# ALLELIC RELATIONSHIPS, PENETRANCE AND EXPRESSIVITY OF GENES CONTROLLING NUMBER OF FLOWERS PER AXIS IN CHICKPEA
## (*Cicer arietinum .L*)

By

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B Sc (Ag.)

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# CERTIFICATE

**Mr. SRINIVASAN .S** has satisfactorily prosecuted the course of research and that the thesis entitled, **"ALLELIC RELATIONSHIPS, PENETRANCE AND EXPRESSIVITY OF GENES CONTROLLING NUMBER OF FLOWERS PER AXIS IN CHICKPEA (*Cicer arietinum .L.*)"** submitted is the result of original research work and is of sufficiently high standard to warrant its presentation to the examination I also certify that the thesis or part thereof has not been previously submitted by him for a degree of any University

Date -08-2005

Place Hyderabad

**(Dr. PM. GAUR)**
**Major Advisor**

# CERTIFICATE

This is to certify that the thesis entitled **"ALLELIC RELATIONSHIPS, PENETRANCE AND EXPRESSIVITY OF GENES CONTROLLING NUMBER OF FLOWERS PER AXIS IN CHICKPEA (*Cicer arietinum* .L)"** submitted in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE IN AGRICULTURE** of the Acharya N. G. Ranga Agricultural university, Hyderabad, is a record of the bonafide research work carried out by **Mr. S. SRINIVASAN** under my guidance and supervision. The subject of the thesis has been approved by the Students Advisory Committee.

No part of the thesis has been submitted for any other degree or diploma or has been published. The published part has been fully acknowledged All the assistance and help received during the course of investigations have been duly acknowledged by the author of the thesis.

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# DECLARATION

I, **SRINIVASAN .S**, hereby declare that the thesis entitled "ALLELIC RELATIONSHIPS, PENETRANCE AND EXPRESSIVITY OF GENES CONTROLLING NUMBER OF FLOWERS PER AXIS IN CHICKPEA (*Cicer arietinum* L)" submitted to the Acharya N.G. Ranga Agricultural University for the degree of **MASTER OF SCIENCE IN AGRICULTURE** is a result of original research work done by me. It is further declared that the thesis or part thereof has not been published earlier in any manner

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## ABSTRACT

Studies were carried out to investigate the allelic relationships, penetrance and expressivity of genes controlling number of flowers per axis in chickpea (*Cicer arietinum* L.) at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, A.P, India during the post rainy season 2004/05.

The material for the present investigation comprised of a double-flowered line (ICC 4929), a triple-flowered line (IPC 99-18) and a multi-flowered line (JGM 7), and six $F_1$s derived from all possible combination of crosses between these lines, and three $F_2$ populations of crosses ICC 4929 x IPC 99-18, IPC 99-18 x JGM 7 and ICC 4929 x JGM 7. These were sown in unreplicated block with a spacing of 60 x 20 cm on 9$^{th}$ October 2004. ICC 4929 has pink-veined white flower, while IPC 99-18 and JGM 7 have pink flower. Standard crop production practices were used and plant protection measures were taken to grow a healthy crop. One light irrigation was given to overcome moisture stress during early flowering stage.

The $F_1$s from double-flowered x triple-flowered cross were double-flowered, whereas $F_1$s from double-flowered x multi-flowered and triple-flowered x multi-flowered crosses were single-flowered. The $F_2$ from double-flowered x triple-flowered cross gave a good fit to a 3:1 ratio for double-flowered and triple-flowered plants. The $F_2$ from double-flowered x multi-flowered cross segregated in a ratio of 9:3:3:1 for single-flowered, double-flowered, multi-flowered and double-multi-flowered plants. The $F_2$ from triple-flowered x multi-flowered cross segregated in a ratio of 9:3:4 for single-flowered, triple-flowered and multi-flowered plants. The results clearly established that two loci control number of flowers per axis in these genotypes. The double-flowered and triple-flowered traits are controlled by a single locus (***Sfl***) and the allele for double-flowered trait (***$sfl_d$***) is dominant over the allele for triple-flowered trait (***$sfl_t$***). The multi-flowered trait in JGM 7 is controlled by a different gene (***cym***). Single-flowered plants have dominant allele at both the loci (***Sfl_ Cym_***).

