

Fig 3. Details of individual plot in phase I experiment

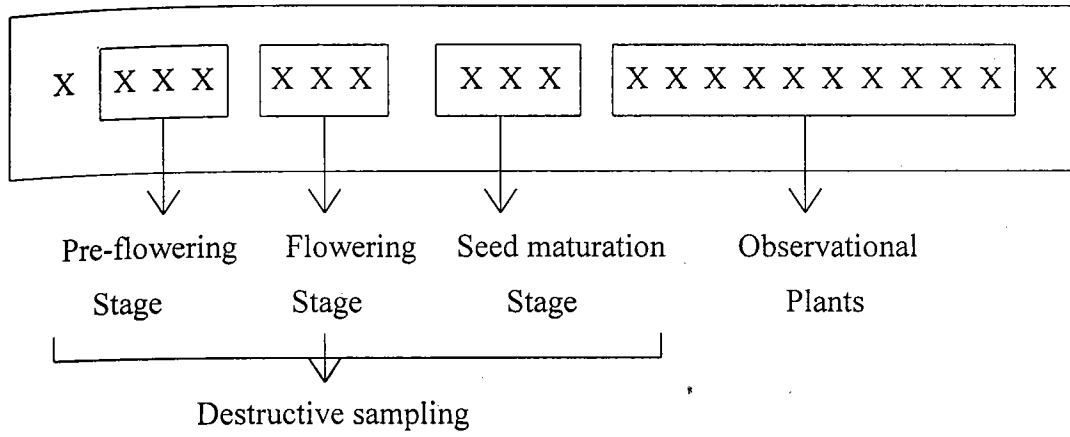


Fig 4. Layout of the phase II experiment

IT 108		IT 106		IT 105
IT 101		IT 111		IT 97
IT 96		IT 108		IT 101
IT 114		IT 97		IT 99
IT 106		IT 96		IT 108
IT 104		IT 101		IT 96
IT 105		IT 104		IT 114
IT 111		IT 99		IT 104
IT 97		IT 114		IT 106
IT 99		IT 105		IT 111
Replication 1		Replication 2		Replication 3

3.4.3.2 Plant Spread (cm)

The distance occupied by the plant in the north-south and east-west direction from its axis was measured and average was worked out.

3.4.3.3 Height at First Branching (cm)

The height at which first branch is produced was measured from the ground level to the position of the first branch.

3.4.3.4 Number of Branches

The total number of branches in a plant was counted and recorded.

3.4.3.5 Girth of Stem (cm)

The girth of main stem at the collar region was taken using a thread and measuring scale.

3.4.3.6 Number of Leaves

The total number of leaves produced in a plant was counted and recorded.

3.4.3.7 Leaf Area (cm²)

Leaf area was calculated by adopting punch method (Watson, 1952). Leaves of the plants earmarked for destructive sampling were separated and 50 punches were made out of them. The discs as well as the leaves were dried in a hot air oven at 70⁰ C and their respective dry weights were recorded. From the data leaf area per plant was computed.

3.4.3.8 Root Length (cm)

Root length was measured from the collar region to the farthestmost tip of the root system.

3.4.3.9 Root Girth at Collar Region (cm)

Root girth at collar region was measured using a thread and measuring scale.

3.4.4 Yield Attributes

3.4.4.1 Fresh and Dry Weight of Shoots (g)

Fresh weight of shoot along with leaves of each observational plant was measured using a counterpoise balance. The samples were then dried in a hot air oven at 70⁰ C until consistent dry weights were obtained.

3.4.4.2 Fresh and Dry Weight of Leaves (g)

Fresh weight of leaves of each observational plant was measured using a counterpoise balance. The samples were then dried in a hot air oven at 70⁰C until consistent dry weights were obtained.

3.4.4.3 Fresh and Dry Weight of Roots (g)

Fresh and dry weight of the root portion of each observational plant was taken in a similar manner as described above.

3.4.4.4 Fresh and Dry Weight of Pods (g)

The pods from observational plants were collected during the seed maturation stage and fresh weight was recorded. The pods were then dried in a hot air oven at 70⁰ C until consistent dry weights were obtained. The seeds were separated from the pods and the shelling percentage was calculated.

3.4.5 Glycoside Estimation

Glycoside content in all the accessions were estimated using the method suggested by Wu *et al.* (1999) with certain modifications using the following treatments:

- 1) Fresh leaves - One gram fresh leaf from the observational plants in each replication was crushed in a mortar filled with liquid nitrogen. It was mixed with 15 ml of 80% methanol. The solution was heated at 70⁰ C for 5 minutes in a water bath. The solution was then stirred for 20 minutes and filtered through a glass funnel. The filtrate was then read at 280 nm in a Spectronic Genesys 5 Spectrophotometer. Indoxyl β -D- glucoside

(indican) standard (Sigma Aldrich) solutions of 0.5 ppm, 1 ppm, 1.5 ppm and 2 ppm were prepared. These were also read at 280 nm in the spectrophotometer. From the standard values, the amount of indican per gram of fresh leaf was calculated.

- 2) Shade dried leaves - Leaves taken from the observational plants in each replication were kept inside brown paper covers and dried under sun for 3 to 5 days till the leaves became completely dried. One gram of leaf from each replication was crushed in a mortar and the above method was followed for glycoside analysis.
- 3) Oven dried leaves - Leaves taken from the observational plants in each replication were kept inside brown paper covers and shade dried for 1 or 2 days and then dried in a hot air oven at 70⁰ C till the leaves became completely dried. One gram of leaf from each replication was crushed in a mortar and the above method was followed for glycoside analysis.
- 4) Fermented leaves - One gram leaf was taken from observational plant in each replication. It was crushed in a mortar using 1 ml distilled water and kept overnight for fermentation. Next day, the above method was followed for glycoside estimation.

The treatment yielding maximum glycoside content was standardised.

3.4.6 Statistical Analysis

A selection index based on leaf yield and glycoside content was worked out to screen the genotypes. The best ten accessions giving maximum shoot yield and glycoside content were selected for the phase II experiment.

3.5 PHASE II EXPERIMENT-CULTURAL TRIAL OF SELECTED ACCESSIONS

The ten selected accessions were raised in replicated trial in open (Plate 1b.) and under shade in coconut garden consisting of palms of 35 years of age (Plate 2).



Plate 2. General view of the crop under shade in pre-flowering stage in Phase II experiment

3.5.1 Experimental Design and Layout

Design	: R B D
Treatments	: 10
Replication	: 3
Plot Size	: 4.5 x 4.05 m
Spacing	: 45 x 45 cm
No. of plants/ plot/ replication	: 90

The layout of the experiment is presented in Fig. 4

From each accession after leaving two border plants, eight plants were retained for data collection at various growth stages namely pre-flowering, flowering and seed maturation stages. Eight plants from each replication was utilised for destructive sampling coinciding with these three stages.

3.5.2 Field Preparation, Sowing and Cultural Operations

Cultural operations were followed as in the case of phase I experiment.

3.5.3 Growth Observations

Biometric observations like plant height, plant spread, height at first branching, number of branches, girth of stem, number of leaves, leaf area, root length and root girth and yield attributes such as fresh and dry weight of shoots, leaves, roots and pods were recorded similar to the phase I experiment. Shelling percentage was also calculated. 100 g mature dry pods were shelled and the weight of seeds thus obtained was recorded.

3.5.4 Root nodule characteristics

3.5.4.1 Number of Root Nodules

Nodules produced in the entire root system of observational plants were detached freshly and their total count per plant was recorded.

3.5.4.2 Number of Effective Root Nodules

With the help of a sharp blade, each nodule was dissected cross sectionally. Nodules, which were firm and possessed a bright red centre were considered as effective root nodules. The count of effective nodules was taken in each observational plant.

3.5.4.3 Fresh and Dry Weight of Root Nodules

Fresh weight of root nodules obtained from each observational plant was taken using an analytic digital weighing balance (Sartorius). The nodules were then dried in a hot air oven at 60⁰ C for three days until consistent dry weights were obtained.

3.5.5 Flowering and Seeding Behaviour of *Indigofera tinctoria*

3.5.5.1 Number of Days for Flowering

Total number of days required for commencing flowering from the date of sowing in fifty per cent of plants in a plot was recorded in each accession.

3.5.5.2 Total Number of Flowers Produced Per Plant

From each accession, after leaving border plants, eight plants were selected and the total number of flowers produced per plant was counted and the mean value recorded.

3.5.5.3 Time of Anthesis

Ten mature flower buds in every accession in a replication were tagged at 6 am and the time of flower opening was noted and the mean value recorded.

3.5.5.4 Time of Anther Dehiscence

Ten inflorescence in every treatment in a replication were cut in the evening and dipped in a vessel containing water and kept for observation.

Mature but unopened flower buds were observed with a hand lens at night for anther dehiscence. Appearance of longitudinal split in the pollen sac indicated the commencement of anther dehiscence. Observations were taken and the mean value recorded.

3.5.5.5 Number of Pollen Grains/Anther

Number of pollen grains/anther was estimated using a haemocytometer, as suggested by Rao and Khader (1962).

Mature flower buds from the observational plants in each replication in an accession were collected. One or two anthers were crushed in 1 ml distilled water. One drop was placed on the haemocytometer and observed through the low power (10x10X) of the compound microscope. Every time, the first large square with 16 small squares was observed for the presence of pollen grains. Total number of pollen grains in that square was recorded.

$$\left[\begin{array}{l} \text{Total number of pollen} \\ \text{grains/anther} \end{array} \right] = \frac{X \times Y \times \text{constant}}{\text{No. of small squares} \times \text{No. of anthers crushed}}$$

Constant for 1st square = 1.6 lakh

X = total number of pollen grains in the first square

Y = dilution factor (here one)

3.5.5.6 Pollen Fertility

Fertility of pollen grains was assessed on the basis of stainability of pollen grains in acetocarmine glycerine mixture (Shivanna and Johri, 1985). Pollen grains were extracted from fully matured anthers using a needle and stained in a drop of acetocarmine glycerine mixture on a clean slide and kept aside for one hour. All the pollen grains that were stained were counted as fertile. Pollen fertility was expressed as percentage.

3.5.5.7 Percentage of Pod Set

The total number of pods produced in an inflorescence in each replication in an accession was noted in the observational plants. The percentage of pod set was calculated using the total number of flowers produced per inflorescence.

3.5.5.8 Number of Days for Seed Set

Total number of days required for 50 per cent of plants in a plot to attain seed set from the date of sowing was recorded in each accession.

3.5.5.9 Number of Days for Seed Maturation

Total number of days required for 50 per cent of plants in a plot to attain seed maturation from the date of sowing was recorded in each accession.

3.5.6 Growth and Yield Analysis

3.5.6.1 Leaf Area Index (LAI)

Leaf area of observational plants, were calculated by adopting punch method. Leaf area index was worked out using the formula proposed by Watson (1952),

$$\text{LAI} = \frac{\text{Leaf area of the plant (cm}^2\text{)}}{\text{Ground area occupied (cm}^2\text{)}}$$

3.5.6.2 Leaf Area Duration (LAD) (days)

Leaf area duration was calculated using the formula of Power *et al.* (1967),

$$\text{LAD} = \frac{(L_1 + L_2) (T_2 - T_1)}{2}$$

where,

L_1 - L A I at time T_1

L_2 - L A I at time T_2

$(T_2 - T_1)$ - time interval in months

3.5.6.3 Net Assimilation Rate (NAR) ($g\ m^{-2}\ day^{-1}$)

Net assimilation rate (NAR) refers to the change in dry weight of the plant per unit leaf area per unit time. The procedure given by Watson (1958) and modified by Buttery (1970) was followed for calculating NAR.

$$NAR = \frac{W_2 - W_1}{(T_2 - T_1) \frac{(A_1 + A_2)}{2}}$$

where,

W_2 - total dry weight of plant (g) at time T_2

W_1 - total dry weight of plant (g) at time T_1

$(T_2 - T_1)$ - time interval in days

A_1 - leaf area (m^2) time T_1

A_2 - leaf area (m^2) at time T_2

3.5.6.4 Crop Growth Rate (CGR) ($g\ m^{-2}\ day^{-1}$)

Crop growth rate (CGR) is the rate of increase in dry weight per unit ground area. CGR was calculated using the formula given by Watson (1958).

$$CGR = \frac{(W_2 - W_1)}{P (T_2 - T_1)}$$

where,

W_1 and W_2 - plant dry weights (g) at time T_1 and T_2 respectively

$(T_2 - T_1)$ - time interval in days

P - ground area in m^2 on which W_1 and W_2 are estimated.

3.5.6.5 Absolute Growth Rate (AGR) ($g\ day^{-1}$)

Absolute growth rate (AGR) gives an idea of daily growth rate. AGR was determined using the formula given by Watson (1958).

$$\text{AGR} = \frac{(W_2 - W_1)}{(T_2 - T_1)}$$

where,

W_1 and W_2 - plant weights (g) at time T_1 and T_2

$(T_2 - T_1)$ - time interval in days

3.5.6.6 Relative Growth Rate (RGR) (g day^{-1})

Relative growth rate (RGR) is the rate of increase in dry weight per unit time. RGR was calculated as per the formula suggested by Williams (1946).

$$\text{RGR} = \frac{\log_e W_2 - \log_e W_1}{(T_2 - T_1)}$$

where,

W_1 and W_2 - plant dry weights (g) at time T_1 and T_2

$(T_2 - T_1)$ - time interval in days

3.5.6.7 Dry Matter Accumulation Per Plant (g plant^{-1})

Shoots, leaves and roots of the uprooted plants were separated and dried to a constant weight at 70° C in a hot air oven during the pre-flowering, flowering and seed maturation stages. The sum of these individual components gave the total dry matter accumulation of the plant.

3.5.6.8 Harvest Index (HI)

Harvest index (HI) was calculated at final harvest using the formula

$$\text{HI} = \frac{\text{Y econ.}}{\text{Y biol.}}$$

where,

Y econ. - dry weight of officinal part

Y biol. - total dry weight of plant

3.5.7 Phytochemical observations

3.5.7.1 Soil NPK Analysis

Common soil samples were taken from the experimental site before the commencement of experiment and after the harvest of the crop from every plot. Samples were taken at 0-15 cm depth, shade dried and passed through a 2 mm sieve.

The available nitrogen status was estimated using the alkaline potassium permanganate method (Subbiah and Asija, 1956) and expressed in kg ha⁻¹. Bray's colourimetric method (Jackson, 1973) was used to estimate the available phosphorous. The available potassium content was estimated using the ammonium acetate method given by Jackson (1973).

3.5.7.2 Plant Analysis for N Content

Nitrogen content of plant samples collected from open and shaded conditions was estimated after drying them in a hot air oven for five days at 60^o C and ground to fine powder. Nitrogen content of the plant was determined by micro kjeldahl digestion in sulphuric acid and distillation (Jackson, 1973).

3.5.7.3 Plant Analysis for Glycoside Content

Glycoside content in the fresh leaves of all the ten accessions was estimated using the method suggested by Wu *et al.* (1999) as described in phase I experiment.

3.5.8 Statistical Analysis

Qualitative and quantitative parameters of 30 accessions under trial was analysed by applying the analysis of variance technique as applied to randomised block design described by Cochran and Cox (1965) and the significance was tested by F test (Snedecor and Cochran, 1967). In cases where the effects were found to be significant, CD values were calculated by using standard technique.

In phase I experiment, a selection index based on leaf yield and glycoside content was worked out to screen the genotypes. In phase II experiment, correlation studies were made so as to find out the factors that contribute to yield and glycoside content.

3.6 ECONOMIC ANALYSIS

Economics of cultivation of *Indigofera tinctoria* both in open and under shade in coconut garden was worked out after taking into account the cost of cultivation and the prevailing market price.

The net income and benefit cost ratio was calculated as follows :

Net income (Rs. ha⁻¹) = Gross income - Cost of cultivation

$$\text{Benefit - cost ratio} = \frac{\text{Gross income}}{\text{Cost of cultivation}}$$

3.7 INCIDENCE OF PESTS AND DISEASES

Throughout the duration of the two crops, periodical surveillance was done for the detection of pests and diseases.

Results

4. RESULTS

The results of the present investigation entitled "Evaluation of Neelayamari (*Indigofera tinctoria* L.) for yield and glycoside content under open and shaded conditions " are presented in this chapter. The study was conducted in two phases during 2001-2003, at the Department of Plantation crops and Spices, College of Agriculture, Vellayani, Thiruvananthapuram.

4.1 SEED CHARACTERIZATION AND GERMINATION STUDY IN *INDIGOFERA TINCTORIA* ACCESSIONS

Seeds of thirty accessions of *Indigofera tinctoria* collected from inside and outside the state, were subjected to characterization and germination study. List of collected accessions and their sources are given in Table 3. The results of the seed germination test are presented in Table 4. Observations on the hundred seed weight and seed characters such as seed colour, external appearance of seeds and germination percentage were recorded for each accession.

Five accessions IT-98, IT-95, IT-105, IT-108 and IT-106 exhibited more than 80 per cent germination. IT-111 and IT-102 exhibited poor germination percentage of 47 and 48 respectively. Hundred seed weight of the accessions ranged from 0.28-0.56g. Of the thirty accessions collected for the study, IT-104 recorded the highest hundred seed weight of 0.56g.

4.2 PHASE I EXPERIMENT- GROWTH AND YIELD ANALYSIS OF THE GENETIC STOCK OF *INDIGOFERA TINCTORIA* AS PURE CROP AND INTERCROP IN COCONUT GARDEN

Thirty accessions of *Indigofera tinctoria* collected from inside and outside the state were raised in lines with two replications each for open as well as shaded condition in coconut garden consisting of palms of 35 years

Table 4. Seed characters and germination of *I. tinctoria* accessions

Sl. No.	Accession No.	Hundred seed weight (g)	Seed characters	Leaf colour characteristics	Germination (%)
1	IT-93	0.38	Light to dark brown, medium size, bold	Bluish green	74
2	IT-94	0.48	Brown to black, bigger, bold	Bluish green	72
3	IT-95	0.43	Light brown, circular to cubic, bold	Bluish green	83
4	IT-96	0.43	Brown, medium, bold	Bluish green	63
5	IT-97	0.32	Light brown, round, bold	Bluish green	74
6	IT-98	0.34	Black, cylindrical, bold	Bluish green	84
7	IT-99	0.28	Brown to black, cubic, depressions on surface	Bluish green	57
8	IT-100	0.52	Brown, round, medium, bold	Bluish green	60
9	IT-101	0.32	Light to dark brown, medium, bold	Bluish green	73
10	IT-102	0.34	Light to dark brown, medium, cubic, bold	Bluish green	48
11	IT-103	0.34	Dark brown to black, larger, depressions on surface	Bluish green	64
12	IT-104	0.56	Light to dark brown & black, cubic, bold	Bluish green	63
13	IT-105	0.48	Light brown, round, bold	Bluish green	83
14	IT-106	0.38	Cream, round, medium, bold	Bluish green	82
15	IT-107	0.28	Grayish brown, elongated, bold	Green	73
16	IT-108	0.36	Light brown, round, bold	Bluish green	83
17	IT-109	0.40	Orange to dark brown, medium, bold	Bluish green	75
18	IT-110	0.36	Brownish black, medium, bold	Bluish green	72
19	IT-111	0.46	Ash, brown & black, larger, bold	Bluish green	47
20	IT-112	0.28	Ash, shiny, elongated, bold	Green	60
21	IT-113	0.30	Dark brown to black, medium	Green	75
22	IT-114	0.38	Brown to black, round, bold	Bluish green	74
23	IT-115	0.36	Dark brown to black, bold, cubic	Green	53
24	IT-116	0.42	Ash, brown & black, bigger, bold	Bluish green	66
25	IT-117	0.39	Brown, cubic, bold	Bluish green	73
26	IT-118	0.37	Orange to dark brown, medium, bold	Bluish green	60
27	IT-119	0.32	Black, cubic, medium, depressions on surface	Bluish green	58
28	IT-120	0.35	Dark brown, round, bold	Bluish green	73
29	IT-121	0.50	Brownish black, bigger, bold	Bluish green	61
30	IT-122	0.30	Light to dark brown, medium, bold	Bluish green	73

of age. The average light intensity under shade was about 35 per cent of that under open condition. Observations were made at three stages of growth viz., pre-flowering stage (90 DAS), flowering stage (150 DAS) and seed maturation stage (240 DAS).

4.2.1 Growth parameters

4.2.1.1 Plant height

At pre-flowering stage, no significant difference could be noticed among the accessions under shade, but significant difference was noticed under open condition (Table 5). Maximum plant height (109.36 cm) was recorded by IT-116 under open condition, followed by IT-106 (95.88 cm), IT-113 (93.99 cm) and IT-105 (92.46 cm), which were statistically on par. The least plant height (53.59 cm) was recorded by IT-117.

At flowering stage, significant difference could be noticed among the accessions under shade only. In shade, maximum plant height (103.90 cm) was recorded by IT-108. It was on par with IT-106 (98.83 cm), IT-105 (97.83 cm) and IT-104 (94.64 cm). The lowest plant height (81.56 cm) was recorded by IT-119.

At seed maturation stage, there was significant difference among the accessions under open and shaded conditions. Under open condition, maximum plant height (232.68 cm) was recorded by IT-113, which was on par with IT-112 (225.60 cm), IT-115 (223.98 cm) and IT-108 (222.7 cm). Least plant height (154.34 cm) was recorded by IT-98. Under shade condition, IT-96 recorded the highest value for plant height (144.20 cm), followed by IT-106 (141.20), IT-104 (139.24 cm), IT-108 (139.09 cm), IT-101 (138.61 cm), IT-113 (137.69 cm) and IT-105 (137.15 cm), which were statistically on par.

Plant height was more in open condition than that under shade at all the three growth stages. IT-106, IT-108, IT-113 and IT-105 showed better performance compared to the other accessions.

Table 5. Growth of *I. tinctoria* accessions under open and shaded conditions: plant height (cm)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT-93	58.69	44.28	100.99	86.76	164.73	123.26
2	IT-94	77.62	50.07	111.75	83.74	170.71	118.07
3	IT-95	79.60	50.76	141.94	86.72	158.06	128.00
4	IT-96	62.75	54.54	131.90	91.81	199.30	144.20
5	IT-97	71.88	49.94	146.58	88.74	169.80	124.94
6	IT-98	81.51	49.10	126.57	89.87	154.34	128.16
7	IT-99	71.03	54.53	116.54	85.02	197.17	118.70
8	IT-100	73.33	54.26	155.75	85.88	176.65	118.96
9	IT-101	79.24	55.27	129.60	92.88	209.19	138.61
10	IT-102	70.36	51.46	101.35	87.96	197.60	120.11
11	IT-103	76.16	52.73	131.49	85.39	175.60	128.67
12	IT-104	73.31	51.29	131.66	94.64	169.48	139.24
13	IT-105	92.46	48.55	154.34	97.83	190.43	137.15
14	IT-106	95.88	48.90	134.14	98.83	183.25	141.20
15	IT-107	71.87	51.38	124.95	91.77	183.82	120.66
16	IT-108	88.99	53.60	147.42	103.90	222.70	139.09
17	IT-109	60.91	42.46	152.39	86.52	195.14	122.44
18	IT-110	69.61	48.79	186.18	85.05	216.62	125.48
19	IT-111	66.63	46.68	136.11	86.11	200.63	133.31
20	IT-112	83.99	40.03	154.96	87.27	225.60	121.06
21	IT-113	93.99	54.28	134.30	92.90	232.68	137.69
22	IT-114	70.86	45.83	121.73	88.94	197.05	131.70
23	IT-115	61.12	51.26	132.05	92.99	223.98	125.96
24	IT-116	109.36	44.54	133.75	86.61	163.34	117.78
25	IT-117	53.59	41.32	123.40	86.61	160.96	121.22
26	IT-118	72.81	44.53	109.70	87.74	168.41	118.02
27	IT-119	68.44	43.79	138.45	81.56	175.86	119.31
28	IT-120	61.18	46.59	120.60	82.82	157.54	121.50
29	IT-121	57.27	49.39	137.87	84.19	176.67	121.01
30	IT-122	65.79	45.04	112.05	86.29	163.35	120.28
	SE	6.24	3.45	14.43	3.27	15.67	4.57
	CD	18.03	NS	NS	9.45	45.24	13.19

4.2.1.2 Plant spread

At pre-flowering stage, significant difference could be noticed among the accessions under open as well as shaded conditions (Table 6). Under open condition, maximum plant spread (116.74 cm) was recorded by IT-113. It was on par with IT-106 (115.07 cm), IT-108 (114.42 cm) and IT-105 (110.61 cm). IT-93 recorded the minimum plant spread (58.27 cm). Under shade, IT-96 recorded the maximum plant spread (59.93 cm) followed by IT-104 (54.71 cm). Least plant spread (29.82 cm) was observed in IT-107.

At flowering stage also, there was significant difference among the accessions under both the conditions. Under open condition, maximum plant spread (151.44 cm) was recorded by IT-106, followed by IT-108 (145.09 cm), IT-105 (140.86 cm) and IT-101 (139.59 cm). Minimum plant spread (81.99 cm) was observed in IT-120. In shade, IT-106 recorded the maximum plant spread (90.31 cm), which was on par with IT-96 (86.84 cm), IT-104 (85.70 cm) and IT-101 (81.76 cm). Least plant spread (51.09 cm) was observed in IT-103.

At seed maturation stage also, significant difference could be noticed among the accessions under both open and shaded conditions. Under open condition, plant spread was maximum in IT-108 (190.38 cm), which was on par with IT-105 (177.51 cm) and IT-96 (176.97 cm). It was minimum in IT-99 (121.04 cm). In shaded condition, IT-118 was found to have maximum plant spread (210.85 cm). Minimum plant spread was (77.00 cm) observed in IT-103.

Plant spread was found to be higher in open condition compared to shade and irrespective of light condition, IT-106 performed better compared to other accessions.

4.2.1.3 Height at first branching

At pre-flowering stage, there was significant difference among the accessions under open and shaded conditions (Table 7). In open condition,

Table 6. Growth of *I. tinctoria* accessions under open and shaded conditions: plant spread (cm)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT-93	58.27	38.99	120.51	70.99	145.79	99.58
2	IT-94	64.09	39.99	105.60	63.28	130.53	88.12
3	IT-95	59.61	41.75	130.01	55.74	139.09	81.07
4	IT-96	92.05	59.93	131.14	86.84	176.97	100.95
5	IT-97	79.91	35.04	127.39	70.51	144.54	90.20
6	IT-98	80.31	37.69	120.09	62.00	149.00	97.18
7	IT-99	71.93	43.15	101.83	59.28	121.04	93.79
8	IT-100	79.50	35.30	125.49	60.71	135.24	79.51
9	IT-101	78.56	31.64	139.59	81.76	165.06	117.03
10	IT-102	63.30	30.74	103.21	58.34	130.45	88.62
11	IT-103	75.82	32.18	104.19	51.09	135.02	77.00
12	IT-104	105.56	54.71	133.42	85.70	158.76	109.43
13	IT-105	110.61	50.31	140.86	80.20	177.51	109.52
14	IT-106	115.07	38.21	151.44	90.31	164.55	118.55
15	IT-107	91.71	29.82	123.20	58.43	148.01	96.57
16	IT-108	114.42	45.65	145.09	77.99	190.38	115.89
17	IT-109	71.82	33.20	111.53	66.24	137.70	92.50
18	IT-110	90.08	31.08	111.07	62.36	130.01	89.06
19	IT-111	87.21	51.01	139.28	76.78	166.00	102.21
20	IT-112	101.56	33.75	135.40	59.58	150.16	91.96
21	IT-113	116.74	36.01	130.48	70.08	149.47	101.56
22	IT-114	100.14	49.99	138.21	72.73	167.12	109.92
23	IT-115	91.25	34.65	115.39	67.20	162.33	90.91
24	IT-116	94.01	47.89	117.28	74.68	140.56	91.89
25	IT-117	79.52	41.04	117.19	72.47	139.15	97.42
26	IT-118	78.48	41.49	119.25	64.51	139.98	110.85
27	IT-119	71.09	40.83	84.64	64.09	131.20	94.91
28	IT-120	69.90	49.56	81.99	66.82	130.07	88.47
29	IT-121	65.87	51.22	91.61	72.08	138.23	94.03
30	IT-122	70.78	51.11	100.50	62.32	149.19	96.03
	SE	6.81	4.87	6.22	4.29	6.58	3.66
	CD	13.92	9.95	12.71	8.76	13.44	7.48

Table 7. Growth of *I. tinctoria* accessions under open and shaded conditions: height at first branching (cm)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT-93	3.76	15.28	5.85	15.78	8.05	16.19
2	IT-94	4.11	13.05	6.13	13.25	7.40	13.41
3	IT-95	5.59	10.34	7.15	11.19	8.13	11.39
4	IT-96	6.92	11.09	7.90	11.69	10.38	12.66
5	IT-97	4.62	11.27	6.39	12.43	7.73	13.46
6	IT-98	4.15	15.32	6.94	16.23	8.10	20.20
7	IT-99	3.62	8.29	6.94	10.57	8.14	16.94
8	IT-100	4.24	9.56	5.88	10.88	7.85	15.28
9	IT-101	6.89	13.50	7.91	14.83	12.10	16.57
10	IT-102	3.80	13.05	5.39	13.92	7.80	15.77
11	IT-103	4.14	16.12	5.59	18.11	7.55	19.19
12	IT-104	6.00	19.71	7.30	20.79	8.53	21.88
13	IT-105	5.78	13.56	7.94	14.60	8.70	16.23
14	IT-106	5.38	13.64	7.19	14.21	9.78	15.44
15	IT-107	5.01	10.59	6.10	12.00	7.99	13.21
16	IT-108	8.05	19.05	8.65	20.31	11.55	21.26
17	IT-109	4.92	13.99	6.18	15.49	8.83	16.90
18	IT-110	4.69	11.46	6.07	12.78	8.04	13.18
19	IT-111	7.09	10.58	8.31	11.58	10.98	12.49
20	IT-112	4.81	9.05	6.35	10.93	7.58	10.99
21	IT-113	6.78	11.75	8.09	12.39	9.82	13.54
22	IT-114	6.57	11.93	7.98	13.49	11.48	13.60
23	IT-115	5.84	12.23	8.05	12.74	12.60	13.02
24	IT-116	4.46	10.01	6.61	11.46	8.03	12.65
25	IT-117	4.47	13.30	6.88	13.88	8.91	14.29
26	IT-118	4.79	9.54	6.14	11.79	8.31	13.03
27	IT-119	5.11	12.51	6.07	13.54	8.77	14.25
28	IT-120	4.76	12.60	5.78	13.55	8.01	15.02
29	IT-121	5.61	10.83	6.78	11.54	7.83	12.02
30	IT-122	4.96	12.39	6.78	12.68	7.50	12.86
	SE	0.65	4.56	0.63	1.25	0.71	0.99
	CD	1.87	4.52	1.81	3.61	2.06	2.87

height at first branching was maximum in IT-108 (8.05 cm). It was on par with IT-111 (7.09 cm), IT-101 (6.89 cm) and IT-113 (6.78 cm). Height at first branching was least in IT-99 (3.62 cm). Under shade, maximum height at first branching was recorded by IT-104 (19.71 cm) followed by IT-108 (19.05 cm). IT-112 recorded the lowest value (9.05 cm).

At flowering stage also, there was significant difference among the accessions under both the conditions. In open condition, height at first branching was maximum in IT-108 (8.65 cm), which was on par with IT-111 (8.31 cm). Lowest value was noticed in IT-103 (5.59 cm). Under shade, maximum height at first branching (20.79 cm) was recorded by IT-104, which was on par with IT-108 (20.31 cm).

At seed maturation stage also, there was significant difference among the accessions under both the conditions. Maximum height at first branching (12.60 cm) in open condition was recorded by IT-115 followed by IT-101 (12.10 cm). Minimum value was observed in IT-94 (7.40 cm). Under shade, maximum value (21.26 cm) was recorded by IT-108 and minimum value (10.99 cm) by IT-112.

Height at first branching was more in shaded condition than that under open at all the three stages of plant growth.

4.2.1.4 Number of branches

At pre-flowering stage, number of branches differed significantly among the accessions under shade, whereas no significant difference was observed under open condition (Table 8). Under shade, maximum number of branches (13.80) was recorded by IT-96 followed by IT-119 (12.94). The lowest number of branches (5.21) was recorded by IT-109.

At flowering stage, all the accessions showed significant difference among them under both open and shade conditions. Under open condition, IT-105 recorded the highest number of branches (84.58). IT-101 (83.28), IT-104 (82.81) and IT-106 (82.69) were on par. The least number of branches (44.28) was recorded by IT-99. Under shaded condition, maximum

Table 8. Growth of *I. tinctoria* accessions under open and shaded conditions: number of branches

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT-93	20.53	4.99	58.02	14.61	87.34	31.66
2	IT-94	19.43	6.80	55.92	17.16	85.75	35.38
3	IT-95	24.91	8.72	62.38	17.86	76.76	30.28
4	IT-96	29.25	13.80	78.03	23.18	103.38	39.73
5	IT-97	21.46	10.43	55.39	16.82	68.68	34.72
6	IT-98	32.01	7.60	50.55	20.19	72.53	39.09
7	IT-99	29.40	8.69	44.28	14.50	91.57	33.35
8	IT-100	25.25	6.91	57.33	14.38	96.99	36.26
9	IT-101	32.11	7.28	83.28	17.93	133.49	38.84
10	IT-102	21.25	7.31	51.76	21.18	86.90	29.06
11	IT-103	23.70	6.89	65.12	22.99	84.50	33.94
12	IT-104	28.22	7.25	82.81	21.96	126.40	38.48
13	IT-105	36.44	4.33	84.58	19.42	110.43	38.23
14	IT-106	35.61	6.50	82.69	19.91	124.87	32.23
15	IT-107	24.17	5.37	68.99	15.35	72.31	29.93
16	IT-108	31.33	4.94	91.67	25.54	151.86	34.40
17	IT-109	17.68	5.21	63.84	13.45	82.06	32.04
18	IT-110	22.86	5.81	60.69	14.04	74.05	36.08
19	IT-111	29.80	8.83	79.94	21.63	112.70	44.74
20	IT-112	26.54	6.53	59.72	18.19	80.29	32.99
21	IT-113	29.79	8.36	75.25	20.71	131.53	36.90
22	IT-114	21.36	9.61	81.92	22.27	119.39	38.55
23	IT-115	20.98	10.20	67.45	22.19	129.92	45.42
24	IT-116	26.65	8.37	67.38	17.02	84.57	31.55
25	IT-117	22.49	9.53	71.24	16.39	89.63	34.04
26	IT-118	21.72	10.27	63.76	18.57	81.59	33.80
27	IT-119	21.85	12.94	66.60	18.09	81.55	37.31
28	IT-120	21.26	8.81	69.83	16.14	82.72	36.50
29	IT-121	20.57	7.03	70.05	17.63	89.23	35.01
30	IT-122	24.25	9.50	68.38	17.75	91.08	34.58
	SE	4.23	0.98	6.19	0.98	8.16	1.73
	CD	NS	2.82	17.87	2.84	23.55	4.99

number of branches (25.54) was observed in IT-108 followed by IT-96 (23.18) and IT-103 (22.99) which were on par. IT-109 recorded the least number of leaves (13.45).

At seed maturation stage, number of branches differed significantly among the accessions both under open and shaded conditions. Under open condition, maximum number of branches was recorded by IT-108 (151.86). It was on par with IT-101 (133.49), IT-113 (131.53) and IT-115 (129.92). IT-97 recorded the lowest number of branches (68.68). Under shade, IT-115 recorded the highest number of branches (45.42) followed by IT-111 (44.74). The least (29.06) was recorded by IT-102.

All the accessions produced more number of branches under open condition at all the three stages of growth, compared to shaded condition.

4.2.1.5 Girth of stem

At pre-flowering stage, significant difference could be observed for stem girth among the accessions under shade, but there was no significant difference among them under open condition (Table 9). In shade, maximum stem girth (1.44 cm) was recorded by IT-106, closely followed by IT-108 (1.41 cm) and IT-96 (1.39 cm) and they were on par. The lowest stem girth (0.84 cm) was recorded by IT-100.

The accessions differed significantly for girth of stem under open as well as shade at flowering stage. Under open condition, IT-101 recorded the highest stem girth (6.03 cm) followed by IT-108 (5.37 cm) and IT-100 (5.34 cm). IT-99 recorded the lowest stem girth (3.03 cm). Under shaded condition, maximum stem girth (2.93 cm) was recorded by IT-108. IT-114 (2.87 cm) and IT-113 (2.70 cm) were on par with IT-108. IT-93 and IT-109 recorded the lowest stem girth (1.60 cm).

Significant difference was observed among the accessions under open as well as shaded condition at seed maturation stage. Under open condition, at seed maturation stage, maximum stem girth (12.99 cm) was recorded by IT-101. It was on par with IT-108 (12.68 cm) and IT-104

Table 9. Growth of *I. tinctoria* accessions under open and shaded conditions: girth of stem

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT-93	1.83	0.88	3.75	1.60	7.06	2.88
2	IT-94	1.84	1.06	3.40	1.84	7.25	2.99
3	IT-95	2.25	1.10	4.30	1.87	7.51	2.90
4	IT-96	2.17	1.39	4.90	2.47	11.15	3.98
5	IT-97	2.03	1.00	4.28	1.94	9.75	3.08
6	IT-98	3.19	1.03	3.45	2.28	6.98	3.87
7	IT-99	2.51	0.88	3.03	1.90	8.37	3.35
8	IT-100	2.99	0.84	5.34	1.81	8.08	3.18
9	IT-101	3.28	1.09	6.03	2.09	12.99	3.80
10	IT-102	2.30	1.02	3.40	1.96	10.12	3.21
11	IT-103	2.93	0.90	4.22	1.92	9.06	3.26
12	IT-104	2.90	1.31	5.01	2.18	12.42	3.63
13	IT-105	2.89	1.33	5.06	2.56	9.15	3.75
14	IT-106	2.62	1.44	5.22	2.61	9.48	3.82
15	IT-107	2.03	0.95	3.38	2.06	6.92	3.39
16	IT-108	1.83	1.41	5.37	2.93	12.68	4.02
17	IT-109	1.95	0.85	4.35	1.60	7.30	3.65
18	IT-110	1.88	0.86	4.18	1.87	7.24	3.29
19	IT-111	2.61	1.09	4.37	2.67	9.18	3.35
20	IT-112	2.57	0.94	4.43	2.07	8.08	3.23
21	IT-113	2.62	1.28	5.16	2.70	10.72	3.90
22	IT-114	2.23	1.34	4.51	2.87	12.00	3.85
23	IT-115	2.65	1.31	4.60	2.41	11.08	3.78
24	IT-116	2.30	0.97	3.87	2.19	6.67	3.29
25	IT-117	1.96	0.85	4.03	2.01	7.75	3.10
26	IT-118	2.11	0.89	4.30	2.19	8.53	3.04
27	IT-119	1.88	1.09	4.32	2.14	8.81	3.22
28	IT-120	2.19	0.93	4.39	2.39	8.37	3.27
29	IT-121	2.38	1.12	4.41	2.27	8.61	3.05
30	IT-122	2.79	0.88	3.54	1.92	8.41	3.23
	SE	0.50	0.10	0.33	0.09	1.14	0.08
	CD	NS	0.28	0.94	0.25	3.29	0.23

(12.42 cm). IT-116 recorded the least stem girth (6.67 cm). In shade, IT-108 recorded maximum girth of stem (4.02 cm), which was on par with IT-96 (3.98 cm), IT-113 (3.90 cm) and IT-98 (3.87 cm). Least stem girth (2.88 cm) was recorded by IT-93.

Stem girth was more in open condition than that in shade at all the three growth stages.

4.2.1.6 Number of leaves

At pre-flowering stage, there was significant difference among the accessions for number of leaves under shaded condition only (Table 10). Maximum number of leaves (107.27) was recorded by IT-104 under shade. IT-104 was on par with IT-98 (106.08), IT-117 (98.33) and IT-111 (98.04). Lowest number of leaves (51.09) was observed in IT-95.

At flowering stage, number of leaves differed significantly among the accessions under open and shaded conditions. Under open condition, maximum number of leaves (680.74) was recorded by IT-104 followed by IT-114 (678.88), IT-113 (662.72) and IT-106 (656.59). The lowest number of leaves (462.58) was recorded by IT-99. In shade, IT-108 (243.13) produced maximum number of leaves. Minimum number of leaves (142.02) was produced by IT-93.

At seed maturation stage, all the accessions showed significant difference under both open and shaded conditions. Under open condition, IT-104 recorded the highest number of leaves (965.78), followed by IT-114 (961.73) and IT-108 (952.71). The least (643.88) was recorded by IT-94. Under shade, maximum number of leaves (444.50) was observed in IT-106 closely followed by IT-105 (444.18). IT-93 recorded the least number of leaves (318.69).

More number of leaves were produced by all the accessions at all stages of growth under open condition than under shade.

Table 10. Growth of *I. tinctoria* accessions under open and shaded conditions: number of leaves

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 93	123.53	54.99	603.84	142.02	665.25	318.69
2	IT- 94	126.61	53.82	513.75	180.29	643.88	361.30
3	IT- 95	179.29	51.09	546.41	162.80	715.55	353.90
4	IT- 96	305.52	98.32	618.42	207.93	907.07	369.08
5	IT- 97	171.46	74.29	616.06	169.53	789.70	336.02
6	IT- 98	234.50	106.08	614.17	190.55	775.29	383.69
7	IT- 99	216.55	67.02	462.58	177.07	670.73	329.22
8	IT-100	261.34	82.92	618.39	187.90	874.99	341.97
9	IT-101	312.78	92.51	605.37	169.39	946.94	384.52
10	IT-102	146.64	88.54	564.44	199.34	777.85	340.22
11	IT-103	161.99	84.31	502.37	183.31	769.59	328.85
12	IT-104	221.94	107.27	680.74	210.27	965.78	414.45
13	IT-105	228.97	97.10	735.23	189.98	839.23	444.18
14	IT-106	248.00	91.64	656.59	210.19	940.93	444.50
15	IT-107	220.90	78.73	482.53	184.02	703.79	381.08
16	IT-108	194.09	96.55	527.78	243.13	952.71	394.12
17	IT-109	190.65	63.02	503.30	189.67	745.55	420.72
18	IT-110	209.27	86.13	614.17	177.68	762.54	388.67
19	IT-111	202.29	98.04	653.05	185.18	869.05	395.11
20	IT-112	206.07	78.52	519.05	173.72	671.11	388.90
21	IT-113	278.85	79.79	662.72	180.36	923.20	412.91
22	IT-114	248.15	83.17	678.88	187.51	961.73	402.71
23	IT-115	207.35	81.19	643.45	175.81	919.31	405.61
24	IT-116	181.80	93.74	512.81	185.70	811.17	380.91
25	IT-117	164.08	98.33	572.28	181.19	771.94	379.91
26	IT-118	171.41	93.74	550.65	191.77	745.67	405.49
27	IT-119	166.07	81.15	498.36	199.29	824.47	375.79
28	IT-120	210.98	83.80	513.55	194.76	815.36	390.66
29	IT-121	197.89	92.78	505.26	193.77	839.65	413.75
30	IT-122	207.00	80.96	528.20	183.04	809.48	400.20
	SE	37.48	5.23	44.43	6.56	42.99	7.67
	CD	NS	15.10	128.30	18.95	124.14	22.16

4.2.1.7 Leaf area

Leaf area differed significantly among the accessions at pre-flowering stage under open condition only (Table 11). In open condition, maximum leaf area (2928.48 cm²) was recorded by IT-108, followed by IT-96 (2890.03 m²). Leaf area (1721.30 cm²) was found to be the least in IT-122.

At flowering stage, there was significant difference among the accessions under shaded condition. Leaf area was found to be maximum (5418.64 cm²) in IT-105 under shade. It was on par with IT-98 (5414.12 cm²), IT-104 (5347.90 cm²) and IT-108 (5208.72 cm²). IT-93 recorded the lowest leaf area (3258.01 cm²).

At seed maturation stage, significant difference was observed among the accessions under open as well as shaded conditions. Under open condition, maximum leaf area (18211.30 cm²) was recorded by IT-108, followed by IT-96 (17987.69 cm²), IT-99 (17702.50 cm²) and IT-111 (17699.62 cm²), which were on par. It was found to be least (10285.37 cm²) in IT-94. In shade, IT-106 recorded the maximum leaf area (16002.77 cm²), which was on par with IT-104 (15953.13 cm²), IT-96 (15942.60 cm²) and IT-101 (15623.66 cm²). IT-94 recorded the least leaf area (7245.02 cm²).

Leaf area was more in all accessions at all the three stages of growth under open condition compared to shade and irrespective of the light condition, IT-108 and IT-96 recorded more leaf area compared to the other accessions.

4.2.1.8 Root length

Significant difference was observed among the accessions for root length under open as well as shaded condition for all the three stages of growth (Table 12).

Under open condition, at pre-flowering stage, maximum root length (33.78 cm) was recorded by IT-96. It was on par with IT-108 (33.01 cm), IT-105 (31.36 cm) and IT-106 (31.03 cm). Lowest root length (20.33 cm)

Table 11. Growth of *I. tinctoria* accessions under open and shaded conditions: leaf area (cm²)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 93	1596.90	1034.92	5467.58	3258.01	14230.93	10072.80
2	IT- 94	1862.00	962.60	4965.64	3310.88	10285.37	7245.02
3	IT- 95	2557.66	1245.68	5754.59	3303.88	14284.97	10726.67
4	IT- 96	2890.03	1332.26	6964.94	5140.38	17987.69	15942.60
5	IT- 97	1788.61	1239.34	6991.42	4422.74	15303.69	13551.97
6	IT- 98	2776.64	1368.56	6951.62	5414.12	16768.04	15365.91
7	IT- 99	2581.34	1160.39	6225.43	4483.23	17702.50	13399.25
8	IT-100	2437.22	1182.98	6069.42	4868.81	15314.40	11790.66
9	IT-101	2565.56	1499.58	7060.60	4818.27	13310.45	15623.66
10	IT-102	2172.47	1369.38	6888.22	3797.63	13459.46	10630.09
11	IT-103	2216.53	1336.64	7300.53	4540.25	13715.33	12203.42
12	IT-104	2110.37	1561.46	7025.40	5347.90	16170.27	15953.13
13	IT-105	1846.87	1529.21	7432.21	5418.64	16783.35	14880.25
14	IT-106	2155.28	1390.50	7367.47	5131.14	16884.52	16002.77
15	IT-107	2070.44	1067.14	6075.09	3753.12	16756.82	9873.22
16	IT-108	2928.48	1491.20	6753.11	5208.72	18211.30	14895.03
17	IT-109	1951.15	1124.31	5505.63	4055.82	15988.49	11281.23
18	IT-110	1854.56	1053.82	5643.92	4437.81	15620.32	11319.12
19	IT-111	1965.14	1308.20	5575.68	3988.31	17699.62	13739.60
20	IT-112	2203.56	1067.56	6001.15	3699.72	15020.69	12547.43
21	IT-113	2183.44	1163.55	6512.22	3697.88	14050.10	10866.49
22	IT-114	2187.82	1320.04	6807.79	4904.69	17649.56	14106.41
23	IT-115	2019.83	1253.35	6233.49	5058.74	15334.16	14166.10
24	IT-116	1976.87	1043.53	6272.35	4824.90	15438.18	12144.52
25	IT-117	1985.53	976.73	6340.89	4737.17	15136.58	13315.16
26	IT-118	1989.99	1178.70	6250.52	4771.61	15022.48	11772.28
27	IT-119	1748.54	896.48	6397.78	4660.88	15076.82	12094.01
28	IT-120	2148.86	1089.10	6136.74	4916.43	14447.58	11750.66
29	IT-121	1964.84	1212.49	6177.59	4075.38	14607.00	13134.49
30	IT-122	1721.30	1068.69	6222.97	4324.17	17136.22	11038.89
	SE	237.18	142.81	4806.79	204.87	1069.05	651.54
	CD	684.95	NS	NS	591.64	3087.23	1881.52

Table 12. Growth of *I. tinctoria* accessions under open and shaded conditions: root length (cm)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT-93	20.91	8.07	33.57	16.79	50.27	25.02
2	IT-94	21.23	9.92	37.80	16.67	46.52	30.15
3	IT-95	26.11	7.37	39.52	16.54	53.74	26.85
4	IT-96	33.78	10.23	49.84	22.01	71.25	33.95
5	IT-97	25.86	8.56	39.59	19.03	50.32	31.82
6	IT-98	20.11	18.55	35.34	21.19	55.87	33.97
7	IT-99	24.24	10.92	32.64	17.21	51.27	28.28
8	IT-100	25.31	9.55	35.07	16.85	56.12	27.18
9	IT-101	29.29	13.20	46.49	18.77	56.93	31.84
10	IT-102	20.33	8.88	36.27	19.55	54.21	28.18
11	IT-103	20.59	7.12	34.97	21.50	52.10	27.27
12	IT-104	25.75	11.59	47.51	23.20	53.04	32.68
13	IT-105	31.36	10.59	40.83	24.03	60.14	34.93
14	IT-106	31.03	12.75	42.01	19.12	61.11	32.73
15	IT-107	25.52	7.93	33.28	19.74	45.75	27.29
16	IT-108	33.01	11.52	38.62	25.92	47.39	27.55
17	IT-109	24.12	13.18	41.15	20.73	44.16	28.25
18	IT-110	21.44	9.25	40.12	18.83	44.27	29.02
19	IT-111	22.47	8.19	43.93	22.18	55.76	33.21
20	IT-112	24.85	7.22	38.17	18.05	48.03	28.15
21	IT-113	27.10	11.92	45.72	24.27	48.42	33.01
22	IT-114	28.81	11.70	38.28	22.90	49.00	33.14
23	IT-115	30.15	12.70	41.31	22.24	48.59	32.64
24	IT-116	24.22	8.04	32.79	19.11	46.34	28.69
25	IT-117	24.13	8.29	37.84	18.44	46.11	28.78
26	IT-118	21.72	8.70	42.76	18.53	46.13	26.57
27	IT-119	26.18	9.20	38.61	20.77	51.07	26.50
28	IT-120	25.32	8.73	40.22	19.72	46.41	29.18
29	IT-121	21.65	8.83	43.75	17.90	46.77	25.34
30	IT-122	21.44	9.34	34.81	18.99	45.01	27.74
	SE	1.52	0.79	2.49	1.38	3.54	0.95
	CD	4.39	2.28	7.18	3.97	10.24	2.76

was observed in IT-102. Under shade, IT-98 produced the longest root (18.55 cm) and IT-112 the shortest (7.22 cm).

At flowering stage, under open condition maximum root length (49.84 cm) was observed in IT- 96, followed by IT-104 (47.51 cm) and IT-101 (46.49 cm). IT-99 recorded the least (32.64 cm). In shade, maximum root length (25.92 cm) was recorded by IT-108, followed by IT-113 (24.27 cm) and IT-105 (24.03 cm). The lowest root length (16.54 cm) was observed in IT-95.

At seed maturation stage, under open condition root length was maximum for IT-96 (71.25 cm), which was on par with IT-106 (61.11 cm). Lowest root length was for IT-109 (44.16 cm). Under shade, IT-105 recorded the maximum root length (34.93 cm), followed by IT-98 (33.97 cm) and IT-96 (33.95 cm). IT-93 recorded the lowest root length (25.02 cm).

IT-96 recorded maximum root length under open condition during all the three stages of growth.

4.2.1.9 Root girth at collar region

At pre-flowering stage, significant difference was observed among the accessions under open condition only (Table 13). Maximum root girth (4.31 cm) was recorded by IT-101 in open condition, followed by IT-111 (4.19 cm), IT-108 (4.12 cm) and IT-115 (4.12 cm). Least value (2.91 cm) was observed in IT-94.

At flowering stage, significant difference was there among the accessions both under open and shaded conditions. Under open condition, root girth was maximum (5.21 cm) in IT-111, which was on par with IT-96 (5.19 cm) and IT-100 (5.13 cm). Root girth was minimum (3.61 cm) for IT-93. Under shade, IT-108 recorded the maximum root girth (2.96 cm) followed by IT-114 (2.88 cm) and IT-106 (2.69 cm). Lowest value (1.60 cm) was observed in IT-93.

At seed maturation stage, accessions varied significantly in root girth under both conditions of light. In open, IT-101 had the highest value

Table 13. Growth of *I. tinctoria* accessions under open and shaded conditions:
root girth at collar region(cm)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 93	3.05	1.06	3.61	1.60	7.75	2.98
2	IT- 94	2.91	1.11	4.20	1.86	7.99	3.06
3	IT- 95	3.10	1.18	4.41	1.92	8.40	3.00
4	IT- 96	3.73	1.46	5.19	2.51	11.33	4.13
5	IT- 97	3.30	1.30	4.61	2.08	9.56	3.15
6	IT- 98	3.48	1.11	4.83	2.42	8.38	3.90
7	IT- 99	4.02	1.04	4.84	2.06	8.87	3.41
8	IT-100	4.02	0.93	5.13	2.13	7.42	3.28
9	IT-101	4.31	1.19	5.05	2.25	13.26	3.80
10	IT-102	3.71	1.15	4.71	2.00	9.23	3.27
11	IT-103	3.91	0.97	4.95	1.94	9.02	3.34
12	IT-104	3.98	1.51	4.99	2.35	9.26	3.68
13	IT-105	3.96	1.34	4.93	2.64	9.54	3.83
14	IT-106	3.95	1.49	4.90	2.69	8.95	3.86
15	IT-107	3.88	1.15	4.63	2.11	8.10	3.48
16	IT-108	4.12	1.52	4.84	2.96	11.25	4.08
17	IT-109	3.84	1.18	4.78	1.93	7.02	3.68
18	IT-110	3.70	1.10	5.06	1.88	7.45	3.38
19	IT-111	4.19	1.45	5.21	2.59	10.83	3.48
20	IT-112	3.62	1.30	4.60	2.10	9.10	3.31
21	IT-113	3.77	1.35	4.76	2.69	11.99	3.96
22	IT-114	3.88	1.20	4.68	2.88	11.75	4.06
23	IT-115	4.12	1.40	4.96	2.43	12.16	3.86
24	IT-116	3.74	1.10	4.33	2.23	8.77	3.37
25	IT-117	3.85	1.09	4.09	2.03	9.10	3.12
26	IT-118	3.39	0.97	4.13	2.26	9.49	3.08
27	IT-119	3.16	1.27	4.39	2.20	9.68	3.25
28	IT-120	3.27	1.18	4.21	2.38	8.88	3.40
29	IT-121	3.24	1.15	4.38	2.32	7.87	3.09
30	IT-122	3.29	1.16	4.58	2.00	8.46	3.27
	SE	0.17	0.13	0.25	0.11	0.75	0.09
	CD	0.49	NS	0.73	0.31	2.17	0.25

of 13.26 cm, followed by IT-115 (12.16 cm). The lowest was recorded in IT-109 (7.02 cm). Under shade, IT-96 recorded the maximum root girth of 4.13 cm followed by IT-108 (4.08 cm) and IT-114 (4.06 cm). IT-93 recorded the lowest value for root girth (2.98 cm). All accessions recorded more than double the root girth under shade, in open condition.

4.2.2 Yield attributes

4.2.2.1 Fresh weight of shoot

Pooled analysis indicated that there was significant difference in shoot fresh weight between shaded and open conditions at all stages of growth (Table 14).

At pre-flowering stage, accessions showed significant difference among them only under shaded condition. In shade, IT-98 recorded the maximum shoot fresh weight (128.5 g). It was on par with IT-101 (122.5 g), IT-99 (115 g) and IT-108 (107.5 g). IT-93 recorded the lowest shoot fresh weight (36 g).

At flowering stage, there was significant difference among the accessions under both conditions. Maximum pooled mean for shoot fresh weight (937.5 g) was observed in IT-104, which was on par with IT-108 (898.3 g), IT-106 (887.5 g) and IT-114 (837.5 g). Pooled mean was minimum (156.25 g) for IT-94.

At seed maturation stage also, there was significant difference between the accessions under both open and shaded condition. IT-101 recorded maximum pooled mean (3087.5 g) for shoot fresh weight followed by IT-108 (2737.5 g), IT-106 (2477.5 g) and IT-96 (2262.5 g). IT-94 recorded the minimum pooled mean (323.75 g).

IT-108 showed good performance in terms of shoot fresh weight at all the three stages of growth.

Table 14. Yield of *I. tinctoria* accessions under open and shaded conditions: fresh weight of shoot (g)

Sl. No	Accession No.	Stages of plant growth							
		Pre-flowering (90 DAS)		Flowering (150 DAS)			Seed maturation (240 DAS)		
		Open	Shade	Open	Shade	Mean	Open	Shade	Mean
1	IT-93	65.00	36.00	227.50	90.00	158.80	637.50	425.00	531.25
2	IT-94	67.00	41.50	225.00	87.50	156.30	392.50	255.00	323.75
3	IT-95	113.50	97.50	212.50	132.50	172.50	472.50	430.00	451.25
4	IT-96	157.00	100.00	980.00	690.00	835.00	2675.00	1850.00	2262.50
5	IT-97	109.00	95.00	502.50	377.50	440.00	905.00	712.50	808.75
6	IT-98	155.00	128.50	605.00	510.00	557.50	782.50	617.50	700.00
7	IT-99	147.50	115.00	605.00	496.50	550.80	1272.50	710.00	991.25
8	IT-100	115.00	97.50	690.00	427.50	558.80	1525.00	610.00	1067.50
9	IT-101	125.00	122.50	900.00	610.00	755.00	4400.00	1775.00	3087.50
10	IT-102	82.50	62.50	557.50	280.00	418.80	1017.50	412.50	715.00
11	IT-103	122.50	58.50	615.00	395.00	505.00	1695.00	852.50	1273.75
12	IT-104	128.50	90.00	1250.00	625.00	937.50	2070.00	1692.50	1881.25
13	IT-105	92.50	94.00	437.50	668.50	553.00	2162.50	1547.50	1855.00
14	IT-106	110.00	97.50	1062.50	712.50	887.50	2975.00	1980.00	2477.50
15	IT-107	80.00	51.50	332.50	132.50	232.50	750.00	442.50	596.25
16	IT-108	220.00	107.50	1147.50	649.00	898.30	3485.00	1990.00	2737.50
17	IT-109	116.50	74.00	720.00	332.50	526.30	985.00	415.00	700.00
18	IT-110	67.50	82.50	796.50	508.00	652.30	915.00	705.00	810.00
19	IT-111	147.50	95.00	885.00	620.00	752.50	2145.00	995.00	1570.00
20	IT-112	85.00	80.00	590.00	371.50	480.80	1497.50	570.00	1033.75
21	IT-113	134.00	85.00	642.50	380.00	511.30	990.00	692.50	841.25
22	IT-114	189.00	106.00	987.50	687.50	837.50	2112.50	2025.00	2068.75
23	IT-115	79.00	51.50	725.00	327.50	526.30	1220.00	765.00	992.50
24	IT-116	72.50	71.00	417.50	374.00	395.80	705.00	762.50	733.75
25	IT-117	67.50	82.50	432.50	422.50	427.50	772.50	757.50	765.00
26	IT-118	65.00	71.50	442.50	430.00	436.30	915.00	797.50	856.25
27	IT-119	142.00	71.00	352.50	332.50	342.50	652.50	750.00	701.25
28	IT-120	107.50	81.00	267.50	430.00	348.08	490.00	682.50	586.25
29	IT-121	100.00	90.50	352.50	372.50	362.50	525.00	765.00	645.00
30	IT-122	79.00	63.50	420.00	267.50	343.80	650.00	520.00	585.00
	Mean	111.40	83.30	612.70	424.70	-	1393.08	916.83	-
	SE	28.92	11.56	-	-	-	-	-	-
	CD	NS	33.38	-	-	-	-	-	-
Pooled analysis				SE	CD	SE	CD		
For light condition				23.47	66.37	75.66	213.99		
For accessions				90.88	257.05	293.02	828.79		
For interaction				128.53	363.53	414.39	1172.09		

4.2.2.2 Dry weight of shoot

Pooled analysis indicated that there was significant difference in shoot dry weight between open and shaded conditions at all stages of growth (Table 15).

At pre-flowering stage, there was significant difference among the accessions under open as well as shaded conditions. Under open condition, the highest value for shoot dry weight (76.5 g) was recorded by IT-108, which was on par with IT-114 (61.5 g) and the lowest value (18.5 g) by IT-118. In shade, IT-98 (36 g) recorded the maximum shoot fresh weight followed by IT-101 (33.5 g) and IT-99 (32 g). Minimum value (12 g) was recorded by IT-93.

At flowering stage also, accessions showed significant difference under both the conditions. Maximum pooled mean for shoot dry weight (248.75 g) was observed in IT-106, which was on par with IT-104 (245.5 g), IT-108 (241.5 g) and IT-114 (233.5 g). Minimum pooled mean (44.5 g) was observed in IT-94.

At seed maturation stage also, accessions showed significant difference under both the conditions. IT-108 recorded the maximum pooled mean (556.5 g) followed by IT-101 (507.50 g), IT-106 (476.00 g) and IT-114 (434.00 g). Minimum value (95 g) was recorded by IT-94.

IT-108 and IT-106 performed better compared to other accessions at all stages of growth.

4.2.2.3 Fresh weight of leaves

Pooled analysis indicated that there was significant difference in fresh weight of leaves between open and shaded conditions at all the three stages of growth (Table 16).

At pre-flowering stage, accessions showed significant difference among them only under shaded conditions. In shade, IT-101 recorded the maximum fresh weight of leaves (71.32 g). It was on par with IT-99 (67.62 g).

Table 15. Yield of *I. tinctoria* accessions under open and shaded conditions: dry weight of shoot (g)

Sl. No	Accession No.	Stages of plant growth							
		Pre-flowering (90 DAS)		Flowering (150 DAS)			Seed maturation (240 DAS)		
		Open	Shade	Open	Shade	Mean	Open	Shade	Mean
1	IT-93	21.50	12.00	66.00	26.50	46.25	177.50	110.50	144.00
2	IT-94	22.00	14.50	66.50	22.50	44.50	119.00	71.00	95.00
3	IT-95	32.50	24.00	63.00	41.50	52.25	130.00	115.00	122.50
4	IT-96	47.50	28.50	248.00	190.00	219.00	455.00	385.00	420.00
5	IT-97	36.50	22.50	131.50	103.00	117.25	276.50	188.50	232.50
6	IT-98	47.50	36.00	178.00	127.50	152.75	244.00	172.50	280.25
7	IT-99	43.00	32.00	165.00	128.00	146.50	320.00	186.00	253.00
8	IT-100	32.50	24.50	197.50	113.00	155.25	365.00	169.00	267.25
9	IT-101	37.00	33.50	255.50	158.00	206.75	642.00	373.00	507.50
10	IT-102	23.00	17.50	140.00	82.50	111.25	273.00	118.00	195.50
11	IT-103	36.50	17.50	171.50	107.50	139.50	377.50	243.00	310.25
12	IT-104	39.50	29.50	326.00	165.00	245.50	455.00	373.50	414.30
13	IT-105	28.00	30.00	115.00	180.00	147.50	437.50	353.50	395.50
14	IT-106	34.50	30.00	297.50	200.00	248.75	548.50	403.50	476.00
15	IT-107	27.00	15.00	84.50	41.00	62.75	204.00	129.50	166.75
16	IT-108	76.50	29.50	305.00	178.00	241.50	715.00	398.00	556.50
17	IT-109	32.40	22.50	188.50	90.00	139.25	280.00	119.00	199.50
18	IT-110	21.00	21.50	205.00	136.00	170.50	249.00	188.50	218.80
19	IT-111	41.50	30.00	245.00	163.00	204.00	497.50	288.00	392.75
20	IT-112	24.50	21.50	158.50	97.00	127.75	362.50	141.50	252.00
21	IT-113	41.50	20.50	177.50	103.50	140.50	290.00	197.00	243.50
22	IT-114	61.50	31.00	275.00	192.00	233.50	465.00	403.00	434.00
23	IT-115	25.00	16.00	188.00	88.00	138.00	310.00	189.50	249.75
24	IT-116	23.50	19.00	107.50	100.50	104.00	188.50	197.50	193.00
25	IT-117	20.50	21.50	114.00	111.00	112.50	212.50	204.00	208.25
26	IT-118	18.50	23.50	118.50	108.00	113.25	256.00	208.00	232.00
27	IT-119	38.50	21.00	101.50	99.00	100.25	170.50	207.00	188.75
28	IT-120	30.00	25.50	78.00	106.00	92.00	137.00	192.00	164.50
29	IT-121	29.00	29.50	98.50	101.50	100.00	149.00	198.00	173.50
30	IT-122	24.00	18.50	104.00	79.50	91.75	181.00	145.00	163.00
	Mean	33.80	23.90	165.70	114.60	-	316.20	222.30	-
	SE	7.35	3.68	-	-	-	-	-	-
	CD	21.24	10.63	-	-	-	-	-	-
Pooled analysis				SE	CD	SE	CD		
For light condition				6.09	17.23	11.52	32.60		
For accessions				23.59	66.72	44.64	126.20		
For interaction				33.36	94.36	63.13	178.50		

Table 16. Yield of *I. tinctoria* accessions under open and shaded conditions: fresh weight of leaves (g)

Sl. No	Accession No.	Stages of plant growth							
		Pre-flowering (90 DAS)		Flowering (150 DAS)			Seed maturation (240 DAS)		
		Open	Shade	Open	Shade	Mean	Open	Shade	Mean
1	IT-93	34.77	21.30	117.80	48.08	82.94	216.89	143.07	179.98
2	IT-94	36.21	25.92	122.17	46.94	84.55	126.21	90.50	108.36
3	IT-95	58.86	60.63	115.61	70.38	92.99	158.74	154.08	156.41
4	IT-96	81.84	57.76	512.79	352.98	432.89	958.38	675.27	816.82
5	IT-97	59.62	53.62	266.31	199.38	232.84	314.12	254.02	284.07
6	IT-98	82.21	80.17	322.40	262.51	292.45	271.38	214.58	242.98
7	IT-99	76.57	67.62	341.89	261.95	301.92	457.90	249.27	353.58
8	IT-100	62.72	58.82	368.73	220.73	294.73	538.99	212.73	375.86
9	IT-101	68.51	71.32	499.26	332.82	416.04	1572.90	639.85	1106.37
10	IT-102	44.98	38.50	301.53	157.28	229.41	360.89	141.25	251.07
11	IT-103	67.11	38.59	332.27	212.83	272.55	600.83	308.43	454.63
12	IT-104	67.62	54.38	690.58	327.55	509.07	736.44	619.75	678.09
13	IT-105	52.70	51.96	219.45	373.08	296.27	773.14	570.35	671.74
14	IT-106	59.50	58.28	589.07	377.46	483.26	1086.08	725.02	905.55
15	IT-107	50.40	33.22	174.90	73.59	124.24	256.94	145.62	201.28
16	IT-108	113.04	63.42	602.25	343.62	472.93	1271.98	727.02	999.50
17	IT-109	64.33	44.57	371.14	181.77	276.45	341.59	226.06	283.83
18	IT-110	38.60	48.74	412.90	272.63	342.76	322.13	239.41	280.77
19	IT-111	84.19	54.00	478.36	325.99	402.18	773.55	375.84	574.69
20	IT-112	50.64	50.70	325.64	201.05	263.35	530.13	207.45	368.79
21	IT-113	72.69	51.60	339.57	207.20	273.38	348.14	237.77	292.96
22	IT-114	104.68	60.80	541.57	360.69	451.13	758.52	747.46	752.99
23	IT-115	46.18	29.58	389.42	185.33	287.37	428.73	259.77	344.25
24	IT-116	41.69	43.32	224.50	204.61	214.56	242.08	274.35	258.21
25	IT-117	37.76	48.53	230.37	243.50	236.94	268.78	268.98	268.88
26	IT-118	37.88	41.67	230.41	237.35	233.88	284.68	284.99	284.84
27	IT-119	77.61	41.54	186.74	187.50	187.12	221.91	265.26	243.39
28	IT-120	56.65	48.47	141.03	236.83	188.93	171.00	239.68	205.34
29	IT-121	55.33	49.75	186.21	203.67	194.94	184.45	264.15	224.30
30	IT-122	42.69	37.82	221.81	152.71	187.26	234.61	180.23	207.42
	Mean	61.08	49.55	328.55	228.73	-	493.74	331.41	-
	SE	15.07	6.16	-	-	-	-	-	-
	CD	NS	17.80	-	-	-	-	-	-
Pooled analysis				SE	CD	SE	CD		
For light condition				13.49	38.16	27.25	77.08		
For accessions				52.25	147.78	105.55	298.53		
For interaction				73.89	208.99	149.27	422.19		

IT-108 (63.42 g), IT-114 (60.80 g) and IT-95 (60.63 g). IT-93 recorded the lowest value (21.30) for leaf fresh weight.

At flowering stage, there was significant difference among the accessions under open as well as shaded conditions. Maximum pooled mean for fresh weight of leaves (509.07 g) was observed in IT-104. It was on par with IT-106 (483.26 g), IT-108 (472.93 g), IT-114 (451.13 g), IT-96 (432.89 g) and IT-101 (416.04 g). Pooled mean was minimum (82.94 g) for IT-93.

At seed maturation stage also, there was significant difference among the accessions under both open and shaded conditions. IT-101 recorded the maximum pooled mean (1106.37 g) for shoot fresh weight followed by IT-108 (999.50 g), IT-106 (905.55 g) and IT-96 (816.82 g).

Fresh weight of leaves was higher in IT-108 and IT-101 at all the three stages of growth compared to the other accessions.

4.2.2.4 Dry weight of leaves

Pooled analysis showed that there was significant difference in dry weight of leaves between open and shaded conditions at all stages of growth (Table 17).

At pre-flowering stage, there was significant difference among the accessions only under shaded conditions. Under shaded condition, the highest value for dry weight of leaves (25.83 g) was observed in IT-98 followed by IT-101 (23.28 g), IT-99 (21.8 g) and IT-108 (20.75 g). The lowest value (6.92 g) was recorded by IT-93.

At flowering stage, there was significant difference among the accessions under open and shaded conditions. Maximum pooled mean for dry weight of leaves (165.57 g) was observed in IT-104, which was on par with IT-106 (159.41 g), IT-108 (153.74 g) and IT-114 (147.50 g). Pooled mean was minimum (27.62 g) for IT-94.