Complete penetrance (100%) was recorded for double-flower, triple-flower and multi-flower traits in respective parental lines. However, variable expressivity was observed for flowering and podding traits. The highest expressivity (96.34%) was observed for double-flower trait in ICC 4929, followed by triple-flower trait (81.15%) in IPC 99-18 and multi-flower trait (51.33%) in JGM 7. the double-pod trait showed higher expression (76.36%) than multi-pod trait (24.7%). The triple-flower line IPC 99-18 did not produce triple-pod at any of the nodes thus the expressivity of triple-pod trait was 0.00%. Average number of flowers per axis and average number of pods per axis were higher in multi-flowered line JGM 7 than other flowering types.

Inheritance studies on flower colour showed the presence of a single gene (***Ifc***) that inhibits the flower colour. This gene in homozygous recessive condition (***ifc ifc***) changes the pink colour petals to white colour without changing the vein colour. Similarly green foliage colour was found dominant over purple foliage. Thus monogenic inheritance was confirmed for two morphological traits, flower colour and foliage colour. Joint segregation analysis showed the presence of a common gene that exhibit pleiotropic effects on flower and foliage colours.

Variability studies showed significant differences among flowering types for characters number of flowers per axis, number of pods per axis, yield per plant and 100-seed weight. Double-multi and multi-flowering types were superior to other flowering types in yield per plant and 100-seed weight.

## LIST OF ABBREVIATIONS

| | | |
|---|---|---|
| 1. | Centimeter | cm |
| 2. | Chi-square | $\chi^2$ |
| 3. | Degree centigrade | °C |
| 4. | Gram | g |
| 5. | Hectare | ha |
| 6. | Hour | hr |
| 7. | Kilogram | kg |
| 8. | Metre | m |
| 9. | Millimetre | mm |
| 10. | Percentage | % |
| 11. | Recombination frequency | r |
| 12. | Standard error | SE |
| 13. | centi Morgan | cM |
| 14. | Random Recombinant Inbred Lines | RILs |

# Chapter I
# Introduction

CHAPTER I

# INTRODUCTION

Chickpea (*Cicer arietinum* L.), a diploid (2n = 2x = 16) member of the family Leguminosae and subfamily Papilionoideae, is the most important pulse crop of South Asia and the third most important pulse crop of the world. It is currently grown on about 10 4 million hectares with production of 8.6 million tons, of which 95% cultivation is in the developing countries (FAOSTAT data 2005).

India is the largest producer of chickpea, accounting for 62.6% of the area (6.50 m ha) and 67.3% of the production (5.77 million tons) of chickpea in the world (FAOSTAT data 2005). Chickpea is known in various parts of the country by different names such as *chana*, *gram*, *chhola* and *Bengal gram*.

Chickpea originated in the temperate regions of southeastern Turkey and adjoining Syria (van der Maesen, 1987). Subsequently two distinct subtypes were evolved. The small, dark-seeded *desi* variant is adapted to South Asia. The large, cream-seeded type called *Kabuli*, predominates across West Asia and North Africa (WANA), America and Europe (van der Maesen, 1987).

In India, chickpea is cultivated in winter season under both, rainfed and irrigated conditions. States like Madhya Pradesh, Rajasthan, Uttar Pradesh, Maharashtra,

Andhra Pradesh, Karnataka, Bihar and Haryana contribute about 95% of the total chickpea production of the country

Andhra Pradesh (6 02%) and Karnataka (4 78%) have registered highest growth rate in productivity during the past decade In Andhra Pradesh, due to the adaptation of short-duration high-yielding varieties, the chickpea area has increased about 5-fold (from 60,000 ha to 288,000 ha) and the production 13-fold (from 28,000 t to 363,000 t) during 1993 to 2002 The yield has increased from 500 to 1300 kg/ha during this period

Chickpea grains provide about 18-24% protein, 4-10% fat, 52-70 9% carbohydrate, 10-23% fiber, minerals, and vitamins Among essential amino acids, lysine, methionine, threonine, valine, isolucine and leucine are major components of seed protein Chickpea contains considerable amount of vitamins such as $B_1$ and $B_2$, ascorbic acid (vitamin C) and niacin Thus, chickpea plays an important role in human nutrition also

The presence of nitrogen fixing bacteria in its roots improves the soil fertility by way of fixing atmospheric nitrogen The deep roots help in opening up the soil to the much deeper strata, which also help in improving soil aeration resulting in better root development and microbial activities in soil

Since decades India ranks first in area and production of chickpea but the average productivity is very low (790 kg/ha) compared to potential yield (5000 kg/ha), this lag mainly

due to several constraints like unstable environmental influence on expression of flowering, long duration of crop season in some areas apart from abiotic and biotic stresses.