At seed maturation stage also, there was significant difference between the accessions under both open and shaded condition. Pooled mean

Table 17. Yield of *I. tinctoria* accessions under open and shaded conditions: dry weight of leaves (g)

Sl. No	Accession No.	Stages of plant growth							
		Pre-flowering (90 DAS)		Flowering (150 DAS)			Seed maturation (240 DAS)		
		Open	Shade	Open	Shade	Mean	Open	Shade	Mean
1	IT-93	13.34	6.92	41.01	16.05	28.53	75.12	47.83	61.48
2	IT-94	13.22	7.86	40.68	14.56	27.62	41.35	39.40	40.38
3	IT-95	20.32	19.10	38.72	22.40	30.56	51.87	50.95	51.41
4	IT-96	27.51	18.22	162.74	112.26	137.50	284.13	225.10	254.61
5	IT-97	19.14	17.58	95.60	64.35	79.97	99.95	83.92	91.93
6	IT-98	27.64	25.83	104.42	82.85	93.63	85.13	68.38	76.75
7	IT-99	25.12	21.80	108.67	85.26	96.96	147.92	79.53	113.72
8	IT-100	20.97	19.44	120.42	71.55	95.98	178.72	67.13	122.93
9	IT-101	25.24	23.28	156.83	107.71	132.27	512.99	206.37	359.68
10	IT-102	16.56	12.30	97.76	53.21	75.49	121.41	45.99	83.70
11	IT-103	23.95	12.08	108.71	67.19	87.95	196.98	100.93	148.95
12	IT-104	24.60	17.98	224.07	107.08	165.57	238.33	205.59	221.96
13	IT-105	17.45	16.58	71.57	121.03	96.30	254.64	190.20	222.42
14	IT-106	21.24	17.84	196.36	122.45	159.41	349.87	237.07	293.47
15	IT-107	17.18	10.16	59.21	24.47	41.84	85.11	47.83	66.47
16	IT-108	38.70	20.75	195.76	111.73	153.74	385.25	228.62	306.94
17	IT-109	23.25	14.48	120.89	58.99	89.94	107.19	72.89	90.04
18	IT-110	13.19	15.44	133.75	89.22	111.48	101.73	76.32	89.03
19	IT-111	28.83	17.29	156.39	106.40	131.39	260.96	120.64	190.80
20	IT-112	19.43	15.79	101.97	63.70	82.83	164.16	63.38	113.77
21	IT-113	25.50	16.68	113.92	64.96	89.44	111.29	77.34	94.31
22	IT-114	34.57	18.37	181.67	113.34	147.50	248.97	246.86	247.91
23	IT-115	15.55	9.21	122.77	58.89	90.83	138.18	85.19	111.69
24	IT-116	13.17	13.22	75.96	56.07	71.01	74.29	87.32	80.80
25	IT-117	13.08	15.48	74.70	77.33	76.01	90.99	90.64	90.81
26	IT-118	13.21	14.10	79.63	76.78	78.20	89.68	88.58	89.12
27	IT-119	27.58	12.43	59.28	59.59	59.43	71.73	82.91	77.32
28	IT-120	19.24	15.10	46.78	77.75	62.27	57.17	80.78	68.98
29	IT-121	17.99	15.94	62.23	66.51	64.37	58.90	88.39	73.64
30	IT-122	13.98	12.75	73.28	49.33	61.30	75.91	59.88	67.89
	Mean	20.99	15.80	107.52	73.77	-	158.66	107.86	-
	SE	5.61	2.19	-	-	-	-	-	-
	CD	NS	6.32	-	-	-	-	-	-
Pooled analysis						SE	CD	SE	CD
For light condition						3.69	10.38	8.81	24.92
For accessions						14.28	40.20	34.12	96.51
For interaction						20.20	56.85	48.25	136.48

for leaf dry weight (359.68 g) was maximum in IT-101. It was on par with IT-108 (306.94 g) and IT-106 (293.47 g). Minimum pooled mean (40.38 g) was observed for IT-94.

IT-108 and IT-101 showed better performance compared to the other accessions.

4.2.2.5 Fresh weight of pods

Based on pooled analysis, significant difference could be observed among the accessions in fresh weight of pods (Table 18). There was significant difference among the accessions under both open and shaded conditions as well. Pooled mean was maximum (717.69 g) for IT-101, which was on par with IT-108 (698.82 g) and IT-106 (631.58 g). Minimum pooled mean (86.80 g) was recorded by IT-94.

4.2.2.6 Dry weight of pods

Pooled analysis showed significant difference among the accessions with regard to pod dry weight (Table 18). There was significant difference among the accessions as well under open and shade. Maximum pooled mean (226.45 g) was recorded by IT-101 followed by IT-108 (192.15 g), IT-96 (183.44 g) and IT-106 (182.71 g).

4.2.2.7 Fresh weight of root

Pooled analysis indicated that there was significant difference in fresh weight of root between open and shaded conditions at all the three stages of growth (Table 19).

At pre-flowering stage, there was significant difference among the accessions only under shaded condition. In shade, IT-106 recorded the maximum root fresh weight (7.92 g). It was on par with IT-111 (7.90 g), IT-110 (7.49 g) and IT-104 (7.49 g). IT-96 recorded the minimum (4.04 g).

At flowering stage also, there was significant difference among the accessions, but only under shaded condition. In shaded condition, IT-106

Table 18 Yield of *I. tinctoria* accessions at seed maturation stage (240 DAS) under open and shaded conditions: fresh and dry weight of pods (g)

Sl. No.	Accession No.	Pod fresh weight (g)			Pod dry weight (g)		
		Open	Shade	Mean	Open	Shade	Mean
1	IT-93	172.30	102.78	137.54	44.40	31.67	38.03
2	IT-94	110.29	63.32	86.80	33.67	20.21	26.94
3	IT-95	129.94	106.29	118.12	41.46	29.70	35.58
4	IT-96	729.41	453.45	591.43	225.37	141.52	183.44
5	IT-97	255.93	172.23	214.08	75.08	50.40	62.74
6	IT-98	204.05	151.75	177.90	63.06	40.16	51.61
7	IT-99	326.30	168.61	247.45	106.39	47.26	76.83
8	IT-100	398.06	147.38	272.72	104.94	39.07	72.00
9	IT-101	980.20	455.18	717.69	334.27	118.63	226.45
10	IT-102	272.68	102.50	187.59	77.05	31.15	54.10
11	IT-103	440.82	208.13	324.48	123.32	64.48	93.90
12	IT-104	559.27	430.30	494.78	134.31	127.03	130.67
13	IT-105	571.97	370.81	471.39	167.18	103.31	135.24
14	IT-106	768.57	494.60	631.58	220.41	145.02	182.71
15	IT-107	215.44	110.13	162.78	64.79	31.78	48.28
16	IT-108	899.28	498.35	698.82	247.78	136.52	192.15
17	IT-109	271.74	104.95	188.34	74.44	32.19	53.31
18	IT-110	240.92	177.17	209.04	61.72	51.81	56.76
19	IT-111	563.00	208.64	385.82	160.84	60.75	110.79
20	IT-112	389.95	139.62	264.78	98.99	39.44	69.21
21	IT-113	263.11	173.11	218.11	63.54	52.59	58.07
22	IT-114	556.75	507.15	531.95	137.91	151.30	144.60
23	IT-115	317.73	190.92	254.32	80.34	57.02	68.68
24	IT-116	183.17	192.79	187.98	55.77	55.32	55.55
25	IT-117	212.54	189.29	200.92	54.33	56.16	55.25
26	IT-118	280.23	196.21	238.22	85.82	57.83	71.83
27	IT-119	169.23	191.33	180.28	42.35	59.50	50.92
28	IT-120	127.40	172.96	150.18	32.02	49.99	41.00
29	IT-121	136.10	194.91	165.50	35.38	56.36	45.87
30	IT-122	168.84	130.08	149.46	42.56	37.28	39.92
	Mean	370.51	226.83	-	102.98	65.85	-
Pooled analysis			SE	CD		SE	CD
For light conditions			20.58	58.20		60.28	17.76
For accessions			79.69	225.40		24.32	68.80
For interaction			112.70	318.76		34.40	97.29

Table 19. Yield of *I. tinctoria* accessions under open and shaded conditions: fresh weight of root (g)

Sl. No.	Accession No.	Stages of plant growth						
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)		
		Open	Shade	Open	Shade	Open	Shade	Mean
1	IT-93	13.05	4.58	38.40	10.74	76.30	24.28	50.29
2	IT-94	9.80	4.99	33.55	12.90	70.45	28.85	49.65
3	IT-95	14.40	4.87	33.50	11.07	79.90	32.81	56.35
4	IT-96	19.15	4.04	40.65	17.01	125.85	38.09	81.97
5	IT-97	11.55	3.86	35.60	13.96	102.70	38.08	70.39
6	IT-98	20.95	6.90	26.85	12.28	94.25	43.74	69.00
7	IT-99	17.70	5.30	34.45	11.27	83.35	32.63	57.99
8	IT100	19.60	5.16	25.45	10.66	94.25	31.23	62.74
9	IT-101	15.75	6.57	38.00	13.55	134.10	39.18	86.64
10	IT-102	14.00	5.29	31.50	10.71	110.40	33.09	71.74
11	IT-103	28.05	5.41	30.20	13.19	83.10	36.00	59.55
12	IT-104	27.10	7.49	50.45	15.41	135.30	39.62	87.46
13	IT-105	20.65	8.28	33.05	16.92	150.45	43.77	97.11
14	IT-106	22.25	7.92	48.70	18.24	147.95	38.51	93.23
15	IT-107	18.15	6.92	27.85	13.28	75.75	31.48	53.61
16	IT-108	26.15	5.76	52.55	13.05	228.95	42.07	135.51
17	IT-109	18.80	6.28	26.30	11.48	94.85	36.03	65.44
18	IT-110	16.45	7.49	26.45	12.58	71.00	32.08	51.54
19	IT-111	20.95	7.90	46.60	16.73	141.75	40.45	91.10
20	IT-112	20.20	6.81	27.65	13.09	95.05	34.09	64.57
21	IT-113	20.65	7.31	22.95	16.03	224.80	34.84	129.82
22	IT-114	26.25	6.71	38.80	16.09	150.25	35.04	92.64
23	IT-115	18.95	5.38	29.85	15.48	183.85	36.33	110.09
24	IT-116	20.75	6.28	30.15	12.18	80.85	30.91	55.88
25	IT-117	17.40	6.79	24.90	13.00	87.15	33.57	60.36
26	IT-118	20.00	6.32	25.75	12.78	91.85	37.25	64.55
27	IT-119	17.55	6.04	25.55	13.18	88.95	34.36	61.65
28	IT-120	23.60	6.03	30.70	13.61	78.50	34.18	56.34
29	IT-121	15.00	5.98	23.95	12.26	84.40	35.78	60.09
30	IT-122	14.80	5.80	26.30	13.32	91.05	36.88	63.96
	Mean	18.99	6.15	32.89	13.53	111.91	35.51	-
	SE	5.64	0.39	6.13	0.88	-	-	-
	CD	NS	1.14	NS	2.53	-	-	-
Pooled analysis						SE	CD	
For light condition						5.24	14.82	
For accessions						20.29	57.39	
For interaction						28.70	81.17	

recorded the maximum value for fresh weight of roots (18.24 g) followed by IT-96 (17.01 g), IT-105 (16.92 g) and IT-111 (16.73 g). IT-100 recorded the minimum (10.66 g).

At seed maturation stage, there was significant difference among the accessions under both open and shaded conditions. Maximum pooled mean for fresh weight of roots (135.51 g) was observed in IT-108. It was on par with IT-113 (129.82 g), IT- 115 (110.09 g) and IT-105 (97.11 g). IT-94 recorded the minimum (49.65 g).

4.2.2.8 Dry weight of root

Pooled analysis indicated that there was significant difference in shoot dry weight between open and shaded conditions at all stages of growth (Table 20).

At pre-flowering stage, accessions showed significant difference among them only under shaded condition. IT-105 recorded the maximum root dry weight (3.92 g) under shade, followed by IT-104 (3.83 g), IT-106 (3.83 g), IT-110 (3.82 g) and IT-111 (3.8 g). IT-97 recorded the minimum root dry weight (2 g).

At flowering stage, there was significant difference among the accessions under open and shaded conditions. Pooled mean was maximum (17 g) for IT-106. It was on par with IT-104 (16.64 g), IT-108 (16.51 g) and IT-111 (16.51 g). Pooled mean was minimum (7.13 g) for IT-103.

At seed maturation stage also, there was significant difference between the accessions under both open and shaded conditions. IT-108 recorded the maximum pooled mean for root dry weight (66.61 g) followed by IT-113 (64.25 g), IT-114 (58.86 g) and IT-115 (57.20 g). IT-110 recorded the minimum pooled mean (25.77 g).

IT-106, IT-108 and IT-104 showed better performance in terms of root dry weight at all stages of growth, irrespective of the light condition.

Table 20. Yield of *I. tinctoria* accessions under open and shaded conditions: dry weight of roots (g)

Sl. No.	Accession No.	Stages of plant growth							
		Pre-flowering (90 DAS)		Flowering (150 DAS)			Seed maturation (240 DAS)		
		Open	Shade	Open	Shade	Mean	Open	Shade	Mean
1	IT-93	5.18	2.08	17.99	5.52	11.75	40.49	12.59	26.54
2	IT-94	4.50	2.59	16.57	5.95	11.26	36.03	16.92	26.47
3	IT-95	7.00	2.59	15.32	5.20	10.25	40.47	14.01	27.24
4	IT-96	8.22	2.02	20.30	8.33	14.31	65.14	18.99	42.07
5	IT-97	5.18	2.00	18.06	7.14	12.51	51.96	18.61	35.38
6	IT-98	9.82	3.78	13.65	6.48	10.06	47.42	21.77	34.50
7	IT-99	8.29	2.60	18.20	5.25	11.72	42.89	16.49	29.69
8	IT-100	8.96	2.37	12.37	5.25	8.81	48.55	15.62	32.08
9	IT-101	7.28	3.73	20.77	7.20	13.98	67.91	19.49	43.70
10	IT-102	7.00	2.56	16.76	5.28	11.02	56.10	16.81	36.45
11	IT-103	12.99	2.63	7.78	6.49	7.13	42.78	15.73	29.26
12	IT-104	13.61	3.83	26.22	7.06	16.64	70.71	19.82	45.26
13	IT-105	10.16	3.92	16.44	8.37	12.40	75.43	21.45	48.44
14	IT-106	9.23	3.83	24.70	9.31	17.00	74.58	18.72	46.65
15	IT-107	7.98	3.27	14.57	6.72	10.54	40.00	15.74	27.87
16	IT-108	12.95	2.63	26.83	6.19	16.51	113.23	20.00	66.61
17	IT-109	8.79	2.72	14.11	5.31	9.71	48.26	17.83	33.04
18	IT-110	9.03	3.82	14.15	6.34	10.24	35.67	15.87	25.77
19	IT-111	10.09	3.80	24.10	8.92	16.51	73.05	20.49	46.77
20	IT-112	10.13	2.75	14.24	6.46	10.35	46.93	16.78	31.85
21	IT-113	10.81	3.74	11.18	7.58	9.38	111.63	16.88	64.25
22	IT-114	13.17	3.26	19.45	8.29	13.87	100.51	17.20	58.86
23	IT-115	9.63	2.49	16.07	7.95	12.01	95.29	19.11	57.20
24	IT-116	10.16	3.71	14.90	5.82	10.36	39.63	14.76	27.19
25	IT-117	8.44	3.73	12.54	6.43	9.48	42.44	16.98	29.71
26	IT-118	10.02	3.20	13.26	6.03	9.64	45.16	18.15	31.66
27	IT-119	9.00	3.25	12.83	6.34	9.58	44.57	16.77	30.67
28	IT-120	12.23	2.99	15.23	7.05	11.14	39.50	16.82	28.16
29	IT-121	7.53	2.62	11.94	6.37	9.15	42.65	17.88	30.26
30	IT-122	7.05	2.61	12.86	6.54	9.70	46.34	18.04	32.19
	Mean	9.15	3.04	16.44	6.70	-	57.51	17.54	-
	SE	2.84	0.28	-	-	-	-	-	-
	CD	NS	0.81	-	-	-	-	-	-
Pooled analysis					SE	CD		SE	CD
For light condition					0.56	1.58		2.79	7.89
For accessions					2.17	6.11		10.80	30.56
For interaction					3.07	8.53		15.28	43.21

4.2.3 Glycoside estimation

Glycoside content in all the accessions as influenced by different leaf treatments, accessions and growing conditions are presented in Table 21.

Considering L x S x V interaction, there was significant difference among the different leaf treatments and between the different varieties (Table 21). Fresh leaves of IT-99 recorded the maximum glycoside content (0.57%). In S x V interaction (Table 22), maximum glycoside content (0.24%) was obtained in IT-99, under open condition and minimum (0.08%) in IT-118 under shaded condition. For L x S interaction, glycoside content was highest for fresh leaves under open condition. Among the accessions, the best accession producing more amount of glycoside (0.21%) was IT-99. All the accessions produced higher glycoside content in open condition compared to shade. Among the different leaf treatments, glycoside content was highest in fresh leaves.

4.2.4 Selection index

Discriminant function technique was adopted for the construction of selection index based on leaf yield and glycoside content. The index value for each accession was determined and the accessions were ranked accordingly. The selection index scores are presented in Table 23 along with the ranking of each accession.

IT-108 ranked first followed by IT-101 and IT-96. Out of the thirty accessions, top ranking ten accessions of the bluish green leaved variety of *Indigofera tinctoria*, which produced high leaf yield and more glycoside content, were selected for the phase-II study. The selected accessions were IT-108, IT-101, IT-96, IT-114, IT-106, IT-104, IT-105, IT-111, IT-97 and IT-99.

4.3. PHASE II EXPERIMENT- CULTURAL TRIAL OF SELECTED ACCESSIONS

The ten selected accessions from the Phase I experiment were raised in randomized block design in open and under shade with three

Table 21. Leaf glycoside content (%) in *I. tinctoria* accessions as influenced by leaf pre- treatments and open and shaded conditions

Sl No	Accession No.	Leaf treatments											
		Sun dried leaves			Oven dried leaves			Fresh leaves			Fermented leaves		
		O	S	M	O	S	M	O	S	M	O	S	M
1	IT-93	0.13	0.10	0.12	0.06	0.06	0.06	0.48	0.33	0.41	0.07	0.07	0.07
2	IT-94	0.13	0.12	0.13	0.05	0.05	0.05	0.49	0.33	0.41	0.06	0.06	0.06
3	IT-95	0.08	0.08	0.08	0.08	0.08	0.08	0.33	0.22	0.28	0.09	0.09	0.09
4	IT-96	0.14	0.13	0.14	0.05	0.05	0.05	0.58	0.45	0.51	0.06	0.05	0.06
5	IT-97	0.10	0.09	0.10	0.05	0.05	0.05	0.41	0.31	0.36	0.07	0.06	0.06
6	IT-98	0.14	0.11	0.13	0.06	0.06	0.06	0.51	0.41	0.46	0.08	0.06	0.07
7	IT-99	0.18	0.14	0.16	0.05	0.04	0.05	0.67	0.47	0.57	0.06	0.05	0.06
8	IT-100	0.09	0.08	0.09	0.04	0.05	0.05	0.38	0.25	0.31	0.06	0.05	0.05
9	IT-101	0.11	0.10	0.11	0.06	0.06	0.06	0.41	0.26	0.33	0.07	0.06	0.07
10	IT102	0.11	0.10	0.11	0.09	0.08	0.09	0.45	0.31	0.38	0.09	0.09	0.09
11	IT-103	0.14	0.12	0.13	0.05	0.05	0.05	0.53	0.43	0.48	0.07	0.06	0.07
12	IT-104	0.09	0.09	0.09	0.06	0.05	0.06	0.36	0.24	0.30	0.07	0.07	0.07
13	IT-105	0.09	0.09	0.09	0.04	0.04	0.04	0.39	0.27	0.33	0.07	0.06	0.07
14	IT-106	0.12	0.11	0.12	0.04	0.03	0.04	0.43	0.32	0.38	0.05	0.07	0.06
15	IT-107	0.05	0.06	0.06	0.03	0.03	0.03	0.31	0.29	0.30	0.05	0.04	0.05
16	IT-108	0.09	0.10	0.10	0.03	0.03	0.03	0.38	0.24	0.31	0.06	0.03	0.05
17	IT-109	0.10	0.09	0.10	0.04	0.04	0.04	0.41	0.31	0.36	0.06	0.05	0.06
18	IT-110	0.14	0.12	0.13	0.06	0.04	0.05	0.59	0.46	0.53	0.07	0.05	0.06
19	IT-111	0.12	0.11	0.12	0.05	0.05	0.05	0.48	0.35	0.41	0.07	0.06	0.06
20	IT-112	0.09	0.09	0.09	0.04	0.03	0.04	0.39	0.28	0.33	0.05	0.05	0.05
21	IT-113	0.09	0.09	0.09	0.03	0.03	0.03	0.30	0.25	0.28	0.05	0.05	0.05
22	IT-114	0.06	0.06	0.06	0.07	0.07	0.07	0.44	0.37	0.40	0.08	0.08	0.08
23	IT-115	0.06	0.06	0.06	0.06	0.06	0.06	0.29	0.22	0.26	0.07	0.07	0.07
24	IT-116	0.08	0.08	0.08	0.06	0.06	0.06	0.31	0.23	0.27	0.08	0.07	0.08
25	IT-117	0.09	0.09	0.09	0.04	0.03	0.04	0.34	0.24	0.29	0.05	0.05	0.05
26	IT-118	0.06	0.06	0.06	0.03	0.03	0.03	0.22	0.20	0.21	0.05	0.04	0.05
27	IT-119	0.07	0.06	0.07	0.05	0.05	0.05	0.27	0.23	0.25	0.06	0.06	0.06
28	IT-120	0.11	0.09	0.10	0.06	0.05	0.06	0.39	0.31	0.35	0.07	0.06	0.07
29	IT-121	0.04	0.06	0.05	0.05	0.05	0.05	0.22	0.20	0.21	0.07	0.07	0.07
30	IT-122	0.08	0.06	0.07	0.03	0.02	0.03	0.35	0.23	0.29	0.06	0.04	0.05
	Mean	0.10	0.09	0.10	0.05	0.05	0.05	0.40	0.30	0.35	0.07	0.06	0.06
Lx Sx V		SE			CD								
		0.0070			0.0195								

O - Open
S - Shade
M - Mean

Table 22. Leaf glycoside content (%) in *I. tinctoria* accessions –
Light × variety interaction

Sl. No.	Accession No.	Open	Shade	Mean
1	IT-93	0.19	0.14	0.16
2	IT-94	0.18	0.14	0.16
3	IT-95	0.15	0.12	0.13
4	IT-96	0.21	0.17	0.19
5	IT-97	0.16	0.13	0.14
6	IT-98	0.20	0.16	0.18
7	IT-99	0.24	0.18	0.21
8	IT-100	0.14	0.11	0.12
9	IT-101	0.16	0.12	0.14
10	IT-102	0.18	0.14	0.16
11	IT-103	0.20	0.16	0.18
12	IT-104	0.15	0.11	0.13
13	IT-105	0.15	0.12	0.13
14	IT-106	0.16	0.13	0.15
15	IT-107	0.11	0.10	0.11
16	IT-108	0.14	0.10	0.12
17	IT-109	0.15	0.12	0.14
18	IT-110	0.21	0.17	0.19
19	IT-111	0.18	0.14	0.16
20	IT-112	0.14	0.11	0.13
21	IT-113	0.12	0.11	0.11
22	IT-114	0.16	0.15	0.15
23	IT-115	0.12	0.10	0.11
24	IT-116	0.13	0.11	0.12
25	IT-117	0.13	0.11	0.12
26	IT-118	0.09	0.08	0.09
27	IT-119	0.11	0.10	0.11
28	IT-120	0.16	0.12	0.14
29	IT-121	0.06	0.09	0.09
30	IT-122	0.13	0.09	0.11
	Mean	0.15	0.12	-
		SE	CD	
	For leaf treatments (L)	0.0009	0.0025	
	For light conditions (S)	0.0006	0.0018	
	For varieties (V)	0.0025	0.0069	
	Lx S	0.0013	0.0035	
	Lx V	0.0050	0.0137	
	Sx V	0.0035	0.0097	

Table 23. Selection index (SI)* scores and ranks of *I. tinctoria* accessions

Sl. No.	Accession No.	SI Score	Rank
1	IT-93	571.94	28
2	IT-94	276.63	29
3	IT-95	203.77	30
4	IT-96	5909.04	3
5	IT-97	2837.34	11
6	IT-98	736.77	25
7	IT-99	2704.52	12
8	IT-100	2336.72	13
9	IT-101	7885.32	2
10	IT-102	1540.95	17
11	IT-103	959.54	22
12	IT-104	3639.52	8
13	IT-105	3346.11	9
14	IT-106	3733.35	7
15	IT-107	1717.07	15
16	IT-108	8766.41	1
17	IT-109	898.83	23
18	IT-110	1095.02	20
19	IT-111	2868.43	10
20	IT-112	2123.23	14
21	IT-113	5480.84	6
22	IT-114	5901.95	4
23	IT-115	5498.92	5
24	IT-116	748.87	24
25	IT-117	1673.61	16
26	IT-118	725.44	26
27	IT-119	584.52	27
28	IT-120	987.78	21
29	IT-121	1209.43	18
30	IT-122	1111.17	19

* - Based on leaf yield and glycoside content

replications. Observations were made at three stages of growth viz., pre-flowering stage (90 DAS), flowering stage (150 DAS) and seed maturation stage (240 DAS).

4.3.1 Growth parameters

4.3.1.1 Plant height

At pre-flowering stage, no significant difference could be noticed among the accessions under open condition, but significant difference was noticed under shade (Table 24). Maximum plant height (91.83 cm) was recorded by IT-108 under shade, followed by IT-106 (86.70 cm) and IT-101 (86.24cm), which were statistically on par. The least plant height (65.13 cm) was recorded by IT-105.

At flowering stage also, significant difference could be noticed among the accessions only under shade. Maximum plant height (105.91 cm) was recorded by IT-108 under shade. It was on par with IT-96 (104.90 cm), IT-101 (104.83 cm) and IT-114 (104.62 cm). The lowest plant height (82.67 cm) was recorded by IT-111.

At seed maturation stage, there was no significant difference between the accessions under both the light conditions. Plant height was more in open condition than that under shade at all the three stages of growth.

4.3.1.2 Plant spread

Pooled analysis indicated that there was significant difference in plant spread between open and shaded conditions at all stages of growth (Table 25). It was more in open condition than that under shade.

At pre-flowering stage, there was significant difference among the accessions under open as well as shaded conditions. Maximum pooled mean for plant spread (88.44 cm) was observed in IT-101, which was on par with IT-111 (87.01 cm) and IT-99 (86.12 cm). Pooled mean was minimum for IT-108 (69.38 cm).

Table 24. Growth of selected *I. tinctoria* accessions under open and shaded conditions: plant height (cm)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 108	93.54	91.83	142.99	105.91	218.79	137.27
2	IT- 101	97.74	86.24	147.40	104.83	212.92	132.22
3	IT- 96	98.89	78.38	141.78	104.90	212.20	132.41
4	IT- 114	92.82	74.74	132.47	104.62	181.65	123.59
5	IT- 106	92.47	86.70	127.73	102.09	189.53	129.18
6	IT- 104	81.90	72.47	120.63	95.60	192.02	114.12
7	IT- 105	90.35	65.13	129.85	86.03	188.89	120.82
8	IT- 111	98.55	77.94	125.37	82.67	202.60	126.74
9	IT- 97	88.73	79.06	122.21	93.63	198.37	111.89
10	IT- 99	88.61	82.78	127.06	89.81	199.67	118.51
	SE	6.39	4.28	8.71	3.06	17.83	8.09
	CD	NS	12.73	NS	9.08	NS	NS

Table 25. Growth of selected *L. tinctoria* accessions under open and shaded conditions: plant spread (cm)

Sl. No.	Accession No.	Stages of plant growth								
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (240 DAS)		
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	76.28	62.47	69.38	107.08	82.23	94.65	163.56	91.59	127.58
2	IT-101	98.07	78.81	88.44	120.10	94.27	107.18	178.58	112.96	145.77
3	IT-96	94.95	72.05	83.50	108.83	87.88	98.36	142.91	97.57	120.24
4	IT-114	86.18	67.99	77.09	116.46	93.97	105.21	139.69	99.80	119.75
5	IT-106	87.05	70.48	78.77	105.56	82.55	94.06	128.98	102.10	115.54
6	IT-104	80.71	66.89	73.80	112.91	93.15	103.03	143.02	108.56	125.79
7	IT-105	108.03	62.86	85.45	133.09	92.70	112.89	156.92	102.28	129.60
8	IT-111	101.51	72.51	87.01	119.12	81.66	100.39	141.07	104.51	122.79
9	IT-97	86.56	75.20	80.88	110.41	82.67	96.54	126.12	94.68	110.40
10	IT-99	94.29	77.94	86.12	111.57	89.46	100.51	150.77	97.85	124.31
	Mean	91.36	70.72	-	114.51	88.05	-	147.16	101.19	-
	Pooled analysis'		SE	CD		SE	CD		SE	CD
	For light condition		2.23	6.41		1.41	4.03		3.35	9.62
	For accessions		5.00	14.34		3.16	9.02		7.49	21.50
	For interaction		7.07	20.28		4.47	12.75		10.59	30.40

At flowering stage also, significant difference could be observed between the accessions under both the light conditions. IT-105 recorded the maximum pooled mean for plant spread (112.89 cm) followed by IT-101 (107.18 cm). IT-106 recorded the minimum pooled mean (94.06 cm).

At seed maturation stage also, there was significant difference among the accessions under both the light conditions. Pooled mean was maximum (145.77 cm) for IT-101, which was on par with IT-105 (129.60 cm), IT-108 (127.58 cm) and IT-104 (125.79 cm). Pooled mean was least (110.40 cm) in IT-97.

IT-101 showed good performance in terms of plant spread at all the three stages of plant growth.

4.3.1.3 Height at first branching

Pooled analysis indicated that there was significant difference in height at first branching between open and shaded conditions at pre-flowering stage and flowering stages (Table 26).

At pre-flowering stage, there was significant difference among the accessions for height at first branching. Maximum pooled mean (9.78 cm) was observed for IT-101, which was on par with IT-96 (9.50 cm) and IT-108 (8.54 cm). Minimum pooled mean (6.24 cm) was observed in IT-97.

At flowering stage also, the accessions showed significant difference among them for height of first branch. IT-96 recorded the maximum pooled mean (10.17 cm), followed by IT-101 (9.9 cm) and IT-108 (9.24 cm). Minimum value (7.25 cm) was recorded by IT-104.

At seed maturation stage, significant difference was observed among the accession only under shaded condition. Under shaded condition, maximum height at first branching (12.43 cm) was observed in IT-96, which was on par with IT-101 (11.14 cm) and minimum in IT-104 (7.42 cm).

IT-96 and IT-101 produced the first branch at higher position than the other accessions.

Table 26. Growth of selected *I. tinctoria* accessions under open and shaded conditions: height at first branching (cm)

Sl. No.	Accession No.	Stages of plant growth							
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (240 DAS)	
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade
1	IT- 108	7.82	9.27	8.54	8.51	9.97	9.24	10.84	9.50
2	IT- 101	7.11	12.45	9.78	8.36	11.44	9.90	10.50	11.14
3	IT- 96	6.95	12.04	9.50	7.93	12.40	10.17	9.58	12.43
4	IT- 114	7.09	9.12	8.11	8.02	8.85	8.44	10.41	9.62
5	IT- 106	6.47	7.37	6.92	7.43	7.88	7.66	9.26	7.68
6	IT- 104	6.23	6.55	6.39	7.01	7.25	7.13	7.98	7.42
7	IT- 105	5.78	7.15	6.47	7.95	7.55	7.75	8.98	7.85
8	IT- 111	7.50	7.33	7.42	8.81	8.11	8.46	9.64	7.99
9	IT- 97	5.21	7.26	6.24	7.07	8.32	7.70	8.56	8.47
10	IT- 99	6.71	6.70	6.70	7.00	7.50	7.25	8.74	8.07
	Mean	6.69	8.52	-	7.81	8.93	-	9.45	9.02
	SE	-	-	-	-	-	-	0.77	0.51
	CD	-	-	-	-	-	-	NS	1.50
Pooled analysis			SE	CD		SE	CD		
For light condition			0.43	1.25		0.35	1.02		
For accessions			0.97	2.79		0.79	2.27		
For interaction			1.37	3.94		1.12	3.21		

Table 27. Growth of selected *I. tinctoria* accessions under open and shaded conditions: number of branches

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 108	33.93	17.90	74.93	27.99	119.39	52.59
2	IT- 101	34.00	17.49	75.05	25.44	123.78	47.32
3	IT- 96	34.20	16.05	75.55	23.52	132.95	47.74
4	IT- 114	32.86	12.97	68.57	22.27	128.83	53.58
5	IT- 106	26.19	13.68	64.21	20.62	100.51	41.22
6	IT- 104	24.74	12.83	57.62	19.16	105.39	39.34
7	IT- 105	28.16	13.21	65.98	19.58	100.60	44.91
8	IT- 111	31.70	14.73	67.33	18.07	83.05	39.01
9	IT- 97	24.69	15.61	57.61	18.40	112.48	37.83
10	IT- 99	28.95	16.74	68.60	20.61	108.68	46.90
	SE	3.95	1.78	7.47	1.86	11.27	3.23
	CD	NS	NS	NS	5.53	NS	9.58

4.3.1.4 Number of branches

There was significant difference in number of branches between open and shaded conditions at all stages of growth (Table 27). More branches were produced by the accessions when grown in open condition compared to shade.

At pre-flowering stage, significant difference was not observed among the accessions for number of branches under both open and shaded conditions.

At flowering stage, accessions differed significantly for number of branches under shaded condition only. Under shade, maximum number of branches (27.99) was recorded by IT-108, which was on par with IT-101 (25.44) and IT-96 (23.52). IT-111 produced minimum number of branches (18.07).

At seed maturation stage also, there was significant difference between the accessions only under shaded condition. IT-114 produced more number of branches (53.58) under shade, followed by IT-108 (52.59). Minimum number of branches (37.83) was produced by IT-97.

4.3.1.5 Girth of stem

There was significant difference in girth of stem between open and shaded conditions at all stages of growth (Table 28) and it was more in open condition.

At pre-flowering stage, accessions differed significantly for girth of stem only under shaded condition. Maximum stem girth (1.92 cm) was recorded by IT-99, followed by IT-104 (1.87 cm) and IT-111 (1.82 cm). Girth of stem was minimum (1.29 cm) in IT-105.

At flowering stage, pooled analysis indicated that there was significant difference among the accessions. Pooled mean was maximum for IT-114 (4.02 cm) followed by IT-106 (3.69 cm) and IT-96 (3.67 cm). Minimum pooled mean (3.22 cm) was recorded by IT-105.

Table 28. Growth of selected *I. tinctoria* accessions under open and shaded conditions: girth of stem (cm)

Sl. No	Accession No.	Stages of plant growth						
		Pre-flowering (90 DAS)		Flowering (150 DAS)			Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Mean	Open	Shade
1	IT- 108	2.83	1.78	4.10	2.35	3.23	10.76	3.82
2	IT- 101	3.72	1.78	4.84	2.48	3.66	10.15	3.80
3	IT- 96	2.94	1.32	5.06	2.27	3.67	8.95	3.56
4	IT- 114	2.69	1.47	5.21	2.83	4.02	9.42	4.02
5	IT- 106	3.14	1.81	4.72	2.65	3.69	8.75	3.47
6	IT- 104	2.91	1.87	4.14	2.70	3.42	8.39	3.47
7	IT- 105	2.82	1.29	4.25	2.18	3.22	7.89	3.51
8	IT- 111	2.78	1.82	4.12	2.52	3.32	7.06	3.88
9	IT- 97	2.93	1.34	3.85	2.73	3.29	8.30	3.64
10	IT- 99	2.98	1.92	4.10	2.65	3.38	8.55	3.95
	Mean	2.98	1.64	4.44	2.54	-	8.82	3.71
	SE	0.31	0.15	-	-	-	1.04	0.15
	CD	NS	0.44	-	-	-	NS	NS
Pooled analysis					SE	CD		
For light condition					0.12	0.33		
For accessions					0.26	0.74		
For interaction					0.37	1.05		

Table 29. Growth of selected *I. tinctoria* accessions under open and shaded conditions: number of leaves

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 108	240.09	182.16	569.74	250.78	957.45	381.24
2	IT- 101	224.65	150.59	508.98	224.59	948.34	371.41
3	IT- 96	283.24	140.95	534.48	195.03	939.26	400.44
4	IT- 114	305.65	123.54	599.23	209.90	855.71	366.12
5	IT- 106	306.64	131.06	475.57	204.12	861.46	353.21
6	IT- 104	278.38	112.00	528.83	188.85	777.15	354.17
7	IT- 105	279.13	118.33	479.96	185.18	855.78	400.20
8	IT- 111	265.48	140.35	527.43	179.49	786.82	376.52
9	IT- 97	261.40	117.80	512.09	180.47	903.93	353.49
10	IT- 99	282.99	127.09	573.09	207.79	889.50	364.19
	SE	52.81	10.84	44.73	11.53	50.79	19.31
	CD	NS	32.22	NS	34.27	NS	NS

At seed maturation stage, there was no significant difference among the accessions under both conditions.

4.3.1.6 Number of leaves

There was significant difference in number of leaves between open and shaded conditions at all stages of growth (Table 29). The accessions produced more leaves when grown in open condition compared to shade.

At pre-flowering stage, accessions showed significant difference among them only under shaded condition. In shade, IT-108 produced maximum number of leaves (182.16), which was on par with IT-101 (150.59). Minimum number of leaves (112.00) was observed in IT-104.

At flowering stage also, there was significant difference among the accessions under shaded condition only. IT-108 produced maximum number of leaves (250.78) under shade, followed by IT-101 (224.59). IT-111 produced minimum number of leaves (179.49).

At seed maturation stage, significant difference could not be observed among the accessions under both conditions.

4.3.1.7 Leaf area

Significant difference could be observed in leaf area between open and shaded conditions at all the three growth stages (Table 30). Higher leaf area was observed under open conditions.

At pre-flowering stage, pooled analysis showed significant difference among the accessions. Pooled mean was highest for IT-101 (1813.68 cm²). It was on par with IT-108 (1787.11 cm²), IT-114 (1655.99 cm²) and IT-96 (1634.22 cm²). Pooled mean for leaf area was least for IT-105 (1147.56 cm²).

At flowering stage also, the accessions showed significant difference among them for leaf area. IT-106 recorded the maximum pooled mean (6078.81 cm²) followed by IT-108 (5607.83 cm²), IT-96 (5551.68 cm²) and IT-104 (5524.59 cm²). Minimum value was recorded by IT-97 (4611.08 cm²).

Table 30. Growth of selected *I. tinctoria* accessions under open and shaded conditions: leaf area (cm²)

Sl No.	Accession No.	Stages of plant growth											
		Pre-flowering (90 DAS)				Flowering (150 DAS)				Seed maturation (240 DAS)			
		Open	Shade	Mean	SE	Open	Shade	Mean	SE	Open	Shade	Mean	SE
1	IT-108	2192.32	1381.90	1787.11	7169.17	4046.49	5607.83	17398.29	14617.55	16007.92			
2	IT-101	2213.91	1413.45	1813.68	6925.10	3934.01	5429.56	17603.54	13857.27	15730.41			
3	IT-96	2025.89	1242.56	1634.22	6918.42	4184.94	5551.68	16682.64	13742.82	15212.73			
4	IT-114	2032.33	1279.64	1655.99	5654.02	4567.79	5110.81	15822.65	17359.17	16590.91			
5	IT-106	1330.59	1245.35	1287.97	6973.02	5184.61	6078.81	16852.97	15515.58	16184.28			
6	IT-104	1443.82	1289.41	1366.62	7328.97	3720.21	5524.59	16070.86	12992.54	14531.70			
7	IT-105	1288.56	1006.59	1147.56	4979.09	4775.40	4977.24	16040.55	12395.83	14218.19			
8	IT-111	1734.67	1190.17	1462.42	6870.89	3777.26	5324.08	15212.46	12170.38	13691.42			
9	IT-97	1276.07	1219.76	1247.92	5955.28	3266.87	4611.08	15199.78	10445.04	12822.41			
10	IT-99	1457.54	1336.94	1397.24	6962.80	2761.37	4862.08	15461.21	10625.64	13043.43			
	Mean	1699.57	1260.58	-	6573.68	4021.88	-	16234.50	13372.18	-			
Pooled analysis			SE	CD		SE	CD		SE	CD			
For light condition			73.61	211.32		268.19	769.94		412.55	1184.37			
For accessions			164.60	472.54		599.69	1721.63		922.49	2648.33			
For interaction			232.78	668.27		848.09	2434.75		1304.59	3745.30			

At seed maturation stage also, there was significant difference among the accessions for leaf area. Maximum pooled mean was observed for IT-114 (16590.91 cm²) which was on par with IT-106 (16184.28 cm²), IT-108 (16007.92 cm²) and IT-101 (15730.41 cm²). IT-97 recorded the minimum pooled mean (12822.41 cm²).

4.3.1.8 Root length

There was significant difference in root length between open and shaded conditions at all the three stages of growth (Table 31). Longer root was recorded under open condition.

At pre-flowering stage, accessions showed significant difference among them only under open condition. Maximum root length (39.67 cm) under open condition was observed in IT-101 and minimum in IT-105 (23.88 cm).

At flowering stage also, there was significant difference among the accessions only under open condition. In open condition, IT-101 recorded the longest root (41.61 cm), which was on par with IT-96 (36.71 cm) and IT-114 (36.56 cm). Lowest root length (26.82 cm) was observed in IT-97.

At seed maturation stage, significant difference could be observed among the accessions under both open and shaded conditions. Under open condition, root length was maximum for IT-101 (43.61 cm) followed by IT-108 (42.12 cm) and minimum for IT-104 (34.54 cm). Under shade, maximum root length (33.92 cm) was recorded by IT-108, which was on par with IT-101 (32.87 cm) and IT-96 (32.32 cm) and minimum by IT-105 (27.95 cm).

IT-101 produced longer roots at all the three stages of growth compared to the other accessions.

4.3.1.9 Root girth at collar region

There was significant difference in root girth at collar region between open and shaded conditions at all the three stages of growth (Table 32).

Table 31. Growth of selected *I. tinctoria* accessions under open and shaded conditions: root length (cm)

Sl. No.	Accession No.	Stages of plant growth					
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 108	28.95	18.75	34.68	26.07	42.12	33.92
2	IT- 101	39.67	21.72	41.61	25.53	43.61	32.87
3	IT- 96	32.32	21.89	36.71	25.89	39.52	32.32
4	IT- 114	31.27	22.13	36.56	25.57	39.69	30.46
5	IT- 106	31.18	22.17	32.70	24.58	34.72	31.58
6	IT- 104	28.75	22.22	31.38	24.99	34.54	29.18
7	IT- 105	23.88	18.00	34.94	22.45	37.51	27.95
8	IT- 111	27.64	19.01	33.79	20.45	39.40	28.12
9	IT- 97	25.93	19.75	26.82	22.31	37.35	29.34
10	IT- 99	29.89	20.64	34.58	22.90	35.97	28.38
	SE	1.79	1.42	1.97	1.60	1.91	1.30
	CD	5.33	NS	5.86	NS	5.68	3.86

Table 32. Growth of selected *I. tinctoria* accessions under open and shaded conditions: root girth at collar region (cm)

Sl. No.	Accession No.	Stages of plant growth							
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (240 DAS)	
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade
1	IT- 108	3.18	1.77	2.48	4.96	2.71	3.84	10.83	3.86
2	IT- 101	4.38	1.83	3.10	5.00	2.86	3.93	10.49	3.91
3	IT- 96	3.31	1.54	2.43	5.40	2.53	3.96	9.23	3.75
4	IT- 114	3.88	1.68	2.78	5.39	2.87	4.13	9.65	3.81
5	IT- 106	3.46	2.06	2.76	5.17	2.85	4.01	8.71	3.62
6	IT- 104	3.61	1.95	2.78	4.61	2.70	3.66	9.14	3.79
7	IT- 105	3.66	1.68	2.67	4.74	2.63	3.69	8.30	3.69
8	IT- 111	3.54	1.93	2.73	4.28	2.91	3.60	7.95	3.70
9	IT- 97	3.14	1.65	2.40	4.34	2.75	3.54	7.26	3.62
10	IT- 99	3.62	1.91	2.77	5.58	2.67	4.13	7.84	3.62
	Mean	3.58	1.80	-	4.95	2.75	-	8.94	3.74
	SE	-	-	-	-	-	-	0.88	9.13
	CD	-	-	-	-	-	-	NS	NS
Pooled analysis		SE		CD	SE		CD		
For light condition		7.77		0.22	9.23		0.26		
For accessions		0.17		0.49	0.20		0.59		
For interaction		0.25		0.70	0.29		0.83		

Root girth at collar region was more under open condition compared to shade.

At pre-flowering stage, pooled analysis showed significant difference among the accessions. Pooled mean was maximum for IT-101 (3.10 cm) followed by IT-114 (2.78 cm) and IT-104 (2.78 cm). Minimum pooled mean (2.40 cm) was recorded by IT-97.

At flowering stage also, pooled analysis showed significant difference among the accessions. Maximum pooled mean (4.13 cm) was recorded by IT-114 and IT-99, followed by IT-106 (4.01 cm).

At seed maturation stage, significant difference could not be observed among the accessions under both conditions.

4.3.2 Root nodule characteristics

Root nodules were found to be present in the roots of *I. tinctoria* only during the pre-flowering and flowering stages of growth of the plant (Plate 3).

4.3.2.1 Number of root nodules/ plant

Pooled analysis showed that there was significant difference for the number of root nodules per plant among the accessions at the flowering stage only (Table 33). IT-101 produced the maximum number of root nodules per plant (76.80) followed by IT- 105 (76.17) and IT-111 (75.63). IT-97 produced the least number of root nodules per plant (68.13).

More number of root nodules were produced by all the accessions under open condition compared to shade.

4.3.2.2 Number of effective root nodules/plant

Pooled analysis indicated that significant difference could be observed for the number of effective root nodules per plant among the accessions at the flowering stage only (Table 33). IT-99 produced the maximum number of effective root nodules per plant (42.75) followed by

Table 33. Root nodule characteristics of selected *I. tinctoria* accessions under open and shaded conditions: number of root nodules/ plant and number of effective root nodules/ plant

Sl. No.	Accession No.	No. of root nodules/ plant						No. of effective root nodules/ plant					
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Pre-flowering (90 DAS)			Flowering (150 DAS)		
		Open		Shade	Open		Shade	Open		Shade	Open		Shade
		Mean	SE	CD	Mean	SE	CD	Mean	SE	CD	Mean	SE	CD
1	IT-108	11.80	5.67	90.46	54.38	72.42	5.71	2.63	49.75	23.63	36.69		
2	IT-101	8.46	4.88	103.34	50.25	76.80	4.09	2.33	57.30	25.50	41.40		
3	IT-96	7.50	4.58	97.25	48.46	72.86	3.59	2.04	56.13	26.59	41.36		
4	IT-114	6.84	4.92	84.00	52.63	68.32	3.79	2.50	47.96	32.71	40.34		
5	IT-106	10.00	6.42	85.34	61.04	73.19	5.55	3.25	48.25	33.79	41.02		
6	IT-104	7.54	4.59	90.50	55.05	72.78	4.08	2.17	49.29	31.17	40.23		
7	IT-105	7.71	4.84	92.21	60.13	76.17	4.00	2.46	51.54	32.50	42.02		
8	IT-111	9.79	5.84	100.25	51.00	75.63	4.75	3.00	51.09	28.63	39.86		
9	IT-97	8.88	5.50	83.79	52.46	68.13	4.46	2.71	47.09	32.50	39.80		
10	IT-99	8.75	6.29	94.84	58.25	76.54	4.75	2.92	52.92	32.92	42.75		
	Mean	8.73	5.35	92.20	54.37	-	4.48	2.60	51.10	29.99	-		
	SE	1.22	0.54	-	-	-	0.57	0.25	-	-	-		
	CD	NS	NS	-	-	-	NS	NS	-	-	-		
	Pooled analysis												
	For light condition				SE	CD				SE	CD		
	For accessions				2.10	6.03				1.39	4.00		
	For interaction				4.69	13.48				3.11	8.94		
					6.64	19.06				4.40	12.64		



Plate 3. Root nodules of *Indigofera tinctoria* at flowering stage

IT-105 (42.02), IT-101 (41.40) and IT-96 (41.36). Minimum number of effective root nodules per plant (36.69) was produced by IT-108.

Under open condition, the number of effective root nodules/ plant were found to be higher compared to shade.

4.3.2.3 Fresh weight of root nodules

At pre-flowering stage, pooled analysis showed that the accessions differed significantly for the fresh weight of root nodules (Table 34). Maximum fresh weight of root nodules (0.045 g) was observed in IT-108 followed by IT-101 (0.043 g) and IT- 111 (0.040 g). Minimum fresh weight of root nodules was recorded by IT-114 (0.024 g).

At flowering stage also, pooled analysis showed significant difference among the accessions for fresh weight of root nodules. Significantly higher fresh weight of root nodules (1.304 g) was recorded by IT-111, which was on par with IT-105 (1.254 g) and IT-104 (1.226 g). Lowest fresh weight of root nodules was observed in IT-108 (1.114 g).

Fresh weight of root nodules was higher under open condition in all the accessions compared to shade.

4.3.2.4 Dry weight of root nodules

Dry weight of root nodules at pre-flowering stage showed significant variation among the accessions on pooled analysis (Table 34). Significantly higher nodule dry weight (0.035 g) was recorded by IT-108, which was on par with IT-101 (0.034 g). IT- 114 recorded the lowest nodule dry weight (0.019 g).

At flowering stage also, significant difference occurred among the accessions for the dry weight of root nodules. Dry weight of root nodules was maximum in IT-111 (1.124 g) followed by IT-105 (1.043 g) and IT-104 (1.019 g).

Dry weight of root nodules was also higher in all the accessions under open condition compared to shade.

Table 34. Root nodule characteristics of selected *I. tinctoria* accessions under open and shaded conditions: fresh weight of root nodules/ plant (g) and dry weight of root nodules/ plant (g)

Sl. No.	Accession No.	Fresh weight of root nodules/ plant (g)						Dry weight of root nodules/ plant (g)					
		Pre-flowering stage (90 DAS)			Flowering stage (150 DAS)			Pre-flowering stage (90 DAS)			Flowering stage (150 DAS)		
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	0.070	0.020	0.045	1.423	0.804	1.114	0.053	0.016	0.035	1.072	0.700	0.886
2	IT-101	0.063	0.022	0.043	1.503	0.867	1.185	0.049	0.018	0.034	0.073	0.774	0.924
3	IT-96	0.057	0.020	0.038	1.487	0.893	1.190	0.043	0.016	0.030	1.161	0.782	0.971
4	IT-114	0.030	0.019	0.024	1.453	0.856	1.155	0.023	0.015	0.019	1.163	0.756	0.959
5	IT-106	0.030	0.021	0.026	1.520	0.764	1.142	0.024	0.017	0.021	1.270	0.667	0.968
6	IT-104	0.043	0.019	0.031	1.553	0.899	1.226	0.033	0.016	0.024	1.252	0.785	1.019
7	IT-105	0.050	0.020	0.035	1.593	0.915	1.254	0.038	0.016	0.027	1.276	0.809	1.043
8	IT-111	0.060	0.019	0.040	1.763	0.844	1.304	0.043	0.016	0.030	1.474	0.775	1.124
9	IT-97	0.050	0.022	0.036	1.357	0.988	1.172	0.035	0.019	0.027	1.048	0.875	0.961
10	IT-99	0.033	0.021	0.027	1.487	0.803	1.145	0.26	0.017	0.022	1.174	0.686	0.930
	Mean	0.049	0.020	-	1.514	0.863	-	0.037	0.017	-	1.196	0.761	-
Pooled analysis		SE	CD	SE	CD	SE	CD	SE	CD	SE	CD	SE	CD
For light condition		0.003	0.009	0.03	0.077	0.002	0.007	0.033	0.095	0.003	0.007	0.033	0.095
For accessions		0.007	0.020	0.06	0.172	0.005	0.015	0.074	0.213	0.005	0.015	0.074	0.213
For interaction		0.010	0.030	0.09	0.243	0.008	0.022	0.105	0.300	0.008	0.022	0.105	0.300

4.3.3 Flowering and seeding behaviour of *Indigofera tinctoria*

Flowers of *I. tinctoria* are numerous and sessile, racemes 5 -10 cm long with 20 to 40 flowers/ inflorescence. The corolla is pink in colour, consisting of a rounded standard petal, brownish and pubescent at the back and two wing petals adherent to the two keel petals which are greenish in colour. Pods 2-3.2 cm long, linear, straight or slightly curved, pale greenish grey when young and dark brown on ripening with 8-12 seeds (Plate 4).

4.3.3.1 Number of days for flowering

Observations on the number of days for flowering are presented in Table 35. Under open condition, IT-96 was found to flower earlier (111 days) than other accessions. IT-101 (116 days) and IT-111 (116 days) took more number of days to flower. Under shade, IT-104 flowered earlier (117 days) than the other accessions while IT-106 (126 days) and IT-97 (126 days) were late in flowering.

4.3.3.2 Total number of flowers produced per plant

The data on the total number of flowers produced per plant are given in Table 35. IT-101 produced more number of flowers (22382.17) under open condition followed by IT-96 (20825.09). Least number of flowers (14378.84) was produced by IT-106. In shaded condition also, IT-101 produced more number of flowers (10764.21) followed by IT-97 (10545.84). Lowest number of flowers (8625.71) was produced by IT-104.

4.3.3.3 Time of anthesis

Observations on time of anthesis are presented in Table 35. IT-97 recorded the earliest time of anthesis (8.27 a.m.) under open condition followed by IT-108 (8.30 a.m.). Flower opening was late in IT-101 (9.12 a.m.). Under shade, earlier flower opening was observed in IT-108 (8.46 a.m.) and IT-97 (9.10 a.m.). IT-111 was late in flower opening (10.08 a.m.).

Table 35. Flowering behaviour and anthesis of selected *I. tinctoria* accessions

Sl. No.	Accession No.	No. of days for flowering		No. of flowers/ inflorescence		Total no. of flowers produced/ plant		Time of anthesis (a.m.)		Time of anther dehiscence (p.m.)	
		Open	Shade	Open	Shade	Open	Shade	Open	Shade	Open	Shade
1	IT- 108	115	124	34.82	22.92	18872.42	10171.67	8.30	8.46	10.57	11.16
2	IT- 101	116	121	42.07	27.25	22382.17	10764.21	9.12	10.03	11.41	11.41
3	IT- 96	111	120	37.56	21.08	20825.09	10104.04	9.05	10.05	11.25	11.58
4	IT- 114	113	124	31.65	26.60	18005.50	9824.38	9.08	10.01	11.50	12.03
5	IT- 106	114	126	35.07	35.56	14378.84	10016.29	8.35	9.27	11.18	12.18
6	IT- 104	113	117	40.50	24.00	19542.29	8625.71	8.43	9.21	11.16	11.41
7	IT- 105	115	124	29.13	34.63	17506.67	8977.13	-	8.50	11.16	11.33
8	IT- 111	116	123	30.06	29.38	19995.50	10225.71	9.01	10.08	11.04	11.53
9	IT- 97	112	126	30.00	23.22	12797.50	10545.84	8.27	9.10	11.24	11.43
10	IT- 99	115	122	39.00	30.75	19261.21	10403.75	8.44	10.00	11.31	11.41



a



b



c

Plate 4. Stages of development of inflorescence in *Indigofera tinctoria*

a. Inflorescence at different stages of development (right to left)

b. A branch with inflorescence

c. A branch with pods

4.3.3.4 Time of anther dehiscence

The data on the time of anther dehiscence are presented in Table 35. Under open condition, anther dehiscence was earliest (10.57 p.m.) in IT-108 followed by IT-111 (11.04 p.m.) and late in IT-114 (11.50 p.m.). Under shade, anther dehiscence was observed first in IT-108 (11.16 p.m.) followed by IT-105 (11.33 p.m.) and last in IT-106 (12.18 p.m.).

4.3.3.5 Number of pollen grains/anther

Observations on the number of pollen grains per anther are recorded in Table 36. Number of pollen grains per anther was highest (2400) under open condition in IT-104, followed by IT-96 (2133.33) and lowest in IT-111 (1066.67). In shade, IT-114 recorded the highest number of pollen grains per anther (2755.56) followed by IT-99 (1955.55) and least in IT-108 (1066.67).

4.3.3.6 Pollen fertility

Pollen fertility was assessed by staining method (Table 36). In open condition, maximum pollen fertility (69.29%) was observed in IT-106 followed by IT-111 (69.22%). IT-105 recorded the minimum (53.67%). In shade, IT-96 recorded the maximum pollen fertility (69.77%) followed by IT-99 (66.41%) and IT-111 (56.67%).

4.3.3.7 Percentage of pod set

The data on the percentage of pod set are presented in Table 36. Highest percentage of pod set (23.78%) was recorded in open condition by IT-106 followed by IT-114 (22.46%). Lowest value (18.03%) was recorded by IT-101. Under shade, IT-114 recorded the highest percentage of pod set (21.04%) followed by IT-97 (20.35%). Percentage of pod set was least in IT-111 (13.55%).

Table 36. Pollen and seeding behaviour of selected *I. tinctoria* accessions

Sl. No.	Accession No.	No. of pollen grains/ anther		Pollen fertility (%)		Percentage of pod set (%)		No. of days for seed set		No. of days for seed maturation	
		Open	Shade	Open	Shade	Open	Shade	Open	Shade	Open	Shade
1	IT-108	1600.00	1066.67	65.42	65.11	21.17	19.33	139	148	237	252
2	IT-101	2044.44	1244.44	68.51	65.52	18.03	14.93	141	147	241	246
3	IT-96	2133.33	1155.55	66.31	69.77	19.60	19.88	136	144	236	242
4	IT-114	1244.44	2755.56	68.96	59.90	22.46	21.04	136	149	238	258
5	IT-106	1689.89	1155.56	69.29	64.41	23.78	16.09	142	149	246	252
6	IT-104	2400.00	1244.44	63.14	61.38	20.63	18.74	136	148	234	251
7	IT-105	1866.67	1333.33	53.67	61.83	21.38	15.44	142	152	244	260
8	IT-111	1066.67	1333.33	69.22	56.67	21.24	13.55	143	151	248	257
9	IT-97	1955.57	1244.44	67.26	63.87	21.55	20.35	133	152	235	259
10	IT-99	1777.78	1955.55	69.25	66.41	18.81	16.52	136	150	235	258

4.3.3.8 Number of days for seed set

Observations on the number of days for seed set from the date of sowing are recorded in Table 36. Under open condition, seed setting was earlier in IT-97 (133 days) and late in IT-111 (143 days). In shaded condition, earlier seed setting was observed in IT-96 (144 days) and late in IT-105 and IT-97 (152 days).

4.3.3.9 Number of days for seed maturation

The data on the number of days for seed maturation from the date of sowing are presented in Table 36. Under open condition, IT-104 took 234 days for seed maturation followed by IT-97 (235 days) and IT-99 (235 days) while IT-111 took 248 days. Under shade, IT-96 recorded less number of days for seed maturation (242 days) and IT-105 recorded more (260 days).

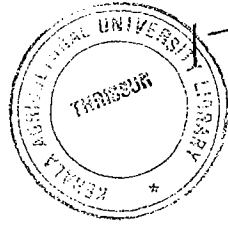
4.3.4 Yield attributes

4.3.4.1 Fresh weight of shoot

Pooled analysis showed that there was significant difference in shoot fresh weight between open and shaded conditions at all stages of growth (Table 37).

At pre-flowering stage, there was significant difference among the accessions. Maximum pooled mean for shoot fresh weight (158.18 g) was observed in IT-101, which was on par with IT-108 (152.49 g), IT-114 (137.79 g) and IT-96 (137.44 g). Pooled mean was minimum for IT-97 (101.63 g). IT-101 recorded the maximum shoot fresh weight under open (191.16 g) and under shade (125.20 g).

At flowering stage also, accessions showed significant difference among them on pooled analysis. IT-104 recorded the maximum pooled mean for shoot fresh weight (644.20 g) followed by IT-106 (639.86 g) and IT-108 (632.63 g). IT-105 recorded the minimum pooled mean (465.57 g).



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Table 37. Yield of selected *I. tinctoria* accessions under open and shaded conditions: fresh weight of shoot (g)

Sl. No.	Accession No.	Stages of plant growth											
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (240 DAS)					
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	186.70	118.29	152.49	723.53	541.73	632.63	3373.58	1927.88	2650.73			
2	IT-101	191.16	125.20	158.18	638.80	497.67	568.24	3480.13	1701.15	2590.64			
3	IT-96	172.74	102.14	137.44	642.47	563.99	603.23	2755.57	1790.67	2273.12			
4	IT-114	170.57	105.02	137.79	536.26	532.83	534.55	2206.38	1985.05	2095.72			
5	IT-106	122.98	101.43	112.21	680.29	599.43	639.86	2963.73	1913.66	2438.70			
6	IT-104	123.46	90.97	107.21	766.34	522.06	644.20	2210.00	1624.77	1917.39			
7	IT-105	114.58	96.10	105.34	422.45	508.69	465.57	2246.12	1502.08	1874.10			
8	IT-111	145.19	96.26	120.72	637.43	514.94	576.18	2008.82	1116.56	1562.69			
9	IT-97	108.11	95.16	101.63	506.15	426.54	466.35	1936.17	1003.25	1469.71			
10	IT-99	132.06	115.77	123.91	610.20	471.98	541.09	2092.32	1038.67	1565.50			
	Mean	146.75	104.63	-	616.39	517.99	-	2527.28	1560.37	-			
Pooled analysis			SE	CD		SE	CD		SE	CD			
For light condition			5.40	15.52		20.60	59.15		96.07	275.79			
For accessions			12.09	34.70		46.07	132.26		214.81	616.68			
For interaction			17.09	49.07		65.15	187.04		303.78	872.12			



Plate 5. Superior accessions
of *Indigofera tinctoria* for
leaf yield and glycoside content

- a. IT - 108
- b. IT - 101
- c. IT - 96

At seed maturation stage also, significant difference was observed among the accessions (Fig. 5). Pooled mean was highest for IT-108 (2650.73 g). It was on par with IT-101 (2590.64 g), IT-106 (2438.70 g) and IT-96 (2273.12 g). Minimum pooled mean (1469.71 g) was observed in IT-97.

4.3.4.2 Dry weight of shoot

Pooled analysis indicated significant difference in shoot dry weight between open and shaded conditions at all stages of growth (Table 38).

At pre-flowering stage, pooled analysis showed significant difference among the accessions. Maximum pooled mean for shoot dry weight (48.75 g) was observed in IT-101, which was on par with IT-108 (47.08 g) and IT-96 (40.94 g). Minimum pooled mean (32.10 g) was observed in IT-97. IT-101 recorded the maximum shoot dry weight under open (58.15 g) and under shade (39.35 g).

At flowering stage also, there was significant difference among the accessions. IT-106 recorded the maximum pooled mean for dry weight of shoot (160.22 g) followed by IT-104 (160.12 g), IT-108 (158.49 g) and IT-96 (157.42 g). Minimum value (114.98 g) was recorded by IT-97.

At seed maturation stage also, significant difference was observed among the accessions (Fig. 6). Pooled mean was highest for IT-101 (500.51 g), which was on par with IT-108 (497.05 g) and IT-106 (480.49 g). IT-97 recorded the lowest value (345.43 g).

4.3.4.3 Fresh weight of leaves

Pooled analysis showed that there was significant difference in fresh weight of leaves between open and shaded conditions at all the three growth stages (Table 39).

At pre-flowering stage, pooled analysis indicated significant difference among the accessions. Maximum pooled mean for fresh weight of leaves (82.44 g) was observed in IT-101. It was on par with IT-108

Table 38. Yield of selected *I. tinctoria* accessions under open and shaded conditions: dry weight of shoot (g)

Sl. No.	Accession No.	Stages of plant growth											
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (240 DAS)					
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	55.87	38.28	47.08	183.85	133.12	158.49	598.87	395.24	497.05			
2	IT-101	58.15	39.35	48.75	179.82	118.69	149.26	628.23	372.78	500.51			
3	IT-96	50.41	31.47	40.94	177.23	137.62	157.42	513.25	382.52	447.89			
4	IT-114	44.84	30.61	37.73	133.03	128.49	130.76	441.59	409.76	425.67			
5	IT-106	36.01	31.56	33.79	174.13	146.31	160.22	571.27	389.70	480.49			
6	IT-104	36.72	28.43	32.57	195.98	124.25	160.12	450.23	362.10	406.17			
7	IT-105	35.46	31.60	33.53	108.26	122.15	115.21	460.59	350.20	405.39			
8	IT-111	41.01	31.49	36.25	176.59	123.76	150.17	410.95	309.81	360.38			
9	IT-97	32.96	31.25	32.10	120.91	109.04	114.98	395.49	295.37	345.43			
10	IT-99	38.67	36.53	37.10	165.51	115.83	140.67	433.13	301.42	367.28			
	Mean	43.01	32.96	-	161.53	125.93	-	490.36	356.89	-			
Pooled analysis		SE	CD		SE	CD		SE	CD		SE	CD	
For light condition		1.53	4.39		6.12	17.56		14.38	41.27		14.38	41.27	
For accessions		3.42	9.81		13.68	39.26		32.15	92.29		32.15	92.29	
For interaction		4.83	13.87		19.34	55.53		45.46	130.51		45.46	130.51	

Table 39. Yield of selected *I. tinctoria* accessions under open and shaded conditions: fresh weight of leaves (g)

Sl. No.	Accession No.	Stages of plant growth											
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (240 DAS)					
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	97.20	60.88	79.04	363.04	284.72	323.88	1175.70	914.25	1044.98			
2	IT-101	101.61	63.26	82.44	331.90	256.41	294.16	1239.53	817.03	1028.28			
3	IT-96	92.46	58.69	75.57	331.21	296.16	313.69	969.96	863.36	916.66			
4	IT-114	92.56	55.33	73.95	273.49	279.13	276.31	794.73	963.93	879.33			
5	IT-106	60.84	55.26	58.05	338.98	308.43	323.71	1060.85	904.37	982.61			
6	IT-104	64.73	51.65	58.19	377.96	275.78	326.87	856.73	764.14	810.44			
7	IT-105	57.74	54.31	56.03	217.64	264.04	240.84	856.73	693.92	775.20			
8	IT-111	76.72	51.89	64.31	316.65	268.00	292.33	749.25	551.38	650.31			
9	IT-97	55.89	51.19	53.54	290.21	237.51	263.86	739.09	512.20	625.65			
10	IT-99	67.09	61.23	64.16	323.28	239.74	281.51	758.32	520.31	639.32			
	Mean	76.68	56.37	-	316.44	270.99	-	920.07	750.49	-			
CD for pooled analysis		SE			SE			SE			SE		
For light condition		3.40			10.03			28.78			33.84		
For accessions		7.59			22.42			64.36			75.66		
For interaction		10.74			31.70			91.01			106.99		
		CD			CD			CD			CD		
		9.75			28.78			97.14			217.20		
		21.80			64.36			307.17			307.17		

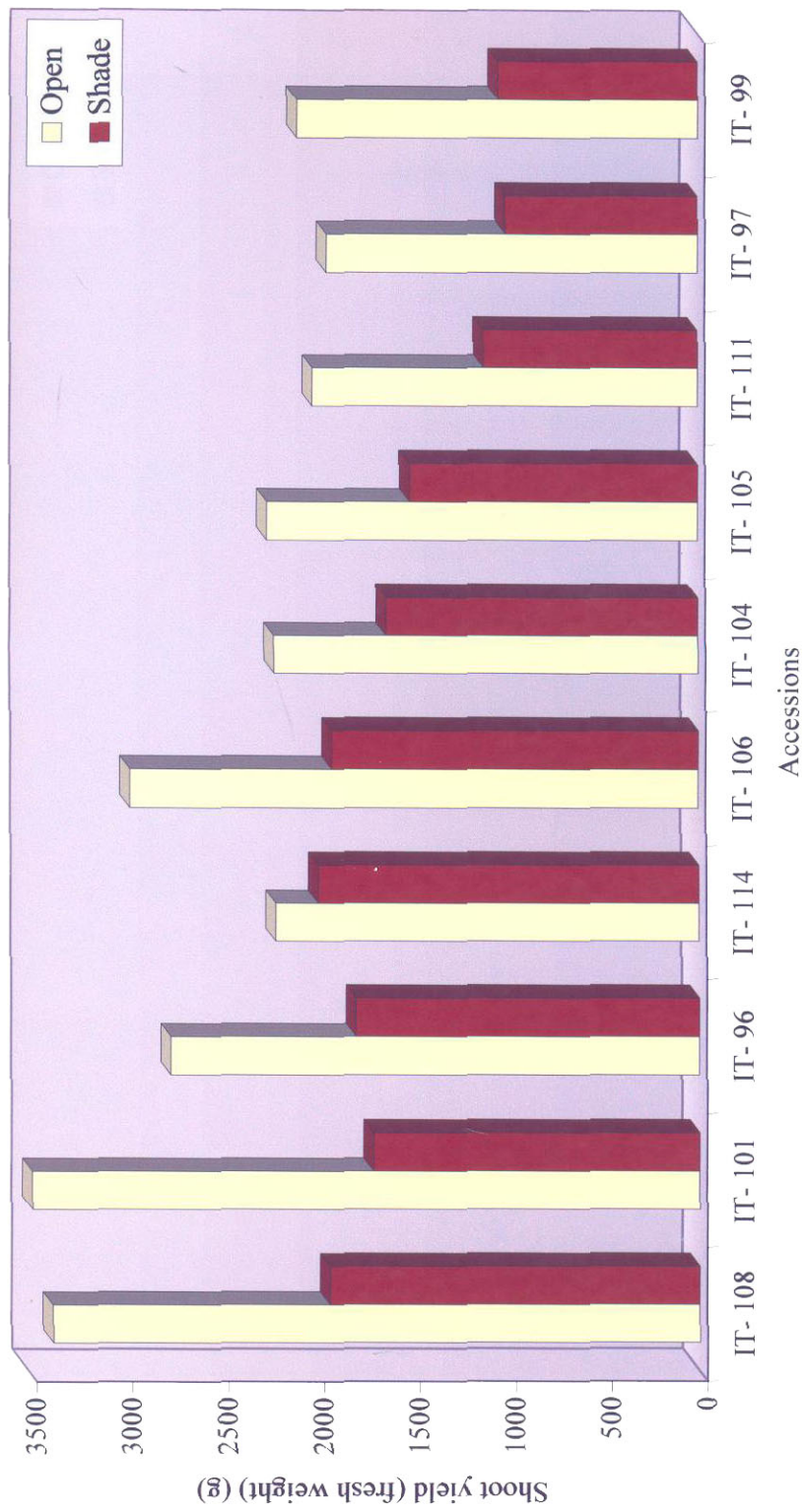


Fig. 5 Shoot yield (fresh weight) (g) of selected *I. tinctoria* accessions at seed maturation stage

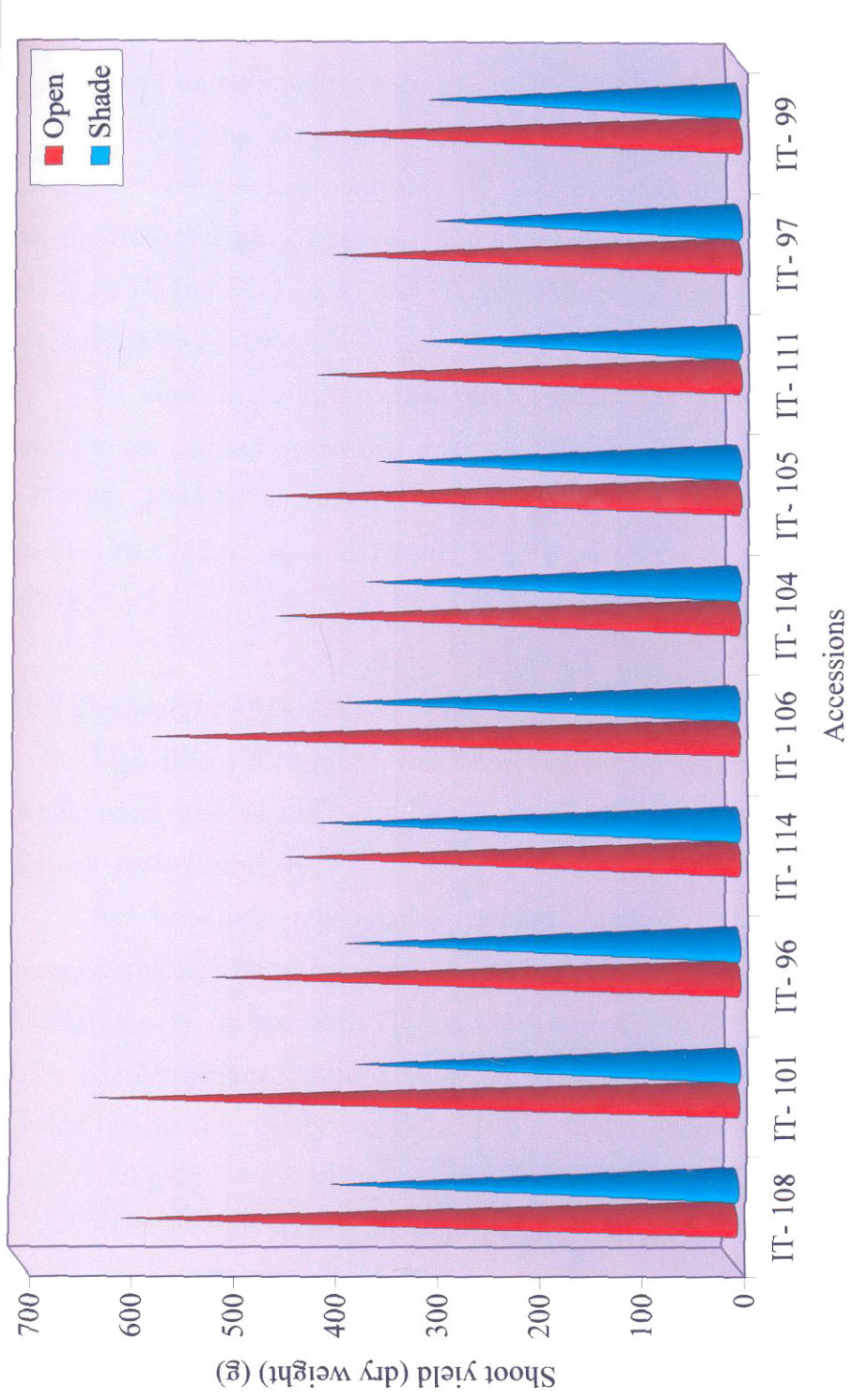


Fig. 6 Shoot yield (dry weight) (g) of selected *I. tinctoria* accessions at seed maturation stage

(79.04 g). IT-96 (75.57 g) and IT-114 (73.95 g). IT-97 recorded the least (53.54 g).