The use of markers in a crop cultivar gives an added advantage in characterizing, selection and in maintaining the genetic purity. Several morphological markers, such as flower colour and pigmentation, are used in different crops. Genetics of flower colour and pigmentation are not well understood as also for many other traits in chickpea

Most efficient way of improving crop yield is through hybrid seed production using male sterile lines. In legumes like beans and peas, there are many seeds in a single pod but in chickpea, each pod produces only a single or two seeds. There are many constraints to the production of hybrid seed in chickpea, i.e. <1% cross pollination (Tayyar et al, 1995) due to cleistogamous flowers, low seed to seed multiplication, and high seed rate required for sowing. Thus, the only approach available for chickpea improvement is the development of an improved plant type by improving traits that contribute to yield potential. The major yield contributing traits in chickpea are number of pods per plant, number of seeds per pod and seed size.

Most of the chickpea germplasm accessions produce a single flower at each flowering node, but some lines produce two, three or more flowers per axis and have the potential to form more than two pods per peduncle, As the sink capacity is more in double, triple and multi-podded plants, exploitation of multi-pod trait may be considered in developing high yielding cultivars. It has been found that there is a yield advantage of double-pod trait over

single-pod trait (Sheldrake et al , 1978, Singh and Rheenen, 1994, Kumar et al , 2000) Meanwhile, triple-flowered and multi-flowered lines have also been identified The multi-flowered plants produce more than two pods at many nodes Preliminary evaluation of this trait showed yield advantage from more number of pods per plant However, more studies would be necessary to examine the effect of multi-flower (multi-pod) trait on seed size and exclusively demonstrate the value of this trait in chickpea breeding (Gaur and Gour, 2002)

A better understanding of the genetics of number of flowers per axis and allelic relationships of genes governing various flowering types will help in improving efficiency of breeding programmes for exploiting these traits The available information on inheritance and linkage of traits is rather limited Thus, the present study was conducted to investigate the inheritance and allelic relationships of genes controlling number of flowers per axis, flower colour and foliage colour with the following objectives

(1) To determine allelic relationships, penetrance and expressivity of genes controlling double, triple and multi-flower traits

(2) To assess the effects of double, triple and multi-flower traits on seed size and grain yield

# Chapter II
# Review of literature

CHAPTER II

# REVIEW OF LITERATURE

The existence of wide morphological diversity in chickpea (*Cicer arietinum* L.) offers ample opportunity for genetic studies. Qualitative characters being less influenced by environmental variations are used widely as markers for quantitative traits. The literature available on genetics of chickpea traits relevant to this study has been reviewed in this chapter

## 2.1. Inheritance

### 2.1.1. Number of flowers per axis

Chickpea (*Cicer arietinum* L.) has a racemose type of inflorescence and most germplasm lines/cultivars produce a single flower at each axis of the raceme. Several germplsm lines/cultivars are known which produce two flowers per axis. Of the 12018 germplasm accessions evaluated at ICRISAT, 100 were double-flowered (Pundir *et al.* 1985).

Varieties having two flowers per pedicel were first described by Shaw and Khan (1931). Later many researchers have studied the inheritance of this trait. Khan and Akhtar (1934) were the first to study genetics of single and double-flower traits and reported that singleness was due to a factor "*S*" dominant to double-flower trait.

Bhapkar and Patil (1962) and Ahmad (1964) also observed dominance of single-pod trait over double-pod trait. They suggested that yielding ability of *kabuli* chickpea could possibly be improved by introducing double-pod trait from *desi* chickpea.

Spontaneously occurring double-flowered mutants have been reported by several researchers. Singh (1965) identified a double-flowered mutant, which gives rise mostly to two unusual smaller pods fused at the base with pear shaped appearance. Similarly, Pundir and van der Maesen (1981), and Rao and Pundir (1983) also reported double-flowered mutants. Recessive nature of double-flower mutant trait was confirmed in all these studies.