IT-101 recorded the maximum fresh weight of leaves under open (101.61 g) and under shade (63.26 g).

At flowering stage also, accessions showed significant difference among them on pooled analysis. IT-104 recorded the maximum pooled mean for fresh weight of leaves (326.87 g) which was on par with IT-108 (323.88 g), IT-106 (323.71 g) and IT- 96 (313.69 g). Lowest value (240.84 g) was recorded by IT-105.

At seed maturation stage also, there was significant difference among the accessions on pooled analysis (Fig. 7). Pooled mean was highest for IT-108 (1044.98 g) (Plate 5a.) followed by IT-101 (1028.28 g) (Plate 5b.), IT-106 (982.61 g) and IT-96 (916.66 g) (Plate 5c.). It was least for IT-97 (625.65 g).

4.3.4.4 Dry weight of leaves

Significant difference was observed in dry weight of leaves/ plant between open and shaded conditions at all the three growth stages, on pooled analysis (Table 40).

At pre-flowering stage, pooled analysis showed significant difference among the accessions. Pooled mean was maximum for IT-101 (30.12 g). It was on par with IT-108 (28.47 g), IT-96 (25.48 g) and IT-114 (24.39 g). Minimum value (17.58 g) was recorded by IT-97. IT-101 recorded the maximum dry weight of leaves under open (38.74 g) and under shade (21.50 g).

At flowering stage also, significant difference was observed among the accessions. IT-104 recorded the maximum pooled mean (113.29 g) followed by IT-108 (110.95 g), IT-106 (106.73 g) and IT-96 (104.52 g). IT-105 recorded the minimum leaf dry weight (83.82 g).

At seed maturation stage also, accessions showed significant difference for dry weight of leaves (Fig. 8). Maximum pooled mean (369.73 g)

Table 40. Yield of selected *I. tinctoria* accessions under open and shaded conditions: dry weight of leaves (g)

Sl. No.	Accession No.	Stages of plant growth											
		Pre-flowering (90 DAS)				Flowering (150 DAS)				Seed maturation (240 DAS)			
		Open	Shade	Mean	CD	Open	Shade	Mean	SE	Open	Shade	Mean	CD
1	IT-108	36.33	20.62	28.47	122.36	99.54	110.95		424.51	314.95	369.73		
2	IT-101	38.74	21.50	30.12	107.69	86.77	97.23		429.28	288.54	358.91		
3	IT-96	31.24	19.72	25.48	107.35	101.68	104.52		339.03	300.41	319.72		
4	IT-114	31.17	17.62	24.39	96.32	97.86	97.09		278.12	325.73	301.93		
5	IT-106	19.78	18.56	19.17	110.87	102.59	106.73		365.08	322.81	343.94		
6	IT-104	21.57	16.96	19.27	131.08	95.50	113.29		299.43	269.99	284.72		
7	IT-105	18.66	18.35	18.51	81.67	85.97	83.82		296.65	227.36	262.01		
8	IT-111	25.46	17.72	21.59	106.12	92.30	99.21		255.05	190.61	222.83		
9	IT-97	18.34	16.82	17.58	99.96	79.52	89.74		250.43	179.92	215.18		
10	IT-99	21.71	20.73	21.22	110.10	79.55	94.83		258.66	185.63	222.14		
	Mean	26.30	18.86	-	107.35	92.13	-		319.62	260.60	-		
	Pooled analysis		SE	CD		SE	CD		SE	CD			
	For light condition		1.48	4.24		2.95	8.48		11.21	32.20			
	For accessions		3.30	9.48		6.60	18.96		25.08	71.99			
	For interaction		4.67	13.41		9.34	26.82		35.46	101.81			



Fig. 7 Leaf yield (fresh weight) (g) of selected *I. tinctoria* accessions at seed maturation stage

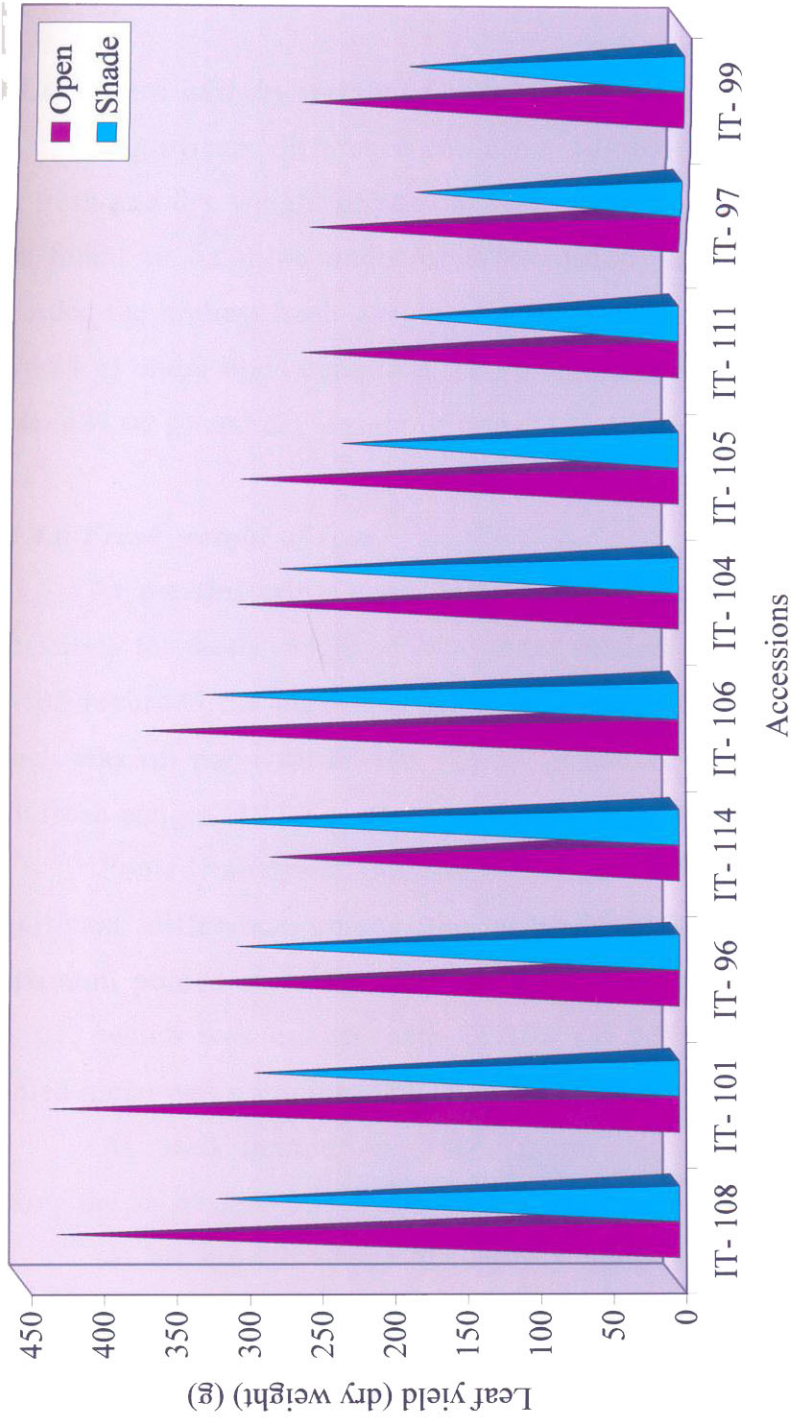


Fig. 8 Leaf yield (dry weight) (g) of selected *I. tinctoria* accessions at seed maturation stage

was observed in IT-108, which was on par with IT-101 (358.91 g), IT-106 (343.94 g) and IT-96 (319.72 g). Minimum pooled mean (215.18 g) was observed in IT-97.

4.3.4.5 Fresh and dry weight of pods

Significant difference could not be observed among the accessions for fresh and dry weight of pods at seed maturation stage (Table 41). But it was found to be more under open condition compared to shade. IT-101 recorded the highest fresh weight of pods (753.28 g) and dry weight of pods (226.88 g) under open condition. IT-96 recorded the highest fresh weight of pods (484.09 g) and dry weight of pods (155.56 g) under shade.

4.3.4.6 Fresh weight of root

At pre-flowering stage, there was significant difference among the accessions for fresh weight of root under shaded condition only (Table 42). IT-108 recorded the maximum fresh weight of root (17.85 g) under shade, which was on par with IT-101 (16.57 g) and IT-106 (14.64 g). Minimum root fresh weight (10.94 g) was recorded by IT-104.

Pooled analysis indicated that at flowering stage there was significant difference among the accessions for fresh weight of root. Maximum pooled mean for fresh weight of root (36.10 g) was observed in IT-101, which was on par with IT-108 (35.17 g) and IT-105 (35.13 g). Pooled mean was minimum (27.31 g) for IT-106.

At seed maturation stage, there was no significant difference among the accessions under both conditions (Fig. 9).

At all growth stages, the accessions recorded higher fresh weight of root under open condition compared to shade.

4.3.4.7 Dry weight of root

At pre-flowering stage, significant difference was there among the accessions both under open and shaded conditions (Table 43). Under open

Table 41. Yield of selected *I. tinctoria* accessions at seed maturation stage (240 DAS) under open and shaded conditions: fresh and dry weight of pods (g)

Sl. No.	Accession No.	Fresh weight (g)		Dry weight (g)	
		Open	Shade	Open	Shade
1	IT- 108	724.14	450.40	214.39	149.96
2	IT- 101	753.28	393.03	226.88	130.51
3	IT- 96	647.92	484.09	176.16	155.56
4	IT- 114	560.36	413.61	160.81	136.91
5	IT- 106	612.75	412.99	173.12	127.14
6	IT- 104	574.09	413.75	172.82	141.57
7	IT- 105	509.28	360.89	138.82	120.33
8	IT- 111	628.36	439.13	210.87	140.51
9	IT- 97	694.49	367.24	203.95	134.41
10	IT- 99	577.82	397.11	171.67	142.98
	SE	76.52	50.64	25.66	15.34
	CD	NS	NS	NS	NS

Table 42. Yield of selected *I. tinctoria* accessions under open and shaded conditions: fresh weight of root (g)

Sl. No.	Accession No.	Stages of plant growth						
		Pre-flowering (90 DAS)		Flowering (150 DAS)			Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Mean	Open	Shade
1	IT- 108	34.92	17.85	46.00	24.33	35.17	134.34	37.60
2	IT- 101	28.64	16.57	44.36	27.83	36.10	121.91	33.41
3	IT- 96	29.58	13.08	32.00	25.39	28.70	114.58	39.59
4	IT- 114	29.03	12.27	41.00	24.63	32.82	86.36	37.71
5	IT- 106	25.99	14.64	33.74	20.87	27.31	91.00	34.85
6	IT- 104	23.68	10.94	36.81	23.18	30.00	126.32	38.93
7	IT- 105	24.85	12.32	47.18	23.07	35.13	101.17	37.31
8	IT- 111	29.92	12.80	48.04	20.49	34.27	95.75	33.88
9	IT- 97	28.17	13.77	37.41	23.71	30.56	117.64	34.40
10	IT- 99	29.08	11.98	43.17	24.64	33.91	105.31	34.83
	Mean	28.39	13.62	40.97	23.81	-	109.44	36.25
	SE	2.03	1.14	-	-	-	18.75	1.48
	CD	NS	3.38	-	-	-	NS	NS
Pooled analysis				CD		SE		
For light condition				1.36		3.91		
For accessions				3.04		8.74		
For interaction				4.31		12.36		

Table 43. Yield of selected *I. tinctoria* accessions under open and shaded conditions: dry weight of root (g)

Sl. No.	Accession No.	Stages of plant growth									
		Pre-flowering (90 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)		Flowering (150 DAS)		Seed maturation (240 DAS)	
		Open	Shade	Open	Shade	Open	Shade	Open	Shade	Open	Shade
1	IT-108	17.95	9.24	23.77	12.99	18.38	17.87	78.41	17.87	48.14	
2	IT-101	14.43	8.12	22.50	14.03	18.26	17.86	70.42	17.86	44.14	
3	IT-96	14.55	6.34	15.99	13.17	14.58	20.63	63.49	20.63	42.06	
4	IT-114	14.70	5.97	21.33	12.69	17.01	18.80	44.00	18.80	31.40	
5	IT-106	12.97	6.74	16.79	10.45	13.62	17.72	48.30	17.72	33.01	
6	IT-104	11.05	5.59	16.99	12.34	14.66	20.61	49.24	20.61	34.93	
7	IT-105	12.10	6.47	24.64	11.31	17.97	19.89	41.64	19.89	30.76	
8	IT-111	15.35	6.23	23.88	10.37	17.13	17.68	35.11	17.68	26.39	
9	IT-97	13.77	6.48	18.69	12.25	15.47	17.97	74.84	17.97	46.40	
10	IT-99	14.74	5.87	22.71	12.68	17.70	17.83	42.54	17.83	30.19	
	Mean	14.16	6.71	20.73	12.23	-	18.69	54.80	-	-	
	SE	1.16	0.67	-	-	-	-	-	-	-	
	CD	3.45	1.99	-	-	-	-	-	-	-	
Pooled analysis				SE		CD		SE		CD	
For light condition				0.79		2.26		3.52		10.11	
For accessions				1.76		5.06		7.88		22.62	
For interaction				2.49		7.16		11.14		31.69	

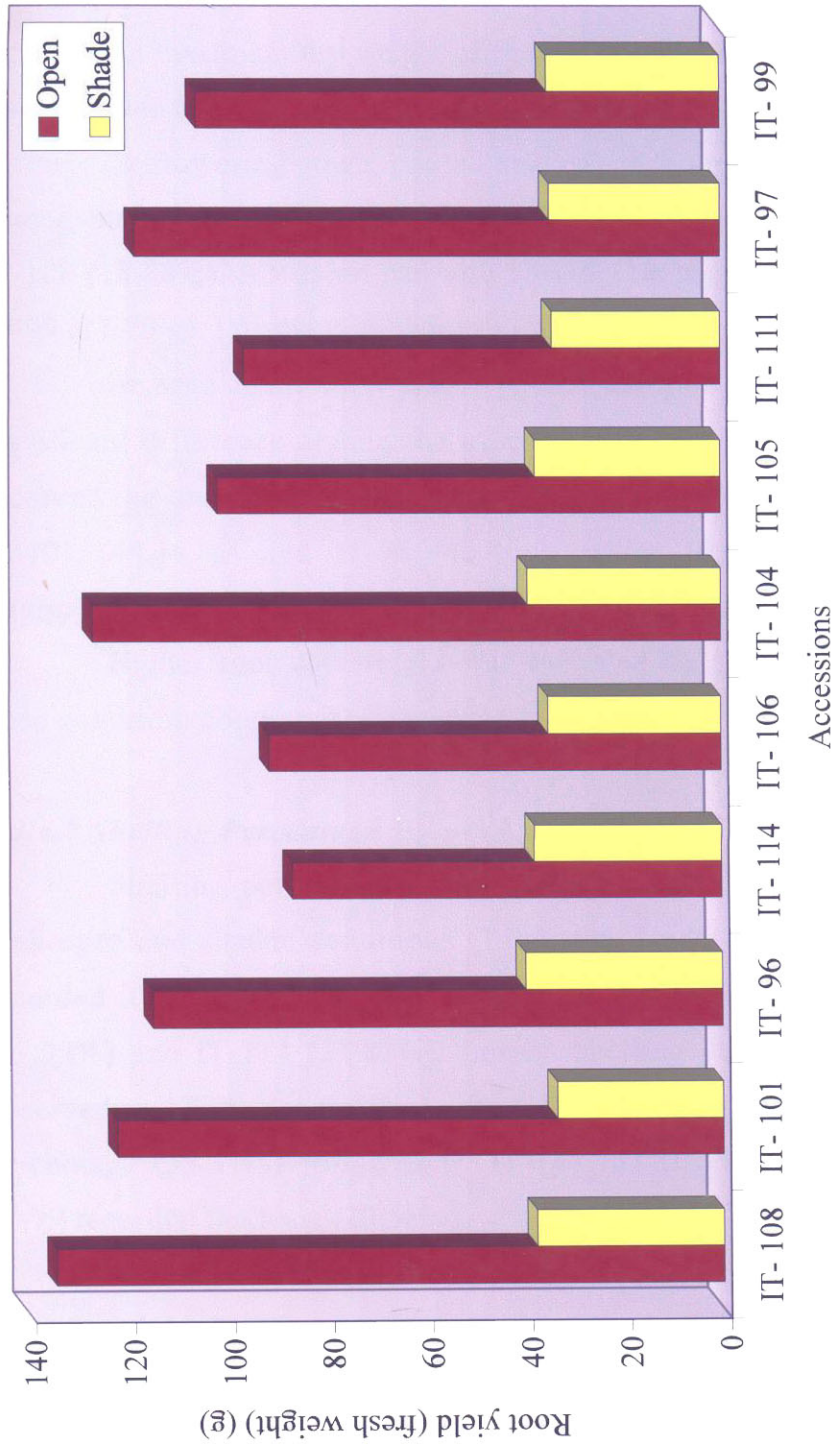


Fig. 9 Root yield (fresh weight) (g) of selected *I. tinctoria* accessions at seed maturation stage

condition, dry weight of root was maximum for IT-108 (17.95 g), which was on par with IT-111 (15.35 g), IT-99 (14.74 g) and IT-114 (14.70 g). Dry weight of root was minimum for IT-104 (11.05 g). Under shade, IT-108 recorded the maximum dry weight of root (9.24 g) followed by IT-101 (8.12 g). Lowest value (5.59 g) was observed in IT-104.

At flowering stage, pooled analysis indicated significant difference among the accessions for dry weight of root. Pooled mean was highest in IT-108 (18.38 g). It was on par with IT-101 (18.26 g), IT-105 (17.97 g) and IT-99 (17.70 g). Lowest pooled mean (13.62 g) was observed in IT-106.

At seed maturation stage, pooled analysis showed that there was significant difference among the accessions for dry weight of root. IT-108 recorded the maximum pooled mean (48.14 g) followed by IT-97 (46.40 g), IT-101 (44.14 g) and IT-96 (42.06 g) (Fig. 10). IT-111 recorded the minimum pooled mean (26.39 g).

Higher root dry weight was recorded by all the accessions under open condition compared to shade.

4.3.4.8 Shelling Percentage

Shelling percentage of the accessions differed significantly under both open and shaded conditions (Table 44). Under open condition, IT-104 recorded the highest shelling percentage (33.81%) followed by IT-108 (31.95%) and IT-114 (31.83%). Lowest shelling percentage (23.27%) was observed in IT-99. Under shade also, IT-104 recorded the highest shelling percentage (33.31%) followed by IT-114 (31.16%) and IT-108 (31.15%). IT-99 recorded the least (23.58%).

4.3.5 Physiological Parameters

4.3.5.1 Leaf area index (LAI)

Pooled analysis indicated that there was significant difference in LAI between open and shaded conditions at all the three growth stages (Table 45).

Table 44. Shelling percentage (%) of selected *I. tinctoria* accessions at seed maturation stage (240 DAS) under open and shaded conditions

Sl. No.	Accession No.	Open	Shade
1	IT- 108	31.95	31.15
2	IT- 101	28.58	28.38
3	IT- 96	26.62	26.46
4	IT- 114	31.83	31.16
5	IT- 106	30.06	29.95
6	IT- 104	33.81	33.31
7	IT- 105	28.63	28.46
8	IT- 111	31.43	30.24
9	IT- 97	25.68	25.70
10	IT- 99	23.27	23.58
	SE	0.74	0.75
	CD	2.19	2.24

Table 45. Leaf area index of selected *I. tinctoria* accessions under open and shaded conditions

Sl. No.	Accession No.	Stages of plant growth											
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (90 DAS)					
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	1.08	0.68	0.88	3.54	2.00	2.77	8.59	7.22	7.91			
2	IT-101	1.09	0.70	0.90	3.42	1.94	2.68	8.69	6.84	7.77			
3	IT-96	1.00	0.61	0.81	3.42	2.07	2.74	8.24	6.79	7.51			
4	IT-114	1.00	0.63	0.82	2.79	2.26	2.53	7.81	8.57	8.19			
5	IT-106	0.66	0.61	0.64	3.45	2.56	3.00	8.32	7.66	7.99			
6	IT-104	0.71	0.64	0.68	3.62	1.84	2.73	7.94	6.42	7.18			
7	IT-105	0.64	0.50	0.57	2.46	2.36	2.41	7.92	6.12	7.02			
8	IT-111	0.86	0.59	0.72	3.39	1.87	2.63	7.51	6.01	6.76			
9	IT-97	0.63	0.60	0.62	2.94	1.61	2.28	7.51	5.16	6.33			
10	IT-99	0.72	0.66	0.69	3.44	1.36	2.40	7.64	5.25	6.44			
	Mean	0.84	0.62	-	3.25	1.99	-	8.02	6.60	-			
	Pooled analysis		SE	CD		SE	CD		SE	CD			
	For light condition		3.62	0.10		0.13	0.38		0.20	0.58			
	For accessions		8.09	0.23		0.30	0.85		0.46	1.31			
	For interaction		0.11	0.33		0.42	1.20		0.64	1.85			

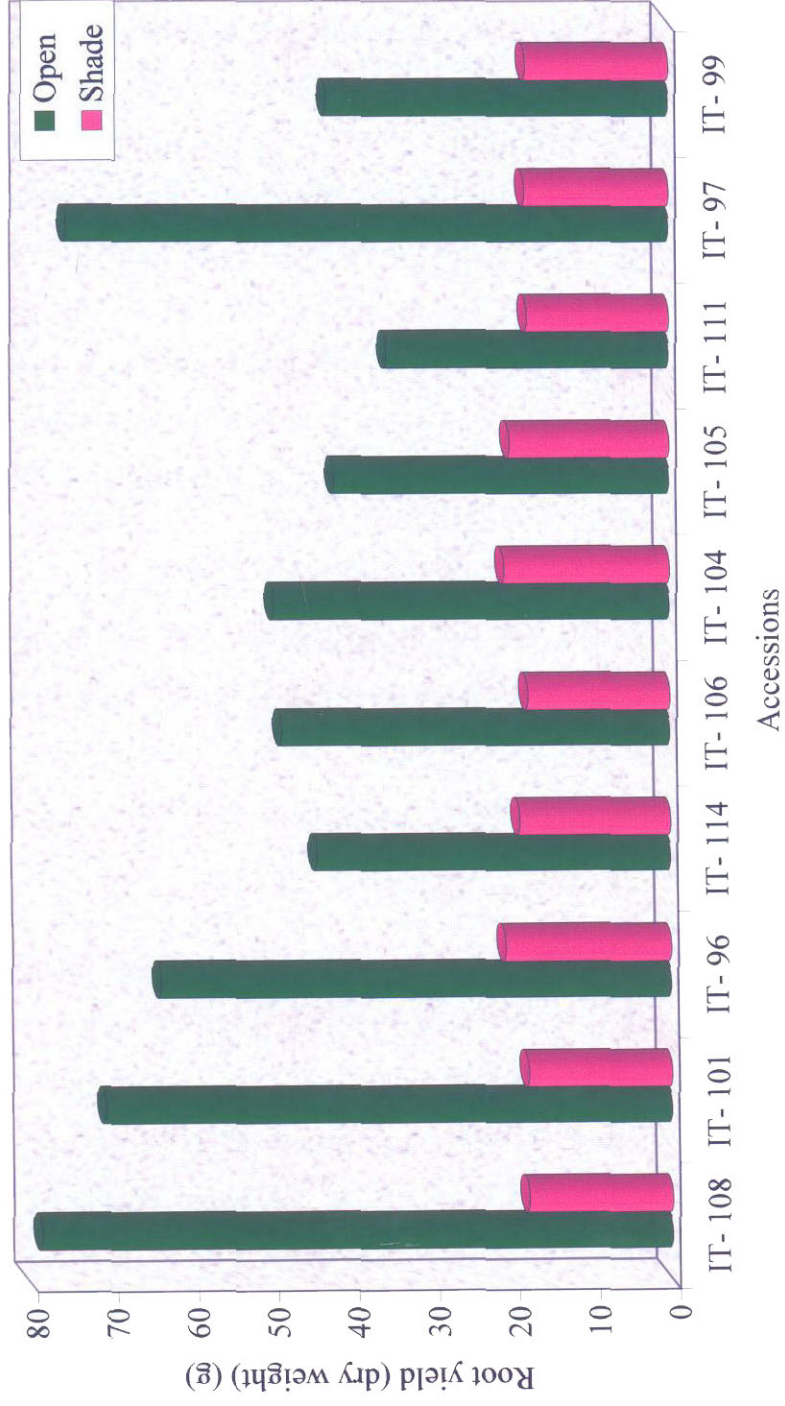


Fig. 10 Root yield (dry weight) (g) of selected *I. tinctoria* accessions at seed maturation stage

At pre-flowering stage, there was significant difference for LAI among the accessions. Maximum pooled mean (0.9) was observed for IT-101, which was on par with IT-108 (0.88), IT-114 (0.82) and IT-96 (0.81). Pooled mean was minimum (0.57) for IT-105.

At flowering stage also, significant difference was observed among the accessions for LAI. IT-106 recorded the maximum pooled mean (3.0) followed by IT-108 (2.77), IT-96 (2.74) and IT-104 (2.73). Minimum pooled mean (2.28) was observed for IT-97.

At seed maturation stage also, accessions showed significant difference among them for LAI. Pooled mean was maximum (8.19) for IT-114. It was on par with IT-106 (7.99), IT-108 (7.91) and IT-101 (7.77). Least value (6.33) was recorded by IT-97.

4.3.5.2 Leaf area duration (LAD)

There was significant difference in LAD between open and shaded conditions at the two periods on pooled analysis (Table 46).

During the period from pre-flowering to flowering stage (period 1), there was significant difference among the accessions for LAD. Maximum pooled mean (3.65 days) was observed in IT-108. It was on par with IT-106 (3.64 days), IT-101 (3.58 days) and IT-96 (3.55 days). Pooled mean was minimum for IT-97 (2.89 days).

For the period from flowering to seed maturation stage (period 2) also, significant difference was observed among the accessions for LAD. IT-106 recorded the maximum pooled mean (16.50 days) followed by IT-114 (16.08 days) and IT-108 (16.02 days). IT-97 recorded the minimum pooled mean (12.92 days).

4.3.5.3 Net assimilation rate (NAR)

Pooled analysis showed significant difference in NAR between open and shaded conditions at the two periods (Table 47).

Table 46. Leaf area duration (days) of *I. tinctoria* accessions under open and shaded conditions

Sl. No.	Accession No.	Period 1 (90-150 DAS)			Period 2 (150-240 DAS)		
		Open	Shade	Mean	Open	Shade	Mean
1	IT- 108	4.62	2.68	3.65	18.20	13.83	16.02
2	IT- 101	4.51	2.64	3.58	18.17	13.18	15.68
3	IT- 96	4.42	2.68	3.55	17.48	13.28	15.38
4	IT- 114	3.79	2.89	3.34	15.91	16.24	16.08
5	IT- 106	4.10	3.17	3.64	17.66	15.34	16.50
6	IT- 104	4.33	2.47	3.40	17.31	12.38	14.85
7	IT- 105	3.09	2.85	2.97	15.57	12.72	14.14
8	IT- 111	4.25	2.46	3.35	16.36	11.81	14.09
9	IT- 97	3.57	2.21	2.89	15.67	10.16	12.92
10	IT- 99	4.16	2.02	3.09	16.61	9.92	13.27
	Mean	4.09	2.61	-	16.90	12.89	-
Pooled analysis			SE	CD		SE	CD
For light condition			0.14	0.39		0.44	1.26
For accessions			0.30	0.87		0.98	2.82
For interaction			0.43	1.23		1.39	3.99

Table 47. Physiological parameters of selected *I. tinctoria* accessions under open and shaded conditions: NAR and CGR

Sl. No.	Accession No.	Net assimilation rate (NAR) ($\text{gm}^2\text{day}^{-1}$)						Crop growth rate (CGR) ($\text{gm}^2\text{day}^{-1}$)					
		Period 1 (90-150 DAS)			Period 2 (150-240 DAS)			Period 1 (90-150 DAS)			Period 2 (90-150 DAS)		
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	4.56	5.82	5.19	3.75	3.12	3.44	10.53	7.80	9.17	22.77	14.38	18.56
2	IT-101	4.44	4.95	4.69	4.05	3.17	3.61	10.01	6.53	8.27	24.60	13.94	19.27
3	IT-96	4.73	6.52	5.63	3.16	3.04	3.10	10.44	8.74	9.59	18.43	13.44	15.94
4	IT-114	3.83	5.58	4.70	3.19	2.85	3.02	7.26	8.06	7.66	16.92	15.43	16.18
5	IT-106	5.54	5.95	5.75	3.71	2.61	3.16	11.37	9.44	10.41	21.79	13.35	17.57
6	IT-104	6.05	6.38	6.22	2.41	3.16	2.79	13.11	7.88	10.50	13.95	13.05	13.50
7	IT-105	3.87	5.22	4.55	3.72	2.95	3.34	5.99	7.45	6.72	19.33	12.51	15.92
8	IT-111	5.25	6.19	5.72	2.35	2.59	2.47	11.16	7.59	9.38	12.86	10.21	11.53
9	IT-97	4.05	5.78	4.92	2.88	3.02	2.95	7.24	6.40	6.82	15.06	10.22	12.64
10	IT-99	5.02	6.53	5.78	2.65	3.08	2.87	10.44	6.61	8.53	14.68	10.18	12.43
	Mean	4.74	5.89	-	3.19	2.96	-	9.76	7.65	-	18.04	12.67	-
Pooled analysis		SE			CD	SE			CD	SE			CD
For light condition		0.13			0.37	0.14			0.39	0.47			1.35
For accessions		0.29			0.83	0.31			0.88	1.06			3.03
For interaction		0.41			1.17	0.43			1.25	1.49			4.28

During the period from pre-flowering to flowering stage (period1), the accessions showed significant difference among them for NAR. Pooled mean was maximum ($6.22 \text{ g m}^{-2} \text{ day}^{-1}$) for IT-104, which was on par with IT-99 ($5.78 \text{ g m}^{-2} \text{ day}^{-1}$), IT-106 ($5.75 \text{ g m}^{-2} \text{ day}^{-1}$) and IT-111 ($5.72 \text{ g m}^{-2} \text{ day}^{-1}$): Minimum pooled mean ($4.55 \text{ g m}^{-2} \text{ day}^{-1}$) was observed in IT-105.

For the period from flowering to seed maturation stage (period 2) also, significant difference was observed among the accessions for NAR. Maximum pooled mean ($3.61 \text{ g m}^{-2} \text{ day}^{-1}$) was recorded by IT-101 followed by IT-108 ($3.44 \text{ g m}^{-2} \text{ day}^{-1}$), IT-105 ($3.34 \text{ g m}^{-2} \text{ day}^{-1}$) and IT-106 ($3.16 \text{ g m}^{-2} \text{ day}^{-1}$). Pooled mean was lowest for IT-111 ($2.47 \text{ g m}^{-2} \text{ day}^{-1}$).

4.3.5.4 Crop growth rate (CGR)

Significant difference was observed between open and shaded conditions at both periods for CGR on pooled analysis (Table 47).

During the period from pre-flowering to flowering stage (period1), there was significant difference among the accessions for CGR. IT-104 recorded the maximum pooled mean ($10.50 \text{ g m}^{-2} \text{ day}^{-1}$) followed by IT-106 ($10.41 \text{ g m}^{-2} \text{ day}^{-1}$), IT-96 ($9.59 \text{ g m}^{-2} \text{ day}^{-1}$) and IT-111 ($9.38 \text{ g m}^{-2} \text{ day}^{-1}$). IT-105 recorded the minimum pooled mean ($6.72 \text{ g m}^{-2} \text{ day}^{-1}$).

For the period from flowering to seed maturation stage (period 2) also, significant difference was observed among the accessions for CGR. Pooled mean was maximum ($19.27 \text{ g m}^{-2} \text{ day}^{-1}$) for IT-101, IT-108 ($18.56 \text{ g m}^{-2} \text{ day}^{-1}$), IT-106 ($17.57 \text{ g m}^{-2} \text{ day}^{-1}$) and IT-114 ($16.18 \text{ g m}^{-2} \text{ day}^{-1}$). Minimum pooled mean ($12.43 \text{ g m}^{-2} \text{ day}^{-1}$) was observed in IT-99.

4.3.5.5 Absolute growth rate (AGR)

There was significant difference in AGR between open and shaded conditions at both the periods on pooled analysis (Table 48).

During the period from pre-flowering to flowering stage (period1), the accessions showed significant difference among them for AGR. Maximum pooled mean (2.13 g day^{-1}) was recorded by IT-104. It was on

par with IT-106 (2.11 g day⁻¹), IT-96 (1.94 g day⁻¹) and IT-111 (1.9 g day⁻¹). Minimum pooled mean (1.38 g day⁻¹) was recorded by IT-97.

For the period from flowering to seed maturation stage (period 2) also, significant difference occurred among the accessions for AGR. IT-101 recorded the maximum pooled mean (3.90 g day⁻¹) followed by IT-108 (3.77 g day⁻¹), IT-106 (3.56 g day⁻¹) and IT-114 (3.28 g day⁻¹). Pooled mean was minimum (2.34 g day⁻¹) for IT-111.

4.3.5.6 Relative growth rate (RGR)

Pooled analysis showed significant difference in RGR between open and shaded conditions at the two periods (Table 48).

During the period from pre-flowering to flowering stage (period1), there was significant difference among the accessions for RGR. Pooled mean was maximum (0.026 g day⁻¹) for IT-106 and IT-104 (0.026 g day⁻¹). They were on par with IT-111 (0.024 g day⁻¹) and IT-96 (0.023 g day⁻¹). Minimum pooled mean (0.019 g day⁻¹) was recorded by IT-101.

For the period from flowering to seed maturation stage (period 2) also, the accessions showed significant difference among them for RGR. IT-101 and IT-105 recorded the maximum pooled mean (0.014 g day⁻¹) followed by IT-108 (0.013 g day⁻¹) and IT-114 (0.013 g day⁻¹). IT-104 and IT-111 recorded the minimum value (0.010 g day⁻¹) for RGR.

4.3.5.7 Dry matter accumulation per plant

Significant difference was observed between open and shaded conditions for dry matter accumulation per plant at all the three growth stages on pooled analysis (Table 49).

At pre-flowering stage, there was significant difference for dry matter accumulation among the accessions. Maximum pooled mean (60.67 g plant⁻¹) was observed for IT-108, which was on par with IT-101 (60.02 g plant⁻¹) and IT-96 (51.38 g plant⁻¹). Pooled mean was minimum (40.90 g plant⁻¹) for IT-104.

Table 49. Physiological parameters of selected *L. tinctoria* accessions under open and shaded conditions: dry matter accumulation per plant (g plant⁻¹)

Sl. No.	Accession No.	Stages of plant growth											
		Pre-flowering (90 DAS)			Flowering (150 DAS)			Seed maturation (240 DAS)					
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	73.81	47.52	60.67	207.62	146.11	176.86	891.67	563.07	727.37			
2	IT-101	72.58	47.47	60.02	202.32	132.72	167.52	925.53	521.15	723.34			
3	IT-96	64.96	37.81	51.38	193.22	150.79	172.00	752.91	558.71	655.81			
4	IT-114	59.54	36.58	48.06	154.37	141.18	147.77	646.40	565.46	605.93			
5	IT-106	48.99	38.30	43.64	190.92	156.76	173.84	792.70	534.56	663.63			
6	IT-104	47.77	34.02	40.90	212.97	136.59	174.78	672.28	524.28	598.28			
7	IT-105	47.56	38.08	42.82	132.90	133.79	133.35	641.05	490.47	565.76			
8	IT-111	56.35	37.72	47.04	200.47	134.13	167.30	656.93	468.00	562.47			
9	IT-97	46.73	37.73	42.23	139.60	121.29	130.45	674.27	447.76	561.01			
10	IT-99	53.41	41.40	47.40	188.22	128.50	158.36	647.34	462.23	554.79			
	Mean	57.17	39.66	-	182.26	138.19	-	730.11	513.57	-			
Pooled analysis			SE	CD		SE	CD		SE	CD			
For light condition			1.64	4.72		6.00	17.22		21.00	60.28			
For accessions			3.68	10.55		13.42	38.51		46.95	134.78			
For interaction			5.20	14.93		18.97	54.47		66.40	190.61			

At flowering stage also, significant difference was observed among the accessions for dry matter accumulation per plant. IT-108 recorded the maximum pooled mean (176.86 g plant⁻¹) followed by IT-104 (174.78 g plant⁻¹), IT-106 (173.84 g plant⁻¹) and IT-96 (172 g plant⁻¹). Minimum pooled mean (130.45 g plant⁻¹) was observed for IT-97.

At seed maturation stage also, accessions showed significant difference among them for dry matter accumulation. Pooled mean was maximum (727.37 g plant⁻¹) for IT-108. It was on par with IT-101 (723.34 g plant⁻¹), IT-106 (663.63 g plant⁻¹) and IT-96 (655.81 g plant⁻¹). Least value (554.79 g plant⁻¹) was recorded by IT-99.

4.3.5.8 Harvest index

Leaf yield

Pooled analysis showed significant difference in leaf yield between open and shaded conditions and among the accessions (Table 50). Maximum pooled mean (0.53) was observed for IT-106. It was on par with IT-108 (0.52) and IT-101 (0.51). Pooled mean was minimum (0.39) for IT-97.

Root yield

There was significant difference in root yield between open and shaded conditions and also among the accessions on pooled analysis (Table 50). IT-97 recorded the maximum pooled mean (0.08) followed by IT-108 (0.06), IT-101 (0.06), IT-96 (0.06), IT-106 (0.06) and IT-105 (0.06).

Leaf + root yield

Significant difference was observed between open and shaded conditions and among the accessions on pooled analysis (Table 50). Pooled mean was maximum (0.58) for IT-108 and IT-106 (0.58), which were on par with IT-101 (0.56), IT-96 (0.56) and IT-114 (0.56). IT-111 recorded the minimum pooled mean (0.45).

Table 50. Harvest index (HI)* of the official parts of selected *I. tinctoria* accessions on per plant basis at seed maturation stage under open and shaded conditions

Sl. No.	Accession No.	Leaf			Root			Leaf + Root		
		Open	Shade	Mean	Open	Shade	Mean	Open	Shade	Mean
1	IT-108	0.48	0.56	0.52	0.10	0.03	0.06	0.56	0.59	0.58
2	IT-101	0.47	0.55	0.51	0.08	0.03	0.06	0.54	0.59	0.56
3	IT-96	0.45	0.54	0.50	0.08	0.04	0.06	0.54	0.58	0.56
4	IT-114	0.43	0.58	0.50	0.07	0.04	0.05	0.50	0.61	0.56
5	IT-106	0.46	0.61	0.53	0.08	0.04	0.06	0.52	0.64	0.58
6	IT-104	0.45	0.52	0.48	0.07	0.04	0.05	0.52	0.56	0.54
7	IT-105	0.46	0.46	0.46	0.07	0.01	0.06	0.53	0.51	0.52
8	IT-111	0.39	0.41	0.40	0.05	0.04	0.05	0.45	0.45	0.45
9	IT-97	0.38	0.40	0.39	0.11	0.04	0.08	0.48	0.44	0.46
10	IT-99	0.40	0.40	0.40	0.06	0.04	0.05	0.47	0.45	0.46
	Mean	0.44	0.50	-	0.08	0.04	-	0.51	0.54	-
	Pooled analysis		SE	CD		SE	CD		SE	CD
	For light condition		0.012	0.035		0.004	0.012		0.012	3.39
	For accessions		0.027	0.078		0.009	0.026		0.026	7.59
	For interaction		0.038	0.110		0.013	0.037		0.037	0.11

Dry weight of official part

HI* =

Dry weight of whole plant

4.3.6 Phytochemical analysis

4.3.6.1 Soil analysis before and after the experiment for content of N, P and K in the experimental field

4.3.6.1.1 Soil nitrogen

Before the commencement of the experiment, the soil analysis revealed a value of 213.25 kg ha⁻¹ of nitrogen under open and 210.11 kg ha⁻¹ of nitrogen under shaded condition.

After the experiment, under open condition, soil nitrogen was the highest in the plot where IT-105 was grown (Table 51). The nitrogen content was 451.58 kg ha⁻¹. This was followed by the plot where IT-101 was grown. It recorded a value of 429.63 kg ha⁻¹. The least was shown by the plot where IT-97 (223.84 kg ha⁻¹) was grown.

In shade, the plot where IT-106 was grown showed the highest nitrogen content (257.15 kg ha⁻¹) followed by IT-99 (250.88 kg ha⁻¹). The lowest soil nitrogen content was recorded in the plot where IT-97 (213.07 kg ha⁻¹) was grown.

4.3.6.1.2 Soil phosphorus content

Before the commencement of the experiment, phosphorous content of the soil was observed to be 57.51 kg ha⁻¹ and 40.46 kg ha⁻¹ under open and shaded conditions respectively.

After the experiment, under open condition, the plot of IT-106 showed the highest phosphorus content of 51.97 kg ha⁻¹ followed by the plot of IT-99 (46.36 kg ha⁻¹) (Table 51). The lowest soil phosphorous content (27.92 kg ha⁻¹) was observed in the plot of IT-108.

Under shade, the soil phosphorus was found to be the highest (35.15 kg ha⁻¹) in the plot of IT-96 followed by the plot of IT-114 (31.08 kg ha⁻¹). The plot where IT-101 was grown recorded the lowest phosphorus content of 22.62 kg ha⁻¹.

Table 51. NPK status (kg ha^{-1}) of the soil of experimental field at final harvest (240 DAS) of selected *I. tinctoria* accessions

Sl. No.	Accession No.	Nitrogen		Phosphorus		Potassium	
		Open	Shade	Open	Shade	Open	Shade
1	IT- 108	333.76	228.93	27.92	22.83	70.00	60.00
2	IT- 101	429.63	250.88	29.56	22.62	80.00	60.00
3	IT- 96	404.54	231.30	32.10	35.15	140.00	70.00
4	IT- 114	263.42	244.61	42.29	31.08	90.00	60.00
5	IT- 106	373.18	228.75	51.97	24.05	130.00	80.00
6	IT- 104	291.65	228.75	40.76	27.72	120.00	80.00
7	IT- 105	451.58	257.15	40.76	24.86	120.00	70.00
8	IT- 111	275.97	247.74	34.14	25.47	70.00	60.00
9	IT- 97	223.84	213.07	43.82	23.23	100.00	100.00
10	IT- 99	272.83	216.98	46.36	29.35	90.00	80.00

Table 52. Nitrogen content (%)^{*} of selected *I. tinctoria* accessions at seed maturation stage under open and shaded conditions

Sl. No.	Accession No.	Seed maturation stage (240 DAS)		
		Open	Shade	Mean
1	IT- 108	5.04	3.87	4.45
2	IT- 101	4.20	4.03	4.12
3	IT- 96	4.54	3.87	4.20
4	IT- 114	3.75	3.64	3.70
5	IT- 106	3.98	3.90	3.94
6	IT- 104	3.19	2.74	2.97
7	IT- 105	4.26	4.09	4.17
8	IT- 111	4.42	2.86	3.64
9	IT- 97	4.43	3.19	3.81
10	IT- 99	4.09	3.75	3.92
	Mean	4.19	3.59	-
Pooled analysis			SE	CD
For light condition			0.12	0.35
For accessions			0.27	0.78
For interaction			0.38	0.10

* - Sample consists of leaves alone

4.3.6.1.3 Soil potassium content

Before the commencement of the experiment, content of soil potassium was 170 kg ha⁻¹ in open and 120 kg ha⁻¹ in shade.

After the experiment, under open condition, the highest soil potassium content was obtained in the plot where IT-96 (140 kg ha⁻¹) was grown followed by the plot where IT-106 (130 kg ha⁻¹) was grown (Table 51). The soil potassium content was lowest (70 kg ha⁻¹) in the plot of IT-108.

Under shade, the plot of IT-97 (100 kg ha⁻¹) recorded the highest soil potassium content. The lowest (60 kg ha⁻¹) was in the plots of IT-108, IT-101, IT-111 and IT-114.

4.3.6.2 Plant nitrogen

Pooled analysis indicated significant difference among the accessions for plant nitrogen content under both open and shaded conditions (Table 52).

IT-108 recorded the highest pooled mean (4.45%) for plant nitrogen content. It was on par with IT-96 (4.20%), IT-105 (4.17%) and IT-101 (4.12%). IT-111 recorded the least pooled mean for nitrogen content (3.64%).

4.3.6.3 Glycoside estimation

Glycoside (indican) content in fresh leaves of the accessions at 280 nm is presented in Table 53 and Fig. 11. There was significant difference for indican content between open and shaded conditions.

Pooled analysis showed significant difference among the accessions for indican content. IT-99 recorded the highest value (0.47%) followed by IT-96 (0.46%), IT-114 (0.44%) and IT-97 (0.44%). Lowest value (0.40%) was recorded by IT-101.

Table 53. Leaf (fresh) glycoside (indican) content (%) in selected accessions of *I. tinctoria* under open and shaded conditions

Sl. No.	Accession No.	Open	Shade	Mean
1	IT- 108	0.45	0.41	0.43
2	IT- 101	0.42	0.37	0.40
3	IT- 96	0.50	0.42	0.46
4	IT- 114	0.46	0.42	0.44
5	IT- 106	0.46	0.39	0.43
6	IT- 104	0.42	0.40	0.41
7	IT- 105	0.42	0.41	0.41
8	IT- 111	0.45	0.40	0.43
9	IT- 97	0.46	0.43	0.44
10	IT- 99	0.49	0.44	0.47
	Mean	0.45	0.41	-
Pooled analysis			CD	SE
For light condition			0.006	0.017
For accessions			0.013	0.038
For interaction			0.019	0.053

Table 54. Leaf indigo content (%)* in selected accessions of *I. tinctoria* under open and shaded conditions

Sl. No.	Accession No.	Indigo content (%)		
		Open	Shade	Mean
1	IT- 108	0.87	0.50	0.69
2	IT- 101	1.04	1.03	1.04
3	IT- 96	2.14	1.21	1.67
4	IT- 114	2.14	0.75	1.45
5	IT- 106	2.15	0.63	1.39
6	IT- 104	1.01	0.77	0.89
7	IT- 105	1.15	0.84	0.99
8	IT- 111	1.01	0.78	0.89
9	IT- 97	1.19	0.79	0.99
10	IT- 99	1.97	1.11	1.54
	Mean	1.47	0.84	-
Pooled analysis			SE	CD
For light condition			0.09	0.27
For accessions			0.21	0.60
For interaction			0.30	0.85

* - Extracted by conventional method

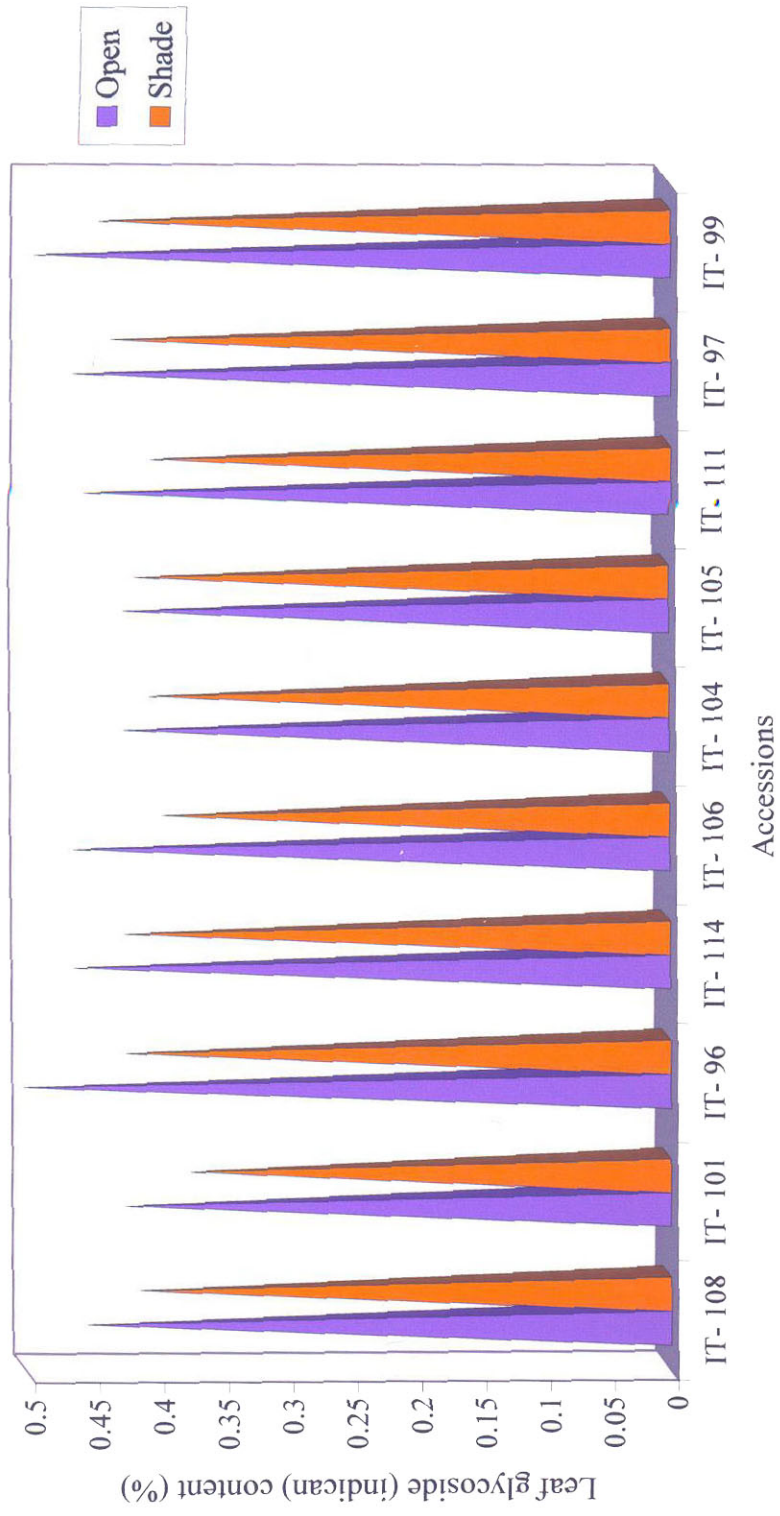


Fig. 11 Leaf (fresh) glycoside (indican) content (%) in selected *I. tinctoria* accessions

4.3.6.4 Extraction of Indigo (conventional method)

On pooled analysis, it was found that significant difference occurred between open and shaded conditions and also among the accessions for indigo content (Table 54 and Fig. 12). Indigo content was maximum for IT-96 (1.67%) It was on par with IT-99 (1.54%), IT-114 (1.45%) and IT-106 (1.39%). Minimum value (0.69%) was recorded by IT-108.

4.3.7 Correlation analysis

The phenotypic correlation coefficient, under open conditions were estimated for nineteen pairs of characters *viz.*, number of branches, girth of stem, leaf area, root length, dry weight of shoot, dry weight of leaves, dry weight of pods, dry weight of roots, number of effective root nodules, leaf area index, leaf area duration, net assimilation rate, dry matter accumulation per plant, harvest index (leaf yield), harvest index (root yield), harvest index (leaf + root yield), plant nitrogen, glycoside content and indigo content. The correlation coefficient is presented in the form of matrix in Table 55.

4.3.7.1 Correlation between shoot yield (dry weight) and other characters

The phenotypic correlation was found to be high and positive for dry weight of leaves (0.9851), leaf area index (0.9709), leaf area (0.9706), dry matter accumulation per plant (0.8957), leaf area duration (0.8559), net assimilation rate (0.8066), harvest index (leaf yield) (0.6958), harvest index (leaf + root yield) (0.6489), dry weight of root (0.5 048), girth of stem (0.4496) and root length (0.3822).

4.3.7.2 Correlation between leaf yield (dry weight) and other characters

The highest positive phenotypic correlation was recorded by dry weight of shoot (0.9851) followed by leaf area index (0.9791), leaf area (0.9788), dry matter accumulation per plant (0.9110), leaf area duration

Table 55. Phenotypic correlation coefficients of growth and yield characters in *I. tinctoria*

	X1	X2	X3	X4	X5	X6	X7	X8	X9	X10	X11	X12	X13	X14	X15	X16	X17	X18	X19	
X1	1.0000																			
X2	0.4670**	1.0000																		
X3	0.2866	0.4761**	1.0000																	
X4	0.2821	0.3476	0.3689*	1.0000																
X5	0.2671	0.4496*	0.9706**	0.3822*	1.0000															
X6	0.2790	0.4765**	0.9788**	0.4218*	0.9851*	1.0000														
X7	-0.2440	0.1856	0.2129	0.2208	0.2325	0.2701	1.0000													
X8	0.4056*	0.4049*	0.5290**	0.3752*	0.5048**	0.5841**	0.3803*	1.0000												
X9	0.1988	0.0475	0.3570	0.3878	0.3619	0.3437	0.2473	0.1275	1.0000											
X10	0.2876	0.4763**	0.9999**	0.3685*	0.9709**	0.9791**	0.2128	0.5287**	0.3561	1.0000										
X11	0.2001	0.4057*	0.8492**	0.2612	0.8559**	0.8601**	0.3139	0.4672*	0.3524	0.8491**	1.0000									
X12	0.2801	0.3772	0.7880**	0.3585	0.8066**	0.7831**	0.0515	0.4349*	0.2272	0.7886**	0.3969*	1.0000								
X13	0.1534	0.4602*	0.8696**	0.4209*	0.8957**	0.9110**	0.6263**	0.6618**	0.3823	0.8698**	0.8178**	0.6715**	1.0000							
X14	0.3540	0.2576	0.7332**	0.2130	0.6958**	0.7052**	-0.4546**	0.1970	0.1430	0.7335**	0.5359**	0.6360**	0.3531	1.0000						
X15	0.0960	0.0116	0.2283	0.1625	0.1850	0.2352	0.1069	0.7251**	0.1791	0.2273	0.1082	0.2928	0.2793	0.0830	1.0000					
X16	0.4920**	0.3410	0.7022**	0.2549	0.6489**	0.6864**	-0.4282*	0.4722**	0.0830	0.7022**	0.5125**	0.6103**	0.3677*	0.9309**	0.3318	1.0000				
X17	0.0970	0.2119	0.2365	0.3410	0.2712	0.3080	0.2758	0.4737**	0.1195	0.2367	0.1527	0.3249*	0.3755	0.0491	0.3480	0.1524	1.0000			
X18	0.0009	0.0409	-0.1343	-0.0216	-0.0978	-0.1533	0.0562	-0.2334	0.0346	-0.1358	0.0296	-0.1921	-0.0511	-0.2781	0.1056	-0.2343	0.3139	1.0000		
X19	0.2645	-0.1119	-0.0522	-0.0713	-0.0441	-0.1368	-0.2208	-0.2334	0.0774	-0.0527	-0.0623	0.0072	-0.1537	-0.0511	-0.0652	-0.0931	-0.1710	0.4293*	1.0000	

X1- Number of branches, X2- Girth of stem, X3- Leaf area, X4- Root length, X5- Dry weight of shoot, X6- Dry weight of leaves, X7- Dry weight of pods, X8- Dry weight of roots, X9- Number of effective root nodules, X10- Leaf area index, X11- Leaf area duration, X12- Net assimilation rate, X13- Dry matter accumulation per plant, X14- Harvest index (leaf yield), X15- Harvest index (root yield), X16- Harvest index (leaf+ root yield), X17- Plant nitrogen, X18- Glycoside content, X19- Crude indigo content

**Significant at 5 % level

* Significant at 1 % level

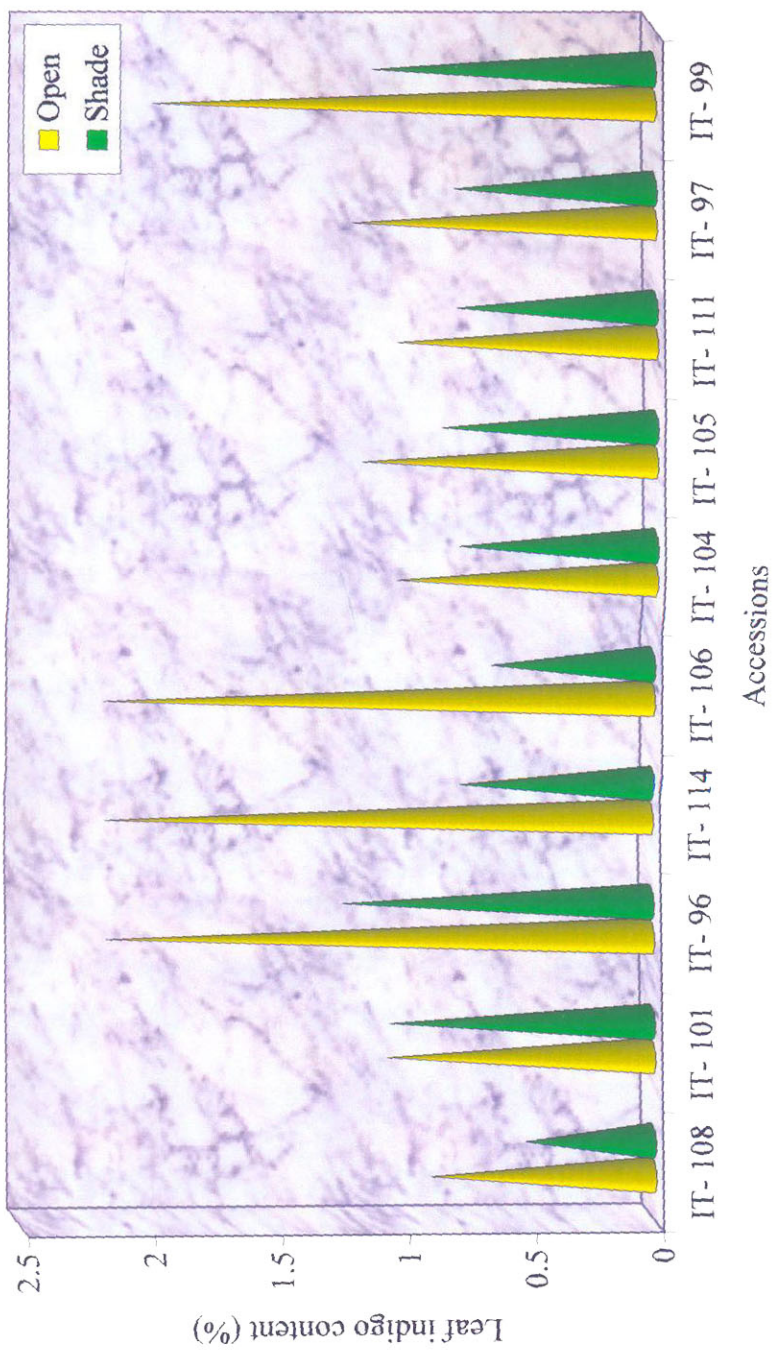


Fig. 12 Leaf indigo content (%) in selected *I. tinctoria* accessions

(0.8601), net assimilation rate (0.7831), harvest index (leaf yield) (0.7052), harvest index (leaf + root yield) (0.6864), dry weight of root (0.5841), girth of stem (0.4765) and root length (0.4218).

4.3.7.3 Correlation between pod yield (dry weight) and other characters

Dry matter accumulation per plant (0.6263) had the highest positive phenotypic correlation with pod yield followed by dry weight of root (0.3803). High negative correlation was observed for harvest index (leaf yield) (-0.4546) and harvest index (leaf + root yield) (-0.4282).

4.3.7.4 Correlation between root yield and other characters

Harvest index (root yield) (0.7251), dry matter accumulation per plant (0.6618), dry weight of leaves (0.5841), leaf area (0.5290), leaf area index (0.5287), dry weight of shoot (0.5048), plant nitrogen (0.4737), harvest index (leaf + root yield) (0.4722), leaf area duration (0.4672), net assimilation rate (0.4349), number of branches (0.4056), girth of stem (0.4049), dry weight of pods (0.3803) and root length (0.3752) showed high positive phenotypic correlation.

4.3.7.5 Correlation between glycoside content and other characters

The phenotypic correlation was found to be high and positive (0.4293) for crude indigo content. Most of the other growth characters expressed negative correlation with glycoside content.

4.3.7.6 Heritability, genetic advance (GA), GCV and PCV

Broad sense heritability, genetic advance (GA), the phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) are shown in Table 56.

High phenotypic coefficient of variation (PCV) was shown by the characters viz, crude indigo content (44.90%), dry weight of root (28.72%), harvest index (root yield) (26.68%), dry weight of leaves (21.13%), net

Table 56. Heritability, genetic advance (GA), genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) in *Indigofera tinctoria*

Sl. No.	Characters	Heritability (%)	GA (%)	GCV (%)	PCV (%)
1	Number of branches	21.24	8.6272	9.0880	19.7173
2	Girth of stem (cm)	2.05	0.8739	2.9628	20.6936
3	Leaf area (cm ²)	94.76	10.6071	5.2895	5.4338
4	Root length (cm)	32.66	7.0665	6.0029	10.5032
5	Dry weight of shoot (g)	98.99	34.4440	16.8031	16.8889
6	Dry weight of leaves (g)	99.09	43.1323	21.0337	21.1303
7	Dry weight of pods (g)	5.25	2.6695	5.6551	24.6836
8	Dry weight of roots (g)	96.64	57.1755	28.2340	28.7201
9	Number of effective root nodules	58.87	9.5278	6.0283	7.8565
10	Leaf area index	94.76	10.6026	5.2872	5.4315
11	Leaf area duration	97.25	11.9776	5.8961	5.9788
12	Net assimilation rate	98.19	38.6626	18.9406	19.1142
13	Dry matter accumulation per plant	83.14	26.5504	14.1350	15.5022
14	Harvest index (leaf yield)(g)	61.02	11.8196	7.3452	9.4029
15	Harvest index (root yield) (g)	54.31	29.8472	19.6603	26.6782
16	Harvest index (leaf + root yield) (g)	51.35	9.0888	6.1569	8.5921
17	Plant nitrogen	75.45	20.0801	11.2220	12.9193
18	Glycoside content	58.70	8.6961	5.5101	7.1915
19	Crude indigo content	56.51	52.2700	33.7536	44.9009

assimilation rate (19.11%) and dry weight of shoot (16.89%). Genotypic coefficient of variation (GCV) was also higher for crude indigo content (33.75%), dry weight of root (28.23%), dry weight of leaves (21.03%), harvest index (root yield) (19.66%), net assimilation rate (18.94%) and dry weight of shoot (16.80%). For most of the characters, heritability values were high and ranged from 51.35 per cent to 99.09 per cent. Genetic advance values were high only for four characters, viz, dry weight of roots (57.18%), crude indigo content (52.27 %), dry weight of leaves (43.13 %) and net assimilation rate (38.66 %).

4.4 ECONOMICS OF CULTIVATION (COST/ BENEFIT RATIO) IN OPEN AND SHADE IN COCONUT GARDEN

Estimated expenditure and returns per hectare of *I. tinctoria* both in open and under coconut shade are given in Table 57. Total expenditure for *I. tinctoria* cultivation comes to Rs. 1,33,950 ha⁻¹ under open condition and under shade. Gross income under open condition was estimated as Rs 4,05,500 ha⁻¹ and under shade in coconut garden as Rs. 2,30,000 ha⁻¹.

In open situation,

$$\begin{aligned} \text{Net income} &= \text{Gross income} - \text{total expenditure} \\ &= \text{Rs } 2,71,550 \end{aligned}$$

$$\begin{aligned} \text{Benefit - cost ratio} &= \frac{\text{Gross income}}{\text{Total expenditure}} \\ &= 3.03 \end{aligned}$$

Under shaded condition,

$$\begin{aligned} \text{Net income} &= \text{Rs } 96,050 \\ \text{Benefit - cost ratio} &= 1.72 \end{aligned}$$

4.5 INCIDENCE OF PESTS AND DISEASES

Surveillance for the occurrence of pests and diseases revealed that *I. tinctoria* crop is susceptible to the attack of the psyllid, *Arytaina punctipennis*

Table 57. Estimated expenditure and return per hectare of *I. tinctoria* under open and shade (as intercrop in coconut garden)

A) Estimated expenditure under open and shade

i) Cost of labour			
Sl. No	Details of Work	Labour	Amount (Rs.) @ Rs 190/ day
1	Preparation of seedlings in poly bags and nursery maintenance	80	15200
Mainfield preparation-			
2	First digging	50	9500
	Second digging & levelling	40	7600
3	Taking furrows for planting	30	5700
4	Drying, powdering, transportation and application of FYM as basal dressing	40	7600
5	Planting of seedlings	40	7600
6	Irrigation for 6 months (mainfield)	130	24700
7	Gap filling	15	2850
Weeding (2 times) –			
8	3 months after sowing	50	9500
	4 months after sowing	50	9500
9	Harvesting	130	24700
		Total	124450
ii) Cost of inputs			
	Item	Quantity	Price
1	Planting material (seeds) (kg) @ Rs 1000/kg	3	3000
2	Cost of farmyard manure (t) @ Rs 500/t	10	5000
3	Cost of PP chemicals	-	1500
		Total	9500
Total Expenditure - (i) + (ii)			133950

B) Gross income

Sl. No.	Item	Quantity (kg)		Price (Rs)	
		Open	Shade	Open	Shade
1	Sale of leaves @ Rs 10/kg	35000	20000	350000	200000
2	Sale of roots @ Rs 15/kg	3700	2000	55500	30000
	Total returns			405500	230000

which causes curling up and drooping of leaves and shoots, leading to wilting of plants. The nymphs secrete honeydew resulting in the colonization of ants. The attack is found to be more in plants growing under coconut shade. Immediate control measures were adopted by spraying Dicofol @ 3 ml l⁻¹.

Discussion

5. DISCUSSION

The present investigation was conducted to evaluate the genetic stock of *Indigofera tinctoria* and to study the performance of selected accessions, so as to identify the best accession(s) yielding high biomass and glycosides. The results of the experiment, presented in the previous chapter are discussed hereunder.

5.1 SEED CHARACTERIZATION AND GERMINATION STUDIES

Of the thirty accessions collected for the study, five accessions *viz.* IT-98, IT-95, IT-108 and IT-106 recorded more than 80 per cent germination. Higher germination percentage of these accessions may be attributed to their high genetic vigour. IT-111 and IT-102 exhibited poor germination (47 and 48 respectively). This may be due to the dormancy resulting from very hard seed coat (Kumari and Francis, 1998).

Hundred seed weight of the accessions ranged from 0.28-0.52 g. Of the thirty accessions collected for the study, IT-104 recorded the highest hundred seed weight of 0.56 g.

Variation was observed in seed colour, seed weight and appearance of different accessions, which may be attributed to the differences in their genetic make up.

5. 2. PHASE I EXPERIMENT- GROWTH AND YIELD ANALYSIS OF THE GENETIC STOCK OF *INDIGOFERA TINCTORIA* AS PURE CROP AND AS INTERCROP IN COCONUT GARDEN

5.2.1 Growth parameters

5.2.1.1 Shoot characters

Shoot characters include plant height, plant spread, height at first branching, number of branches, girth of stem, number of leaves and leaf area.

Considerable variation was noticed in plant height among the accessions under open condition at pre-flowering stage, under shade at flowering stage and under both these conditions at seed maturation stage. Bose (1963) reported that variation in plant height within legumes is purely a function of genetic make up. The accessions IT-106, IT-108, IT-113 and IT-105 were found to show better performance with respect to plant height compared to the other accessions, irrespective of the growing condition. Hence these accessions may possess more genetic vigour than the others.

Plant height of all the accessions exhibited an increasing trend throughout the growth period under shade as well as in open condition. A similar trend was recorded in *Andrographis* sp. as reported by Singh *et al.* (2001a).

Higher plant height was observed in open condition in all the accessions than under shade at all the three growth stages. Reduction in plant height under shade in coconut garden may be due to the physiological and ecological influence, especially light, which limits growth of crops in mixtures (Blackman and Black, 1959). Existence of a tight competition for eco-physiological requirements like water, nutrients and light might have resulted in an unfavourable situation for rapid vegetative growth, thereby causing a reduction in plant height (Anilkumar, 1984). Lalithabai (1981) has reported maximum plant height at full illumination in *Colocasia esculenta* compared to shade under coconut garden. Similarly a negative trend on the shade response to plant height has been reported by George (1982) in *Vigna radiata* and *Vigna unguiculata*.

The accessions differed significantly with respect to plant spread at all the three growth stages under open as well as under shaded conditions. Among the accessions, those that revealed a trend to dominate in plant spread were IT-106, IT-108, IT-96 and IT-105, irrespective of the growing conditions. Under open condition, those accessions possessing genetic vigour in terms of plant height were found to possess greater plant spread also. Plant spread was found to be higher in open condition

compared to shade. This may be due to more branching and sidewise growth of plants resulting from their profuse growth under open conditions. The primary reason for the decrease in the spread of plants under shade is the reduction in the number of branches under shaded conditions (Pillai, 1990). Greater plant spread in *Sida rhombifolia* and *Solanum melongena* var. *incanum* in open condition than under shade was reported by Sarada (2000).

Under open as well as shaded conditions, the accessions differed significantly for the height of first branching. Under shade at all the three growth stages, IT-104 and IT-108 produced the first branch at a greater height. This may be attributed to the lanky nature of growth of the accessions under shade. These two accessions recorded a higher plant height also, under shade. All the accessions were found to produce the first branch at a greater height under shaded condition compared to open. The shade effect on plant height is believed to be due to auxin enhancement, probably acting synergistically with gibberellic acid (Leopold, 1964). Theoretically, photodestruction of auxin is less in shaded stands. Shading tends to increase auxin levels which could affect the height of first branching (Evans, 1973).

There was significant difference among the accessions for number of branches under shade at pre-flowering stage and at flowering and seed maturation stages under open and shaded conditions. IT-105 and IT-108 produced more branches irrespective of open and shaded conditions. According to George (1981), the number of branches is usually related to the height of the plants. The accessions with good genetic make up may have greater capacity to utilize the environmental resources and produce more number of branches. Number of branches was influenced significantly due to open and shaded conditions also. Open condition recorded the maximum number of branches. Reduction of branching in shade has been reported by Beinhart (1963) in white clover (*Trifolium repens*) (a leguminous plant), Nalawadi *et al.* (1988) in *Ixora coccinia*, Pillai (1990) in

Ocimum gratissimum, Sunitha (1996) in *Clitoria ternatea* and Sarada (2000) in *Sida rhombifolia* and *Solanum melongena* var. *incanum*. The possible reason that can be attributed to this phenomenon is that the available low light intensity in shade may be inadequate to stimulate the production of new branches. The absence of competition for light, space, water or nutrients in a pure cropping system may result in higher production of branches. As sunlight passes through the tree canopy in plantations, spectral changes perceived by the plants through the phytochrome system, may induce marked morphogenetic changes in plants and inhibit branching (Deregibus *et al.*, 1985).

Considerable variation was noticed among the accessions for stem girth under shaded condition at pre-flowering stage and under open as well as in shaded conditions at flowering and seed maturation stages. Irrespective of the above conditions, IT-108 and IT-101 were found to produce thicker stems compared to the other accessions. Increased branching was also observed in these two accessions. Thicker stem was produced in open compared to shade. Increased vegetative growth under open condition tends the cambium to become more active and form a thicker stem.

Significant difference was observed among the accessions for number of leaves under shade at pre-flowering stage and under open as well as shaded conditions at flowering and seed maturation stages. IT-104, IT-108 and IT-106 produced more number of leaves at all the growth stages, which can be attributed to their increased genetic vigour. Variation in number of leaves among the accessions may be attributed to the fact that it is purely a function of genetic make up and environmental conditions (Gardner *et al.*, 1988). In *I. tinctoria*, Sunitha (1996) reported the production of more number of leaves (623) under coconut shade than under open condition (619), 180 days after sowing. Varghese (1989) observed a decrease in the number of leaves under shade in *Zingiber officinale* and *Curcuma longa*. The plants of *Centella asiatica* produced a greater number

of leaves under high light intensity than under low light intensity (Wankher and Tripathi, 1990). Similar results were also observed by Sheela (1992) in turmeric and Sarada (2000) in *Sida rhombifolia* and *Phyllanthus amarus*.

Accessions that maintained a steady superiority in leaf area were IT-108, IT-106, IT-104 and IT-96, in all the three growth stages. These accessions were found to produce more number of leaves, which resulted in higher leaf area. This factor is beneficial to the plant as a larger leaf surface serves to capture more solar radiation and hence, increase photosynthetic rates. Leaf area was found to be more in open. Increased light intensity resulting in greater leaf area has been reported by Beinhart (1963) in white clover, a leguminous plant.

From pre-flowering stage to flowering stage, the increment in the number of leaves and leaf area was found to be higher compared to that between flowering and seed maturation stage. This may be attributed to the transition from vegetative to reproductive stage, which is characterized by leaf senescence and leaf fall. Similar observations were recorded by Samuel (2000) in *Mucuna pruriens* and Nair (2000) and Resmi (2001) in *Clitoria ternatea*.

5.2.1.2 Root characters- root length and girth

Root length showed significant variation among the accessions under open as well as shaded conditions at all the three stages of plant growth. Under open condition, IT-96 recorded the longest root during all the growth stages. Irrespective of growing condition, IT-108, IT-105 and IT-96 performed better in the case of root length compared to other accessions. Increased vegetative growth in terms of height, spread, number of branches *etc.* might have stimulated the production of bigger roots in these accessions under open condition. A root length of 32.85 cm under open and 30.90 cm under coconut shade has been reported in *Indigofera tinctoria*, 180 days after sowing by Sunitha (1996). Root length was found to be higher under open condition than under shade. This is in confirmation with the findings of Sunitha (1996) and Resmi (2001) in *Clitoria ternatea*

and Sarada (2000) in *Sida rhombifolia* and *Phyllanthus amarus*. This might be due to more vigorous and faster growth rate of the plants under open condition when compared to that under shade in coconut garden.

Those accessions that dominated in plant length and leaf production were found to lead in root length measurements. These accessions would be genetically more vigorous. Similar findings were reported by Babalola (1980) and Rajan (1999) in legumes.

The accessions varied significantly for root girth under open condition at pre-flowering stage and under open as well as shaded conditions at flowering and seed maturation stages. Thicker roots at collar region were observed in IT-108, IT-96 and IT-101, irrespective of open and shaded condition. Also, more root girth was observed under open condition, which might be due to the lesser competition for nutrients. Sunitha (1996) reported a root girth of 5.93 cm in open and 5.8 cm under coconut shade in *Indigofera tinctoria*, 180 days after sowing.

5.2.2 Yield and yield attributes

5.2.2.1 Shoot yield

Pooled analysis showed that fresh weight of shoot varied significantly among the accessions at flowering and seed maturation stages and under shade at pre-flowering stage. Dry weight of shoot varied significantly among the accessions at flowering and seed maturation stages on pooled analysis and at flowering stage under open and shaded conditions.

Irrespective of the growing conditions, IT-108, IT-101, IT-106, IT-104 and IT-98 recorded higher shoot fresh weight and dry weight at all stages of growth. These accessions were found to produce better vegetative growth in terms of height, spread, number of branches, number of leaves *etc.* Biomass yield is mainly a function of vegetative growth.

During the period from flowering to seed maturation stage, covering about three months, the increase in fresh and dry weight of shoot

was found to be proportionately less compared to that from pre-flowering to flowering stage covering two months. This decline can be attributed to the leaf shedding and senescence as reported by Kulkarni and Karadge (1991). When senescence slowly sets in, the assimilate produced gets diverted for seed set (Cock and Yoshida, 1972) and (Evans, 1973).

Fresh and dry weight of shoot was comparatively superior under open condition. The environmental conditions under which a plant grows control the productivity of the plant to a great extent. Out of the various physical environmental factors, the light regime is one, which has much influence on the growth and productivity of a plant (Bindra and Brar, 1977). Yield is a complex character, which is the outcome of a number of genetic factors and environmental conditions. Reduced yield under the stress of shade is reported as a regular phenomenon in various tropical vegetables (Nair, 1991) and pepper (Leonardi, 1996).

5.2.2.2. Leaf yield

There was significant variation among the accessions for both fresh and dry weight of leaves at flowering and seed maturation stages on pooled analysis and at pre-flowering stage under shade.

IT-108, IT-101, IT-106, IT-104 and IT-98, which dominated in shoot fresh and dry weights, were also found to produce higher leaf fresh and dry weights. This points to the fact that higher shoot yield relates to higher leaf yield also. Hence, in the case of leaf fresh and dry weights also, highest increment was observed during pre-flowering to flowering stage as reported by Samuel (2000) in *Mucuna pruriens* and Resmi (2001) in *Clitoria ternatea*. Leaf yield was also found to be higher under open condition. Smith (1977) reported that greater the light intensity, higher the yield since more light reaches the lower leaves and even the lower layers of chloroplasts within the leaf. In cassava, Ramanujam and Jose (1984) observed that the photosynthetic apparatus per unit leaf area was curtailed under low light intensity. In summer, one per cent reduction in light

intensity imposed 0.36 per cent yield loss, because the rate of photosynthesis decreased with decreasing light intensity (Akimovo *et al.*, 1986).

5.2.2.3 Pod yield

Pod yield was found to differ significantly among the accessions as indicated by pooled analysis at seed maturation stage.

IT-101, IT-108, IT-106 and IT-96, which produced good vegetative growth gave higher pod yield also. This may be attributed to the overall genetic superiority of these accessions over the others. Fresh and dry weight of pods were superior under open condition. According to Cathedral and Lantican (1977), artificial shade of 40-50 per cent reduced sunlight and caused a yield reduction of 30 per cent compared to that in full sunlight for soyabeans and about 70 per cent for mungbeans. Usha (1990) reported that there was a slight reduction in the vegetative as well as yield attributes when horsegram was grown as intercrop in coconut garden compared to sole crop. The grain yield of black gram as intercrop in coconut-casuarina agroforestry system was also lesser than the sole crop yield.

5.2.2.4 Root yield

Analysis of the data on root yield revealed that fresh weight of root varied significantly among the accessions under shade at pre-flowering and flowering stages and on pooled analysis at seed maturation stage. Dry weight of root varied significantly among the accessions under shade at pre-flowering stage and on pooled analysis at flowering and seed maturation stages.

IT-106, IT-108 and IT-104, which showed good performance in terms of shoot and leaf yield, were also found to dominate in root yield. The increase in root weight is proportional to the increase in root length and root girth as reported by Resmi (2000) in *Clitoria ternatea*. Under open

condition, root yield was found to be higher. The superiority in root yield under open condition can be attributed to more number of roots, greater root length and girth under open condition. Fresh and dry weight of officinal part was found to be higher in open condition for *Alpinia galanga*, *Plumbago rosea*, *Costus speciosus*, *Ocimum* sp. and *Kaempferia galanga* compared to that under shade as intercrop in coconut plantation (Nair *et al.*, 1991b). Nelson (1964) observed that root girth is usually inhibited by low light intensities and this can lead to a reduction in assimilate flow to the root system.

5.2.3 Glycoside content

Glycoside content in *I. tinctoria* was found to vary among the accessions, between the leaf treatments and between open and shaded conditions. Jose (2002) recorded that leaf tissues of *Indigofera tinctoria* are more potent sites of indican production than the tissues of other parts, the callus tissue derived from leaf explants showed more or less similar capabilities to synthesise indican. Maximum content of β glucosidase was also recorded from leaf tissue.

IT-99 showed superior performance in terms of glycoside content at both the wavelengths. But the vegetative characters in terms of plant spread, number of branches, number of leaves and root length were found to be the least in IT-99. According to Wijesekera (1993), morphological functions that may be a good guide in the breeding of other plants may be misleading in the case of medicinal and aromatic plants. In *Digitalis*, there was no correlation between thick, wide and healthy looking leaves and a high content of the desired glycoside. Similarly in the case of citronella or lemon grass, luxuriant foliage does not imply a higher content of essential oil and often it is quite the reverse (Wijesekera, 1993).

Maximum amount of glycoside was obtained from the fresh leaves of IT-99 (0.57%). This is in conformity with the findings of Khazhakyian

and Egibyan (1988). They obtained indican 5 to 15 mg g⁻¹ fresh leaf of *I. tinctoria*, determined by spectrophotometry.

Among the different leaf treatments, fresh leaves of *I. tinctoria* was found to possess more indican content. This shows that the glycoside content decreases on drying. Gupta (1995) obtained similar results in foxglove. A rise in drying temperature to 40⁰ C or more induced degradation of glycosides in foxglove. Srivastava *et al.* (1980) reported that slow drying of senna leaves under the sun induces blackening, indicating loss of sennosides by up to 50 per cent. The indican content of fermented leaves was also less. During fermentation, indican on hydrolysis liberates indoxyl, which in turn oxidizes to the dye indigo (Epstein *et al.*, 1967). This may be the reason for low indican content in fermented leaves.

Higher glycoside content was obtained in the open condition in all the accessions and for all the leaf treatments. This agrees with the findings of Gupta and Chadha (1995) that ample sunshine is necessary in mints at maturity to enable plants to synthesis higher content of oil and menthol. Varghese (1989) found that curcumin content of turmeric rhizome showed a progressive decrease with increase in shade. Similar reports were made by Li *et al.* (1996) in sage and thyme, Jaggi and Kapoor (1997) in *Solanum laciniatum* and Lambers *et al.* (1998) in tea.

5.2.4 Selection index

For detailed study on the influence of shade and open conditions on the growth and glycoside content in *I. tinctoria* in the Phase II experiments, the best ten accessions as determined using the selection index scores based on leaf yield and glycoside content in open condition was used. The ten accessions (blue green leaved) of *I. tinctoria*, which showed superior performance in open condition were IT-108, IT-101, IT-96, IT-114, IT-106, IT-104, IT-105, IT-111, IT-97 and IT-99.

5.3 PHASE II CULTURAL TRIAL OF SELECTED ACCESSIONS UNDER OPEN AND SHADED CONDITIONS

5.3.1 Growth parameters

5.3.1.1 Shoot characters

Shoot characters include plant height, plant spread, height at first branching, number of branches, girth of stem, number of leaves and leaf area.

Considerable variation was noticed in plant height among the accessions under shade at pre-flowering and flowering stages only. IT-108, IT-106, IT-96 and IT-101 showed better performance with respect to plant height compared to the other accessions. This may be attributed to the genetic vigour of these accessions as observed by Bose (1963) in legumes.

Plant height of all the accessions exhibited an increasing trend throughout the growth period under open and shaded conditions, which was more obvious during the later stages of growth as recorded by Singh *et al.* (2001a) in *Andrographis* spp.

More plant height was observed in open condition in all the accessions than that under shade at all the three growth stages. Blackman and Black (1959) have reported that the reduction in plant height under shade in coconut garden may be due to the physiological and ecological influence, especially light, which limits growth of crops in mixtures. According to Nair (1979), almost all tropical grain legumes are very sensitive to the partial shade existing in coconut gardens. Existence of a tight competition for eco-physiological requirements like water, nutrients and light might have resulted in an unfavourable situation for rapid vegetative growth, thereby causing a reduction in plant height (Anilkumar, 1984). Maximum plant height at full illumination was also reported by Lalithabai (1981) in *Colocasia esculenta* and George (1982) in *Vigna radiata*

and *Vigna unguiculata*. In general, shade affects the growth and morphological development of leguminous plants.

Pooled analysis showed that the accessions differed significantly with respect to plant spread at all the three growth stages. The accessions that dominated in plant spread were IT-101 and IT-105. Plant height was also higher in the case of IT-101. This may be attributed to its increased genetic vigour. But IT-105 recorded only lesser plant height. This may be due to more branching and sidewise growth of the plants at the expense of plant height resulting from their profuse growth under open conditions. Plant spread was generally less under shade. The primary reason for the decrease in the spread of plants under shade is the reduction in the number of branches under shaded conditions as observed by Pillai (1990) in *Ocimum gratissimum*. Similar observation was also made by Sarada (2000) in *Sida rhombifolia* and *Solanum melongena* var. *incanum*.

Significant difference was observed among the accessions at pre-flowering and flowering stages on pooled analysis and at seed maturation stage under shade. IT-101, IT-96 and IT-108 were found to produce the first branch at greater heights. These accessions recorded more plant height also. Hence it can be concluded that plants, which grow taller will produce the first branch also at a greater height. This shows the vigorous nature of growth of those accessions. But when growing conditions were compared, plants growing in shade produced first branch at higher points on the stem, which resulted in lanky nature of growth. The shade effect is believed to be due to auxin enhancement, probably acting synergistically with gibberellic acid (Leopold, 1964).

There was significant difference among the accessions for number of branches under shade at flowering and seed maturation stages of growth. IT-108, IT-101 and IT-114 were found to produce more number of branches. These accessions produced greater plant height also under shade. This is in conformity with the observations of George (1981) that the number of branches is usually related to the height of the plant. This shows

the greater capacity of accessions with good genetic make up to utilize the environmental resources efficiently and to produce more number of branches. Plants growing under shade recorded lesser number of branches since the low light intensity in shade may be inadequate to stimulate the production of new branches. Crop competition for light, space, water and nutrients also exists under shade.

Considerable variation was noticed among the accessions for stem girth under shaded condition at pre-flowering stage and on pooled analysis at flowering stage. IT-99, IT-114, IT-104 and IT-106 produced more stem girth compared to the other accessions. IT-114 also produced more number of branches under shade at seed maturation stage. IT-99 and IT-104 were found to possess lesser plant height at pre-flowering stage, which may be the reason for greater stem girth at this stage in these two accessions. Thicker stem was produced under open condition compared to shade. Increased vegetative growth under open condition favoured by improved synthesis of growth hormones like cytokinins which promote cell division and gibberellins which promote cell elongation, may tend the cambium to become more active and form a thicker stem.

Significant difference was observed among the accessions for number of leaves under shade at pre-flowering and flowering stages of growth. IT-108 and IT-101 produced more number of leaves, which can be attributed to their increased genetic vigour in terms of plant height, spread, height at first branching and number of leaves also. Gardner *et al.* (1988) opined that variation in number of leaves among the accessions may be attributed to the fact that it is purely a function of genetic make up and environmental conditions. More number of leaves were produced under open conditions compared to shade. Similar observations were made by Varghese (1989) in *Zingiber officinale* and *Curcuma longa*; Wankher and Tripathi (1990) in *Centella asiatica*; Sheela (1992) in turmeric and Sarada (2000) in *Sida rhombifolia* and *Phyllanthus amarus*.

In the present study, accessions that showed superiority in leaf area were IT-101, IT-106, IT-108 and IT-114. These accessions were found to produce more number of branches and number of leaves which resulted in higher leaf area and finally in increased photosynthetic rates. Leaf area was found to be more in open as reported by Beihart (1963) in white clover. Photosynthetic rate per unit leaf area is low under shade. This could have resulted from a reduced surface area available for light reception and gas exchange (Neerakkal *et al.*, 2003).

The increment in the number of leaves and leaf area was found to be more from the pre-flowering to the flowering stage than that between flowering and seed maturation stage. The transition from vegetative to reproductive stage is characterized by leaf senescence and leaf fall (Samuel, 2000; Nair, 2000 and Resmi, 2001).

5.3.1.2 Root characters- root length and girth

Root length showed significant variation among the accessions under open at pre-flowering and flowering stages and under open as well as shaded conditions at seed maturation stage. Under open condition, IT-101 produced the longest root during all the growth stages. Irrespective of growing conditions IT-108, IT-96 and IT-101 performed better in the case of root length. Genetic vigour of these accessions in terms of plant height, plant spread, number of branches and number of leaves might have resulted in the production of longer roots in these accessions. Increased root length was observed under open conditions which suggests the vigorous growth under sunlight compared to shade as observed by Sunitha (1996). Sarada (2000) and Resmi (2001). Babalola (1980) and Rajan (1999) have reported that in legumes, increased plant height and leaf production leads to increased root length.

Pooled analysis showed that the accessions varied significantly for root girth at pre-flowering and flowering stages. Root girth was more in IT-101 and IT-114. IT-114 also dominated for number of branches, girth of

stem and leaf area. IT-101 also dominated in plant height, spread, number of branches, number of leaves, leaf area and root length. More root girth was observed under open condition, which might be due to the lesser competition for nutrients.

5.3.2 Root nodule characteristics

Root nodule characteristics consist of number of root nodules, number of effective root nodules, fresh weight and dry weight of nodules.

Root nodules were found to be present in the roots of *I. tinctoria* only during the pre-flowering and flowering stages of the plant. At seed maturation stage, nodules were absent. This conforms to the findings of Russell (1961), that the nodules of annual legumes tend to die out during active flowering and seed setting stages. Similar results were obtained by Nair (2000) and Resmi (2001) in *Clitoria ternatea*.

Nodule count and nodule weight were found to be the least at pre-flowering stage. From pre-flowering to flowering stage, it showed an increasing trend and then it was completely absent at seed maturation stage, which may be due to sloughing of nodules after their life span. Similar findings were reported by Venkataraman and Tilak (1990).

Amount of nitrogen fixed by the plant is related to the number of effective nodules. In all the accessions, the effective nodule count was highest at the flowering stage. During pre-flowering stage, roots are immature, when the nodule number and weight were less. It was found to be the highest during flowering stage, when the roots increased in length and girth.

Allen and Allen (1981) recorded that *I. arrecta* produces a deep root system with plentiful number of root nodules. Sen (1993) found that in *I. tinctoria* root nodules were abundant, diffused and globose in structure.

Pooled analysis indicated significant variation among the accessions at flowering stage for the number of root nodules and number of effective root nodules and at pre-flowering and flowering stages for fresh

and dry weight of root nodules. Number of root nodules and number of effective root nodules were higher for IT-101 and IT-105. Fresh and dry weight of root nodules were higher for IT-108, IT-101 and IT-105. Less nodule count and more nodule weight in IT-108 showed that it produces bigger sized nodules. IT-108 and IT-101 performed well in terms of fresh and dry weight of shoot and leaves. IT-108, IT-101 and IT-105 showed superiority for root yield at flowering stage. Hence it can be concluded that nodule count and weight are related to the shoot, leaf and root yield of the plant. This is in conformity with the findings of Samuel (2000) in *Mucuna pruriens* that the plant, which records maximum yield has significantly more number of nodules and weight at all stages of observations.

Both nodule count and weight were found to be higher under open condition. Whyte and Trumble (1953) reported that the infection of plants by nodule bacteria and the effectiveness of nodulation in terms of nitrogen fixation improve as the supply of light increases. Sunitha (1996) had also recorded higher nodule count and weight in *I. tinctoria* grown in open condition compared to that under coconut shade.

5.3.3 Flowering and seeding-behaviour of *I. tinctoria*

Flowering in *I. tinctoria* was observed about 111 days after sowing (DAS) to 116 DAS in open condition and about 117 DAS to 126 DAS under shade. Profuse flowering occurred about 150 DAS, which was fixed as the second stage of plant growth. According to Benvenuti *et al.* (1994), shading caused species dependant delay in the onset of flowering in *Datura stramonium*. *I. tinctoria* accessions IT-101 and IT-96. which dominated in shoot and leaf yield produced more number of flowers under open condition. *I. tinctoria* flowers open early in the morning, close before noon and fall in the same day. Anthesis starts by slight cracking at the top of the bud. The process of opening of corolla takes 45-60 minutes. Anthesis was spread over from 8.27 a.m. to 9.12 a.m. under open condition and from 8.46 a.m. to 10.08 a.m. under shade. Swarnapiriya *et al.* (1995) stated that temperature and relative

humidity (RH) had influence on anthesis in *Gloriosa superba*. When the temperature is low and RH is high, the flower opening is delayed for a few hours in *G. superba*. Lower temperature and higher RH under shade compared to open condition may be the reason for the delay in anthesis in shade. In *I. tinctoria*, anthers mature and start dehiscing about 7-8 hours before the time of opening of corolla. Pollen is released through openings such as splits, transverse cracks or pores. Temperature and relative humidity regulate the duration of pollen discharge in *Ricinus communis* L. (Meinders and Jones, 1950). The peak anther dehiscence was between 10.57 to 11.50 p.m. in *I. tinctoria* in open condition and between 11.16 to 12.18 p.m. under shade, the day before anthesis. Anther dehiscence was completed in 1 to 2 hours. Increased temperature caused earlier dehiscence and lower temperature delayed the dehiscence. The results of the estimation of pollen production revealed that the average pollen count per anther under open was 1733.34 and that under shade was 1561.12. Higher pollen production and more number of flowers in open might have contributed to the better pod set under open condition. The mean pollen fertility values obtained by staining method under open and shade were 61.48 and 63.22 per cent respectively. Fertile pollen were deeply stained and more or less rounded in shape. The sterile pollen were irregular and less stained. Sreekala (1997) observed that in *Allamanda*, the fertile pollen grains were bigger in size and deeply stained, while the sterile ones were smaller, less stained and irregular in shape. In *I. tinctoria*, the flower shedding percentage was high. This led to a poor pod setting percentage of 20.91 and 17.30 per cent respectively under open and shaded condition. Average number of days for pod set was 138 days in open and 148 days under shade. Higher temperature in open condition might have caused the rapid pod development process in *Moringa oleifera* Lam. (Sindhu, 2002). *I. tinctoria* pods are harvested when they turn brown. It will not split longitudinally on attaining maturity. Hence seed dehiscence does not occur. Average number

of days for seed maturation was 241 DAS in open condition and 251 DAS under shade.

5.3.4 Yield and yield attributes

5.3.4.1 Shoot yield

Pooled analysis showed that fresh and dry weight of shoot varied significantly among the accessions at all the three growth stages.

IT-101, IT-108, IT-106 and IT-104 were found to produce higher shoot fresh and dry weight at all growth stages irrespective of light conditions. The performance of these accessions in vegetative characters such as plant height, plant spread, number of branches, number of leaves and leaf area also showed superiority. This shows that higher yield is related to increased vegetative growth.

Fresh and dry weight of shoot was found to increase from the pre-flowering to the flowering stage and from flowering to seed maturation stage. Even though leaf fall occurs during the seed maturation stage, the thickening of shoots and increase in number of leaves from flowering to seed formation stage contributed to the increase in shoot yield during the seed maturation stage.

Open condition recorded higher fresh and dry weight of shoot compared to shade. According to Bindra and Brar (1977), out of the various physical environmental factors, the light regime is one, which has much influence on the growth and productivity of a plant. The capacity of plants to accumulate soluble carbohydrate reserves is greatly diminished under shade (Wilson, 1982). The immediate effect of low light is to reduce the carbohydrate or assimilate status of the plant and therefore, the supply of substrate available to support respiration (Lambers *et al.*, 1998).

5.3.4.2 Leaf yield

There was significant variation among the accessions for both fresh and dry weight of leaves at all growth stages on pooled analysis.

Higher leaf fresh and dry weights were produced by the accessions IT-108, IT-101, IT-106 and IT-104. These were the accessions, which dominated in the case of shoot fresh and dry weights also. Fresh and dry weight of leaf was also highest during seed maturation stage, as was the case for shoot fresh and dry weights. This can be explained by the fact that the leaf fall, which occurred during the seed maturation stage was not sufficient enough to reduce the number of leaves and leaf area in the seed maturation stage when compared to flowering stage. Leaf fall will be maximum just after the seed maturation stage.

Leaf yield was found to be reduced under shade. As evident from the data on yield, a reduction in photosynthesis might have occurred under shade resulting in a lower sugar concentration in leaves, which might have led to higher senescence under shade (Quirino *et al.*, 2000). Resmi (2001) also obtained lesser leaf yield in *Clitoria ternatea* under shade.

5.3.4.3 Pod yield

The accessions of *I. tinctoria* did not show any significant difference in fresh and dry weight of pods under open and shaded conditions. Open condition favoured the production of higher pod yield. This conforms to the findings of Usha (1990) that the grain yields of *Dolichos uniflorus* and *Vigna mungo* were less when grown as intercrop in coconut compared to the sole crop yield. Jessykutty (2003) recorded highest per plant yield in *Piper longum* under open condition compared to shade under oil palm.

5.3.4.4 Root yield

From the data on fresh and dry weight of roots it can be observed that the accessions varied significantly for fresh weight of roots under shade at pre-flowering stage and on pooled analysis at flowering stage. Significant variation for dry weight of roots among the accessions was observed under

open as well as shaded conditions at pre-flowering stage and on pooled analysis at flowering and seed maturation stages.

The accessions IT-108 and IT-101 dominated for fresh and dry weight of roots. These accessions were found to produce longer roots compared to others. This is in agreement with Resmi (2001) that the increase in root weight is proportional to the increase in root length in *Clitoria ternatea*. Open condition recorded higher root yield similar to the findings of Sunitha (1996) and Resmi (2001). The superiority in root yield under open condition can be attributed to the increased number of roots, greater root length and girth under open conditions. Geetha (2004) obtained maximum yield of *Alpinia calcarata* under full sunlight and yield decline was observed under shade.

5.3.4.5 Shelling percentage

Shelling percentage of the accessions at seed maturation stage differed significantly under both open and shaded conditions. Highest shelling percentage was recorded by IT-104, both under open and shaded conditions. Irrespective of the light conditions, IT-104, IT-108 and IT-114 were found superior in terms of shelling percentage. Lowest shelling percentage was observed in IT-99 under both conditions. IT-104 was superior in terms of fresh and dry weight of shoot and leaves during flowering stage. IT-114 was found to produce more number of branches, highest leaf area and maximum root girth at collar region at seed-maturation stage. IT-108 was superior for all vegetative and yield characters.

5.3.5 Physiological parameters

5.3.5.1 Leaf area index (LAI)

Leaf area index was found to differ significantly among the accessions at all stages of growth on pooled analysis.

Leaf area index was least in the first stage of growth, increased and was maximum during the last stage. This is in conformity with the findings of Jessykutty (2003) in medicinal plants like *Adathoda beddomei*, *Alpinia calcarata*, *Solanum indicum*, *Coleus aromaticus*, *Strobilanthes heynianus*, *Plumbago rosea*, *Pogostemon patchouli* and *Asparagus racemosus* grown as intercrops in oil palm plantations. The trend of variation in leaf area index was observed to be similar to that of leaf number and leaf yield as LAI is a function of these characters.

The observations on LAI of *I. tinctoria* at all the growth stages were found to be higher when compared to the LAI obtained in works on other crops like *Clitoria* and *Mucuna*. This may be due to the reduced spacing and higher plant density in this experiment. According to Jessykutty (2003), plant density significantly influences leaf area index. At higher population level, the leaf area produced for the given area was higher due to narrow spacing resulting in higher leaf area index.

The accessions IT-101, IT-108, IT-106 and IT-114 recorded higher leaf area index at all stages of growth. These accessions were found to produce more number of leaves and higher leaf area also. This shows that leaf area index is a function of leaf number and leaf area.

The data indicated higher values of leaf area index at all stages under open condition compared to shade. This is in conformity with the report of Bleasdale (1973) that the optimum LAI depends not only on the arrangement of leaves in the canopy but also on the light intensity that the canopy receives. Beinhart (1963) also recorded that decreased light intensity resulted in lower LAI in clover, a leguminous plant. Decrease in LAI in cassava under high shade levels have been reported by Fukai *et al.* (1984). Pushpakumari (1989) stated that coconut shade significantly influenced the LAI at all stages of growth in *Dioscorea sp.*, and *Amorphophallus peoniifolius*. Steindler *et al.* (1999) reported that the reduction in LAI under shade might be due to a change in auxin

distribution, which might have caused the inhibition of leaf production and leaf expansion under shade.

Under full sunlight, the leaves are vertically inclined. This minimises the probability of photo inhibition and increases light penetration to lower leaves in high light environments, thereby maximizing whole canopy photosynthesis, whereas under shade leaves are more or less horizontally inclined leading to canopy overlapping and mutual shading (Terashima and Hikosaka, 1995).

5.3.5.2 Leaf area duration (LAD)

Leaf area duration showed significant difference among the accessions for the period from pre- flowering to flowering stage and from flowering to seed maturation stage on pooled analysis. As in the case of LAI, IT-108, IT-106, IT-114 and IT-101 recorded higher values for leaf area duration. LAD is a function of LAI, which is related to the number of leaves. LAD appears to be an important plant trait to enhance the rate of photosynthesis.

LAD was observed to be less between pre-flowering and flowering stages and reached a maximum between flowering and seed maturation stages. This conforms to the findings of Nair (2000) and Resmi (2001) in *Clitoria ternatea*.

LAD also decreased under shade. LAD depends mainly on leaf production and senescence. Here, under shade, the leaf production was less as observed from the data on number of leaves. Shading leads to reduced sugar content in the leaf. This reduced sugar content coupled with decline in photosynthesis might have triggered the leaf senescence (Geetha, 2004).

5.3.5.3 Net assimilation rate (NAR)

Net assimilation rate varied significantly among the accessions from pre- flowering to flowering stage and from flowering to seed maturation stage on pooled analysis. Maximum NAR was observed in IT-96

(6.52 g m⁻² day⁻¹) under shade during the period from pre-flowering to flowering stage. For the period from flowering to seed maturation stage, IT-101 and IT-108 recorded higher NAR, 3.61 g m⁻² day⁻¹ and 3.44 g m⁻² day⁻¹ respectively. NAR showed a declining trend towards seed formation and seed maturation stages. Downward drift of NAR with plant age was observed by Gardner *et al.* (1988) and Resmi (2001) in *C. ternatea*.

The photosynthetic efficiency per unit leaf area (NAR) was found to be higher in shade during the period from pre-flowering to flowering stage in all the accessions. A positive trend, *i.e.* an increase in NAR with increase in shade intensity, was reported in *Ipomea batatas* and *Coleus zeylanicus* by Lalithabai (1981) and in *Xanthosoma sp.* by Valenzuela (1990). For the period from flowering to seed maturation stage, all the accessions except IT-104, IT-111, IT-97 and IT-99 recorded a decline in NAR with shade. Shading reduced NAR in potato (Nosberger and Humphries, 1965).

5.3.5.4 Crop growth rate (CGR)

Pooled analysis showed that the accessions differed significantly for CGR from pre-flowering to flowering stage and from flowering to seed maturation stage. The accessions IT-104, IT-101, IT-106 and IT-108, which produced higher NAR were also found to be superior in terms of CGR.

CGR showed an increasing trend towards the second period of growth, which corresponds to LAI. Pearce and Mitchell (1990) opined that CGR was closely related to LAI and that it could be considered as the most meaningful term for growth analysis.

Sun grown plants recorded higher CGR compared to the shade grown plants. This is in conformity with the findings of Ramadasan and Sathesan (1980) in turmeric, Fukai *et al.* (1984) in cassava, Valenzuela (1990) in cocoyam and Jessykutty (2003) in *Piper longum*.

5.3.5.5 Absolute growth rate (AGR)

There was significant difference for absolute growth rate among the accessions from pre-flowering to flowering stage and from flowering to seed maturation stage on pooled analysis. During the first period, IT-104, IT-106, IT-96 and IT-111 recorded maximum AGR, similar to CGR. From flowering to seed maturation stage, AGR was maximum for IT-101, IT-108 and IT-106 which recorded higher CGR also during the second period. AGR also showed an increasing trend towards the second period of growth, similar to CGR. This is because CGR is a function of AGR.

AGR was less under shade for all the accessions at both the periods, except for IT-114 and IT-105 for the period from pre-flowering to flowering stage. Jessykutty (2003) also observed lower AGR under shade in oil palm plantations compared to open for *Plumbago rosea*.

5.3.5.6 Relative growth rate (RGR)

Significant difference was observed on pooled analysis for relative growth rate among the accessions from pre-flowering to flowering stage and from flowering to seed maturation stage. IT-106, IT-104, IT-101 and IT-108, which showed superior performance for NAR, CGR and AGR recorded higher values for RGR also.

RGR was maximum during the period from pre-flowering to flowering stage under both conditions of light. Then it showed a declining trend towards the second period. This conforms to the findings of Haloi and Baldev (1986) that RGR was higher during the initial growth stages in chickpea. Resmi (2001) also obtained similar results in *Clitoria ternatea*.

Except for the accessions IT-108, IT-96, IT-114 and IT-105 during the initial growth period and IT-104 and IT-111 during the final growth period, RGR was found to decrease with shade. Murata (1961) reported that RGR was practically free from the influence of solar radiation as long as the level of radiation was above one third of full incident radiation. At different growth phases, significantly higher RGR was recorded in open, 20

and 40 per cent shade in ginger (Ajithkumar, 1999). Jessykutty (2003) also observed lower RGR under shade in oil palm plantations compared to open for thippali and koduveli. Okoli and Owasu (1975) observed that RGR was maximum for cocoa plants grown under medium shade.

5.3.5.7 Dry matter accumulation per plant

There was significant difference for dry matter accumulation per plant among the accessions in all the three growth stages on pooled analysis. IT-108, IT-101, IT-104, IT-106 and IT-96, which produced higher yields in terms of shoot, leaves and root were found to produce higher dry matter. This shows that dry matter accumulation follows the same pattern of response as that of final yield. According to Neerakkal *et al.* (2003), increased photosynthetic rate due to increased leaf area enhances the dry matter production.

Dry matter accumulation in open was more than that under shade in all the three stages of growth. Blackman and Wilson (1951) reported that the ability of plants to tolerate shade depends on the efficiency of total dry matter production. Highest dry matter production was noticed under open condition for *Alpinia calcarata* during last stages by Jessykutty (2003). The report of Monteith (1969) that maximum amount of dry matter production by a crop is strongly correlated with the amount of light intercepted by its foliage conforms this. Lesser dry matter production at higher shade levels was reported in plants such as cowpea by Dolan (1972) and beans by Crookston *et al.* (1975) and turmeric by Varghese (1989). Sunlight being the source of energy for plants for photosynthesis, the rate and subsequent dry matter accumulation in general was found to be adversely affected by shading. According to Bjorkman (1981) most of the sun plants have higher light saturated rate of photosynthesis, higher light compensation points and lower rates of photosynthesis at low light. This might be the reason for lower dry matter production under shade. *Aloe vera* plants grown under full sun produced numerous and larger axillary shoots, resulting in twice the

total dry mass than those grown under partial shade. Limitation in light availability, primarily affected total dry mass production and allocation (Paez *et al.* 2000).

5.3.5.8 Harvest index

Harvest index (HI) showed significant variation on pooled analysis, among the accessions for leaf yield, root yield and leaf + root yield taken together. The accessions IT-108, IT-106 and IT-101 recorded higher harvest index for leaf yield and for leaf + root yield. Harvest index for root yield was maximum for IT-97, IT-108 and IT-101. HI is related to the economic yield. Hence as the leaf and root yield increases, HI also increases.

HI for root yield was highest in open condition. This is in agreement with Susan (1989), who obtained the highest HI in ginger under open condition:

5.3.6 Phytochemical analysis

5.3.6.1 Soil N, P and K status

According to IITA (1993), the encouraging results of three decades of cover legume research in the tropics, has resulted in the promotion and increased use of legumes for the improvements of soil fertility.

The initial soil nitrogen content of the experimental site was 213.25 kg ha⁻¹ in open and 210.11 kg ha⁻¹ under shaded conditions.

Analysis of soil nitrogen content revealed that a range of 10.59 kg ha⁻¹ to 238.33 kg ha⁻¹ of nitrogen was fixed by various accessions under open. The amount of nitrogen fixed under shade ranged from 2.96 kg ha⁻¹ to 47.04 kg ha⁻¹. The amount of nitrogen fixed in the soil by an accession is given by the difference between the final and initial soil nitrogen content. Irrespective of the growing conditions, the accession IT-105 fixed comparatively higher soil nitrogen, followed by IT-101. Number of effective root nodules in these two accessions during the flowering stage was also found to be more. Nitrogen fixing potential was least in IT-97.

Jefferies *et al.* (1981) stated that nitrogen fixing plants in the family Leguminosae play an important role in providing nitrogen into nutrient poor soils. Venkataraman and Tilak (1990) reported that legumes can fix around 50 to 300 kg ha⁻¹ of nitrogen, resulting in the addition of more nitrogen to agricultural soil.

Soil nitrogen content was found to be lower under shade than in open. Low light intensity may be the reason for low nitrogen fixation under shade. The low rate of photosynthesis under coconut shade may be the reason for lesser nodule count and weight, which leads to lower nitrogen fixation. No chemical fertilizers were applied to the crop.

5.3.6.1.2 Soil phosphorus Content

Before the commencement of the experiment, the soil phosphorus content was 57.51 kg ha⁻¹ and 40.46 kg ha⁻¹ under open and shade respectively.

After the experiment, soil phosphorus status was maximum in the plot of IT-106 under open (51.97 kg ha⁻¹) and IT-96 (35.15 kg ha⁻¹) under shade. This shows that these accessions had low phosphorous uptake. Least phosphorous content was observed in the plots of IT-108 in open (27.92 kg ha⁻¹) and IT-101 in shade (22.62 kg ha⁻¹), which indicates high phosphorous uptake of these accessions. Those accessions, which had higher phosphorous uptake had better root growth in terms of root length, girth and yield. Nair (2000) reported similar observations in *Clitoria ternatea*. According to Rajasree (1994), phosphorous influence the growth and development of root system in legumes, which in turn increases rate of nitrogen fixation (due to higher nodule count and weight) and herbage yield.

Phosphorous uptake was found to be less under shade than in open. Misra and Naskar (2003) reported that when tuber crops were grown under partial shade in coconut garden, nutrient uptake followed a decreasing trend with increase in shade intensities.

5.3.6.1.3 Soil potassium content

Soil potassium content before the commencement of the experiment was 170 kg ha⁻¹ in open 120 kg ha⁻¹ in shade.

After the experiment, under open condition, the highest soil potassium content was obtained in the plot where IT-96 (140 kg ha⁻¹) was grown, which indicates low potassium uptake and the lowest potassium content in the plot where IT-108 was grown (70 kg ha⁻¹), which indicates high potassium uptake. Under shade, IT-97 recorded the highest soil potassium content (100 kg ha⁻¹) indicating low potassium uptake and IT-108, IT-101, IT-111 and IT-114 recorded lowest potassium content (60 kg ha⁻¹), showing high potassium uptake. Those accessions with high potassium uptake recorded increased nitrogen fixing capacity also. This conforms to the findings of Yahiya (1996) that grain legumes require high quantity of potassium for enhancing nodulation and nitrogen fixation.

5.3.6.2 Plant nitrogen

Pooled analysis showed significant difference among the accessions for plant nitrogen content under both open and shaded conditions. Pooled mean was highest for IT-108 (4.45%), which was on par with IT-96, IT-105 and IT-101. Analysis of *I. tinctoria* leaves gave 5.11 per cent nitrogen (CSIR, 1959). Lower leaf nitrogen content was recorded in plants grown under shade.

5.3.6.3 Glycoside estimation

Pooled analysis showed significant difference among the accessions for indican content. IT-99 recorded the highest indican content (0.47 %), which was on par with IT-96 (0.46 %), IT-114 (0.44 %) and IT-97 (0.44 %). But IT-99 was poor in the case of vegetative characters like plant spread, number of branches, number of leaves etc. Wijesekara (1993) has rightly suggested that a plant breeder should not be misled by

morphological functions in the case of medicinal and aromatic plants. He found that in *Digitalis*, there was no correlation between thick, wide and healthy looking leaves and a high content of the desired glycoside.

Plants of all accessions grown in open condition recorded higher glycoside content compared to shade. Jessykutty (2003) recorded that shade under oil palm plantation significantly influenced the oleoresin content of *Kaempferia galanga* rhizomes and the highest content was recorded under open conditions. Similar reports were made by Lambers *et al.* (1998) in tea; Jaggi and Kapoor (1997) in *Solanum laciniatum* and Li *et al.* (1996) in sage and thyme. Gupta and Chadha (1995) also reported that ample sunshine is necessary in mints at maturity to enable plants to synthesise higher content of oil and menthol.

5.3.6.4 Indigo content estimation (conventional method)

Pooled analysis showed significant difference among the accessions and between open and shaded conditions for indigo content. IT-96, IT-99, IT-114 and IT-106 recorded the maximum value of 1.67%, 1.54 %, 1.45 % and 1.39 % respectively for indigo content. IT-108 recorded the minimum (0.69 %). Shade grown plants recorded lower indigo content compared to open.

5.3.7 Correlation analysis

Yield and glycoside content are complex characters that depend on several component characters, either in a positive or negative direction. Therefore, direct selection for yield and glycoside content are often not effective. So the related characters should also be taken into account, when selection for yield is being done. Correlation provides information on the nature and extent of relationship between pairs of characters. Therefore, yield analysis in terms of genotypic and phenotypic correlation coefficient of component characters leads to the understanding of the character that can form the basis for selection. The important characters in this study are shoot

yield, leaf yield, root yield and glycoside content. The extent of association between yield and glycoside content with other characters are measured by phenotypic correlation coefficients.

5.3.7.1 Correlation between shoot yield (dry weight) and other characters

Shoot yield was found to show high positive phenotypic correlation with dry weight of leaves, leaf area index, leaf area and stem girth, but high negative correlation was exhibited by glycoside (indican) content and indigo content. The above relationship suggests that higher shoot yield is correlated with more leaf dry weight, leaf area index, leaf area and stem girth and lower glycoside content and indigo content. This is in agreement with the findings of Dwivedi *et al.* (1999b) in *Catharanthus roseus* that the total herbage yield showed positive and significant association with total leaf yield.

5.3.7.2 Correlation between leaf yield (dry weight) and other characters

High positive phenotypic correlation of leaf yield (dry weight) was observed with shoot yield (dry weight), leaf area, leaf area index, dry matter accumulation per plant and girth of stem. Negative correlation was exhibited by glycoside content and indigo content. According to Dwivedi *et al.* (1999b), total leaf yield (dry weight) showed negative and significant association with total alkaloid percentage. The findings of Wijesekara (1993) in *Digitalis* and *Citronella* also confirms this.

5.3.7.3 Correlation between pod yield (dry weight) and other characters

Pod yield showed high positive phenotypic correlation with dry weight of roots.

5.3.7.4 Correlation between root yield and other characters

High positive phenotypic correlation of root yield with girth of stem and dry weight of pods was observed.

5.3.7.5 Correlation between glycoside content and other characters

Glycoside content was found to show high positive phenotypic correlation with crude indigo content. Most of the other characters showed negative correlation with glycoside content.

5.3.7.6 Heritability, Genetic advance, GCV and PCV

The success of a breeding program usually depends upon the quantum of genetic variability present in the germplasm. The knowledge on genetic variability, heritability and genetic advance is very essential for a breeder to choose good parents and to decide the correct breeding methodology for crop improvement.

Indigo content, dry weight of root, harvest index (root yield), dry weight of leaves, net assimilation rate and dry weight of shoot showed high GCV and PCV compared to other characters. In general, PCV was higher in magnitude than GCV, which indicates considerable influence of environment on these characters.

The coefficient of variation indicated only the extent of variability present in different characters and did not indicate its heritable position. This can be ascertained from the heritability estimates in broad sense. The knowledge of heritability of a trait is helpful to enable a plant breeder to decide the selection procedure to be followed to improve the trait under given situation (Dwivedi *et al*, 1999a). For most of the characters, heritability values were high and ranged between 51.35 per cent to 99.09 per cent. Dry weight of leaves, dry weight of shoot, net assimilation rate, leaf area duration, dry weight of root, leaf area and leaf area index showed high heritability. Heritability values were low for girth of stem and dry weight of pods. The experimental materials comprised diverse ecotypes, which have not undergone any selection process. This might be the reason for higher values of heritability. Similar findings were reported by Farooqi *et al*. (1999) in glory lily.

Johnson *et al.* (1955) pointed out that heritability estimate along with high GA was more useful than the heritable values alone in predicting the resultant effect for selecting the best individual, since the heritability determines the component of heritable variation and GA measures the extent of its stability under selection. These two parameters should be considered in conjunction, so as to bring effective improvement. In the present investigation, dry weight of leaves, dry weight of root, net assimilation rate, harvest index (root yield) and crude indigo content had high heritability and high GA. Therefore these traits might be highly amenable to direct selection for their genetic improvement over a short span of time. These results are in agreement with Lal *et al.* (1999) in *Plantago* sp.

5.4 ECONOMICS OF CULTIVATION (COST/ BENEFIT RATIO) IN OPEN AND SHADE IN COCONUT GARDEN

Economic analysis is important to ascertain whether a system is sustainable or not. The best way of economically analyzing a cropping system is by way of cost-benefit analysis and calculation of net return (Hoekstra, 1985). Cost-benefit relationship in open as well as under shade in coconut garden was analysed in order to identify the prospects of cultivation as pure crop or intercrop and thereby enable the farmer to undertake this enterprise on a commercial basis.

Benefit-cost analysis showed that for every one rupee spent on the crop, the net returns was found to be 3.03 in open condition and 1.72 under shade. Thus, even though yield per plant was reduced under coconut shade compared to open condition, the cost-benefit relationship of the crops clearly suggests that *I. tinctoria* cultivation is a profitable enterprise both in open condition and under coconut shade and farmers can adopt it in a commercial basis. Thomas (2000) also obtained high B/C ratio for medicinal plants like *Plumbago rosea*, *Rauvolfia serpentina* and *Catharanthus roseus*, when grown as pure crop.

The results of the present study indicated that the accession IT-108 showed superior performance in terms of leaf yield and glycoside content followed by IT-101 and IT-96. Evaluation of shade response of *I. tinctoria* accessions indicated that there was a decline in yield with shade. Maximum yield was obtained from plants grown in open condition. A factor that decides the compatibility of crop combinations in a cropping system such as coconut culture is the ability of associated crops to come up under shade. This ability has been reported to vary widely between species of plants, there being varying degrees of yield decline in most of the common 'sun plants'. Generally shade imposes a limitation to biological productivity in plants although the extent of limitation varies with shade tolerance of the species. In the present study NAR, CGR, AGR, RGR etc. were highest in open condition, which indicates that for optimum growth it prefers open condition compared to shade. But benefit-cost analysis indicates that the crop can be cultivated as a profitable enterprise, both in open condition and under coconut shade, the net income being lesser under shaded condition.

The accessions which were found to be best in the present study may further be used for multilocational trials to evolve varieties which can be released for cultivation by farmers.

Summary

6. SUMMARY

The present study was conducted at the College of Agriculture, Vellayani during the period 2001-2003 to evaluate the genetic stock of *Indigofera tinctoria* both in open condition and under shade in coconut garden and to study the performance of selected accessions for identifying the best accession(s) yielding high biomass and glycosides. In the phase I experiment, growth and yield analysis of the genetic stock of *Indigofera tinctoria* as pure crop in open and as intercrop in coconut garden were evaluated. Thirty geographical accessions of *Indigofera tinctoria* collected from inside and outside the state were raised in lines with two replications each for open as well as shaded condition. In the phase II experiment, cultural trial of selected accessions, ten selected accessions selected based on yield and glycoside content from the phase I experiment were raised in randomized block design in open and under shade with three replications.

6.1 PHASE I EXPERIMENT

Observations on the hundred seed weight, seed colour, external appearance of seeds and germination percentage were recorded for each accession. Five accessions IT-98, IT-95, IT-105, IT-108 and IT-106 exhibited more than 80 percent germination. Of the thirty accessions collected for the study, IT-104 recorded the highest hundred seed weight of 0.56 g.

Growth and yield parameters were studied under open and shaded conditions. The accessions IT-106, IT-108, IT-113 and IT-105 recorded more plant height compared to the other accessions at all the three stages *viz.* pre flowering, flowering and seed maturation stages. Higher plant spread was observed in the accessions IT-106, IT-108, IT-96 and IT-105, irrespective of growing conditions. Under shade, at all the three growth stages, IT-104 and IT-108 produced the first branch at a greater height.

IT-108 and IT-105 produced more number of branches, irrespective of open and shaded conditions. Increased stem girth was recorded by the accessions IT-108 and IT-101 under both open and shaded conditions. Maximum number of leaves were produced by IT-104, IT-108 and IT-106 at all growth stages. Accessions that maintained a steady superiority of leaf area were IT-108, IT-106, IT-104 and IT-96. These accessions were found to produce more number of leaves, which resulted in higher leaf area. From the pre-flowering stage to flowering stage, the increment in the number of leaves and leaf area was found to be more compared to that between flowering and seed maturation stage. Under open condition, IT-96 recorded the longest root during all the growth stages. Irrespective of growing conditions, IT-108, IT-105 and IT-96 performed better in the case of root length compared to the other accessions. Those accessions that dominated in plant length and leaf production were found to lead in root length measurements. Thicker roots at collar region were observed in IT-108, IT-96 and IT-101, under both open and shaded conditions. The accessions recorded higher values for all growth parameters except height at first branching, under open conditions compared to that under coconut shade.

Pooled analysis showed that, the accessions, IT-108, IT-101, IT-106, IT-104 and IT-98 recorded higher shoot and leaf fresh and dry weights at all stages of growth, irrespective of the light conditions. These accessions were also found to produce better vegetative growth in terms of height, spread, number of branches, number of leaves etc. During the period from flowering to seed maturation stage, which covers three months, the increase in fresh and dry weights of shoot and leaves was found to be proportionately less compared to that from pre-flowering to flowering stage (two months). IT-101, IT-108, IT-106 and IT-96, which produced good vegetative growth, gave higher pod yield also. IT-106, IT-108 and IT-104, which showed good performance in terms of shoot and leaf yield, were also found to dominate in root yield. The increase in root weight was found to be proportional to the increase in root length and root girth. Shoot yield, leaf

yield, pod yield and root yield of all the accessions were found to be more under open condition compared to shade under coconut garden.

IT-99 showed superior performance in terms of glycoside content at both 280 nm and 224 nm. But the vegetative characters in terms of plant spread, number of branches, number of leaves and root length were found to be poor in IT-99. Among the different leaf treatments, maximum glycoside content was obtained from the fresh leaves. This shows that the glycoside content decreases on drying and on fermentation. Higher glycoside content was obtained in the open condition in all the accessions and for all the leaf treatments.

The ten accessions of the bluish green leaved variety of *I. tinctoria*, which showed superior performance based on selection index scores were IT-108, IT-101, IT-96, IT-114, IT-106, IT-104, IT-105, IT-111, IT-97 and IT-99.

6.2 PHASE II EXPERIMENT

The ten selected accessions from the phase I experiment were put under cultural trial under two conditions *viz.* as pure crop and as intercrop in a coconut garden. Growth and yield parameters, physiological parameters and flowering behaviour were studied under both conditions.

Pooled analysis was carried out for growth and yield parameters and physiological parameters. IT-108, IT-106, IT-96 and IT-101 showed better performance with respect to plant height compared to the other accessions at pre-flowering and flowering stages, under shade. The accessions that dominated in plant spread were IT-101 and IT-105, on pooled analysis. IT-101, IT-96 and IT-108 were found to produce the first branch at greater heights on the shoot, irrespective of the light conditions. More number of branches were produced under shade by IT-108, IT-101 and IT-114 during flowering and seed maturation stages. These accessions were found to produce higher plant height also, under shade. IT-99 and IT-104 produced more stem girth at pre-flowering stage. Plant height was

found to be less at this stage for these accessions. More number of leaves were produced under shade during the first two stages by IT-108 and IT-101. Accessions that showed superiority in leaf area were IT-101, IT-106, IT-108 and IT-114 on pooled analysis. These accessions were found to produce more number of branches and number of leaves, which resulted in higher leaf area. Under open condition, IT-101 produced the longest root during all the growth stages. Irrespective of the growing conditions, IT-108, IT-96 and IT-101 performed better in the case of root length. Root girth was maximum in IT-101 and IT-114. IT-101 also dominated in plant height, spread, number of branches, number of leaves, leaf area and root length. IT-114 also dominated in number of branches, girth of stem and leaf area. The accessions recorded higher values for all growth parameters except height at first branching, under open conditions compared to that under coconut shade.

Pooled analysis showed that, the accessions, IT-108, IT-101, IT-106, IT-104 and IT-98 recorded higher shoot and leaf fresh and dry weights at all stages of growth, irrespective of the growing conditions. The performance of these accessions in vegetative characters such as plant height, spread, number of branches, number of leaves and leaf area also showed superiority. Fresh and dry weight of shoot and leaf was found to be highest during seed maturation stage. The accessions did not show any significant difference in fresh and dry weight of pods under open and shaded conditions. The accessions IT-108 and IT-101 dominated for fresh and dry weight of roots. These accessions also produced longer roots compared to others. Observations on yield parameters of all the accessions were found to be more under open condition compared to shade under coconut garden. Highest shelling percentage was recorded by IT-104, both under open and shaded conditions. Irrespective of the light conditions, IT-104, IT-108 and IT-114 were found superior in terms of shelling percentage.

Flowering in *I. tinctoria* was observed about 111 days after sowing (DAS) to 116 DAS in open condition and about 117 DAS to 126 DAS under shade. Anthesis was spread over from 8.27 to 9.12 a.m. under open condition and from 8.46 to 10.08 a.m. under shade. Anthers mature and start dehiscing about 7-8 hours before the time of opening of corolla. The peak anther dehiscence was between 10.57 to 11.50 p.m. in open and between 11.16 to 12.18 p.m. in shade, the day before anthesis. The average pollen count per anther under open was 1733.34 and that under shade was 1561.12. The mean pollen fertility values obtained by staining method under open and shade were 61.48 per cent and 63.22 per cent respectively. Pod setting percentage was 20.91 per cent under open and 17.3 per cent under shade. Average number of days for pod set was 138 days in open and 148 days under shade. Average number of days for seed maturation was 241 DAS in open and 251 DAS under shade.

Root nodules were found to be present in the roots of *I. tinctoria* only during the pre-flowering and flowering stages of growth of the plant. Both nodule count and weight was found to be maximum at flowering stage. Number of root nodules and effective root nodules were higher for IT-101 and IT-105. Fresh weight and dry weight of root nodules were higher for IT-108, IT-101 and IT-105. IT-108 produced bigger sized nodules. Both nodule count and weight were found to be higher under open condition.

The accessions IT-101, IT-108, IT-106 and IT-114 recorded higher LAI and LAD at all growth stages. Higher NAR, CGR and RGR was observed in IT-104, IT-101, IT-106 and IT-108. NAR and RGR showed a declining trend towards seed maturation stage while CGR and AGR showed an increasing trend towards the second period of growth. Except NAR during the first growth period, all physiological parameters were higher in open condition. IT-108, IT-101, IT-104, IT-106 and IT-96, which produced higher yields in terms of shoot, leaves and root were found to produce higher dry matter. The accessions IT-108, IT-106 and IT-101 recorded

higher HI for leaf yield and for leaf and root yield. HI for root yield was maximum for IT-97, IT-108 and IT-101.

Irrespective of the growing condition, the accessions IT-105 fixed comparatively higher soil nitrogen, followed by IT-101. Soil nitrogen content was found to be lower under shade than in open. High P uptake was observed in the plots of IT-108 in open and IT-101 under shade. P uptake was less under shade than in open. High K uptake was observed in the plot of IT-108 in open and in the plots of IT-108, IT-101 IT-111 and IT-114 under shade. Pooled mean for plant N content was maximum for IT-108 followed by IT-96, IT-105 and IT-101.

IT-99 showed superior performance in terms of glycoside content. Plants of all accessions grown in open condition recorded higher glycoside content compared to shade. IT-96, IT-99, IT-114 and IT-106 recorded the maximum value for indigo content. Shade grown plants recorded lower indigo content compared to open.

The phenotypic correlation coefficient under open conditions were estimated for nineteen pairs of characters. The extent of association between yield and glycoside content with other characters was measured by genotypic and phenotypic correlation coefficients. Shoot yield was found to show high positive phenotypic correlation with dry weight of leaves, leaf area index, leaf area and stem girth and high negative correlation with glycoside content and indigo content. High positive phenotypic correlation of leaf yield (dry weight) was observed with shoot yield (dry weight), leaf area, leaf area index, dry matter accumulation per plant and girth of stem and negative correlation with glycoside content and indigo content. Pod yield showed high positive phenotypic correlation with dry weight of roots. High positive phenotypic and genotypic correlation of root yield with girth of stem and dry weight of pods was observed. Glycoside content was found to show high positive phenotypic correlation with indigo content. Most of the other characters showed negative correlation with glycoside content.

Indigo content, dry weight of root, HI (root yield), dry weight of leaves, net assimilation rate and dry weight of shoot showed high GCV and PCV compared to other characters. PCV was higher in magnitude than GCV. For most of the characters, heritability values were high and ranged from 51.35 per cent to 99.09 per cent. Dry weight of roots, crude indigo content, dry weight of leaves, net assimilation rate and harvest index (root yield) showed high heritability. Heritability values were low for girth of stem and dry weight of pods. In the present investigation, leaf area, dry weight of shoot, dry weight of leaves and dry matter accumulation per plant had high heritability and high genetic advance. Therefore these traits are highly amenable to direct selection for their genetic improvement.

In the phase I experiment, the ten accessions of the bluish green leaved variety of *I. tinctoria*, which showed superior performance based on selection index scores were IT-108, IT-101, IT-96, IT-114, IT-106, IT-104, IT-105, IT-111, IT-97 and IT-99. Benefit-cost analysis showed that, for every one rupee spend on the crop, the net returns was found to be 3.05 in open condition and 2.34 under shade. The accession IT-108 showed superior performance in terms of leaf yield and glycoside content followed by IT-101 and IT-96. NAR, CGR, AGR, RGR etc. were found to be highest in open condition, which indicates that for optimum growth it prefers open condition compared to shade. But benefit- cost analysis indicates that the crop can be cultivated as a profitable enterprise, both in open condition and under coconut shade, the net income being more under open condition.

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*Original not seen

Appendix

APPENDIX – I

Weather parameters during the crop growth period (monthly averages)

Month	Temperature (°C)		Relative humidity (%)	Total precipitation (mm)	Evaporation (mm day ⁻¹)
	Maximum	Minimum			
I year (2001-2002)					
October	30.00	24.00	83.21	256.90	3.40
November	30.39	23.49	82.42	238.10	2.90
December	30.85	22.35	79.90	20.60	2.81
January	31.10	22.20	75.65	0	3.50
February	31.60	22.30	75.80	15.00	3.70
March	32.90	23.50	75.95	16.70	4.60
April	33.20	24.80	77.85	50.60	4.20
May	31.50	25.00	82.40	200.10	3.60
II year (2002-2003)					
June	30.50	24.10	82.30	161.10	3.60
July	30.40	28.90	82.30	33.20	3.90
August	29.90	23.40	81.70	101.40	3.70
September	31.20	23.30	75.50	32.40	4.70
October	30.10	23.30	86.30	416.50	2.70
November	30.20	23.40	85.50	95.50	2.10
December	30.90	21.50	72.80	3.20	2.80
January	31.50	21.40	75.30	1.60	3.60

**EVALUATION OF NEELAYAMARI (*Indigofera tinctoria* L.)
FOR YIELD AND GLYCOSIDE CONTENT UNDER
OPEN AND SHADED CONDITIONS**

SARADA.S.

**Abstract of the
thesis submitted in partial fulfilment of the requirement
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ABSTRACT

The present study entitled "Evaluation of Neelayamari (*Indigofera tinctoria* L.) for yield and glycoside content under open and shaded conditions" was conducted in two phases viz., phase I experiment and phase II experiment. In the phase I experiment, thirty geographical accessions of *Indigofera tinctoria* collected from inside and outside the Kerala state were raised in lines with two replications each for open as well as shaded condition. Observations on hundred seed weight, seed colour, external appearance of seeds and germination percentage were recorded for each accession. Growth and yield parameters were studied under open and shade under coconut garden. Glycoside content in all the accessions were estimated using fresh leaves, shade dried leaves, oven dried leaves and fermented leaves. A selection index based on leaf yield and glycoside content was worked out to screen the genotypes.

In the phase II experiment, ten accessions, selected based on yield and glycoside content from the phase I experiment were raised in randomized block design in open and under shade with three replications. Growth and yield parameters of the selected accessions were recorded under open as well as under coconut shade similar to phase I experiment. Observations on flowering and seeding behaviour were also made. Root nodule characteristics and physiological parameters were estimated. Soil NPK analysis, plant analysis for nitrogen content and plant analysis for glycoside content were done. Correlation studies were made to find out the factors that contribute to yield and glycoside content. Economics of cultivation of *Indigofera tinctoria* both in open and under shade in coconut garden was worked out.

Five accessions IT-98, IT-95, IT-105, IT-108 and IT-106 exhibited more than 80 per cent germination. Of the thirty accessions collected for the

study, IT-104 recorded the highest hundred seed weight of 0.56 g. Higher growth parameters were recorded by the accessions IT-108, IT-106 and IT-104, irrespective of open and shaded conditions. Pooled analysis showed that these accessions dominated in the case of shoot, leaf, pod and root, fresh and dry weights also. IT-99 showed superior performance in terms of glycoside content. Among the different leaf treatments, maximum glycoside content was obtained from the fresh leaves. The ten accessions of the blue-green leaved variety of *I. tinctoria*, which showed superior performance based on selection index scores were IT-108, IT-101, IT-96, IT-114, IT-106, IT-104, IT-105, IT-111, IT-97 and IT-99.

The ten selected accessions from the phase I experiment were put under cultural trial under open and shade in phase II experiment. The accessions IT-108, IT-106, IT-104, IT-101 and IT-96 recorded higher values for all growth parameters. Pooled analysis showed that, the accessions, IT-108, IT-101, IT-106, IT-104 and IT-98 recorded higher shoot and leaf fresh and dry weights at all stages of growth, irrespective of the growing conditions. IT-108 and IT-101 dominated for fresh and dry weight of roots. All the accessions recorded higher values for almost all growth and yield parameters under open condition compared to that under coconut shade.

Flowering in *I. tinctoria* was observed about 111 days after sowing (DAS) to 116 DAS in open condition and about 117 DAS to 126 DAS under shade. Anthesis was spread over from 8.27 a.m. to 9.12 a.m. under open condition and from 8.46 a.m. to 10.08 a.m. under shade. Pod setting percentage was 20.91 per cent under open and 17.3 per cent under shade. Average number of days for pod set was 138 days in open and 148 days under shade. Average number of days for seed maturation was 241 DAS in open and 251 DAS under shade.

Root nodules were found to be present in the roots of *I. tinctoria* only during the pre-flowering and flowering stages of growth of the plant.

Both nodule count and weight was found to be maximum at flowering stage. Number of root nodules and effective root nodules were higher for IT-101 and IT-105. Fresh weight and dry weight of root nodules were higher for IT-108, IT-101 and IT-105. IT-108 produced bigger sized nodules. Both nodule count and weight were found to be higher under open condition.

The accessions IT-101, IT-108, IT-106 and IT-114 recorded higher LAI and LAD at all growth stages. Higher NAR, CGR and RGR were observed in IT-104, IT-101, IT-106 and IT-108. The accessions IT-108, IT-106 and IT-101 recorded higher HI for leaf yield and for leaf and root yield. HI for root yield was maximum for IT-97, IT-108 and IT-101. Irrespective of the growing condition, the accessions IT-105 fixed comparatively higher soil nitrogen, followed by IT-101. Soil nitrogen content was found to be lower under shade than in open. Pooled mean for plant N content was maximum for IT-108 followed by IT-96, IT-105 and IT-101.

IT-99 showed superior performance in terms of glycoside content. Plants of all accessions grown in open condition recorded higher glycoside content compared to shade. IT-96, IT-99, IT-114 and IT-106 recorded the maximum value for indigo content. Shade grown plants recorded lower indigo content compared to open.

The phenotypic and genotypic correlation coefficients under open conditions were estimated for nineteen pairs of characters. The extent of association between yield and glycoside content with other characters was measured by genotypic and phenotypic correlation coefficients. High positive phenotypic and genotypic correlation of leaf yield (dry weight) was observed with shoot yield (dry weight), leaf area, leaf area index, dry matter accumulation per plant and girth of stem and negative correlation with glycoside content and indigo content. Glycoside content was found to show high positive phenotypic and genotypic correlation with indigo content. Most of the other characters showed negative correlation with glycoside content.

Indigo content, dry weight of root, HI (root yield), dry weight of leaves, net assimilation rate and dry weight of shoot showed high GCV and PCV compared to other characters. PCV was higher in magnitude than GCV. In the present investigation, leaf area, dry weight of shoot, dry weight of leaves and dry matter accumulation per plant had high heritability and high genetic advance. Therefore these traits are highly amenable to direct selection for their genetic improvement.

Benefit-cost analysis showed that, for every one rupee spend on the crop, the net returns was found to be 3.05 in open condition and 2.34 under shade. The accession IT-108 showed superior performance in terms of leaf yield and glycoside content followed by IT-101 and IT-96. NAR, CGR, AGR, RGR etc. were found to be highest in open condition, which indicates that for optimum growth it prefers open condition compared to shade. But benefit- cost analysis indicates that the crop can be cultivated as a profitable enterprise, both in open condition and under coconut shade, the net income being less under shaded condition.