Genetics of double-flower trait was also studied by Patil (1966), D'Cruz and Tendulkar (1970), More and D'Cruz (1976), ICRISAT (1977), Yadav *et al.*, (1978), Rao *et al.*, (1980), Pawar and Patil (1983), Singh and van Rheenen (1994), Kumar *at al.*, (2000a) and Gaur and Gour (2002). These studies further confirmed that the double-floweredness is controlled by a recessive allele. D'Cruz and Tendulkar (1970) proposed a gene symbol "*sfl*" for the recessive gene for double-flower trait. It appears that "*s*" (earlier proposed by Khan and Akhtar 1934) and "*sfl*" represent the same gene.

Mutants have been identified that produce more than two flowers per axis. First report on triple-flower trait was by Singh and Chaturvedi (1998). They demonstrated that triple-flower trait is controlled by a single recessive gene "*rtf*". The allelic relationship between "*s*" or "*sfl*" and "*rtf*" is not known.

Gaur and Gour (2002) identified a spontaneous mutant that produces 3 to 9 flowers, arranged in a cymose inflorescence, at many axis of the raceme. The mutant was isolated from $F_2$ population of an interspecific cross ICC 5783 (*C. arietinum*) x ICCW 9 (*C. reticulatum*) in

which both the parents involved were single-flowered. The number of pods set varied from 0 to 5 in each cyme. Inheritance studies indicated that a single recessive gene, designated '*cym*', was responsible for cymose inflorescence. The allelic relationship of *cym* and *sfl*, a gene for double-flower trait, was studied from a cross involving multi-flowered plants and the double-flowered line ICC 4929. The *cym* gene was not allelic to *sfl*, suggesting that two loci control the number of flowers per peduncle in chickpea. The *cym* locus segregated independently of the loci *sfl*, *ifc* (inhibitor of flower color) and *blv* (bronze leave).

**2.1.2 Flower colour**

Flower colour is the most important diagnostic character in chickpea and is widely used as a marker in genetic studies and breeding work. Observations on the largest assemblage of chickpea material at ICRISAT recognize three main flower colours in chickpea: pink in 71.0%, white in 18.9% and light pink in 9.4% of the germplasm accessions. The other flower colours (dark pink, blue and light blue) occur in smaller proportion of germplsm. Geographically, the pink flower colour dominates in the Indian subcontinent and the white flower colour in the Mediterranean and Andean regions, and Mexico (Pundir *et al.*, 1985).

Pimplikar (1943), Khan *et al.* (1950), Argikar (1955), Argikar and D'Cruz (1963), Bhapkar and Patil (1963), More (1964), Patil (1964), Athwal and Brar (1967), Patil (1967), Khosh-Khui and Niknejad (1971), Mian (1971), Jagtap *et al.* (1973), More and D'Cruz (1976b), Nayeem *et al.* (1977), Reddy and Nayeem (1978), Yadav *et al.* (1978), Pawar and Patil (1982 and 1983), Kidambi *et al.* (1988), Singh *et al.* (1988), Gil and Cubero (1993) and

*Pundir and Reddy (1997), Tefera (1998), Sabaghpour (2000) and Kiran kumar (2001)* reported that a single locus is responsible for pink and white/blue/light velvet colours in chickpea.

Khan and Akhtar (1934) reported that flower colour is controlled by two pairs of genes (*P* and *B*) in chickpea. The genotypes *P_ B_*, *p p B_*, *P_ b b* and *p p b b* produce pink, blue, white and white flowers, respectively. The $F_2$ gave a ratio of 9 pink : 3 blue : 4 white-flowered plants. Pal (1934) and Kadam *et al.* (1941) observed similar segregation ratio (9:3:4) for pink, salman and white flower colours. More and D'Cruz (1970, 1976a) and Patil and Deshmukh (1975) also suggested that flower colour in chickpea is controlled by two loci, *Sco* and *Bco*.

Deshmukh *et al.* (1972) proposed two loci *Lvco* and *Wco* for flower colour. It was suggested that the genotypes *Lvco Wco_*, *lvco lvco Wco_*, *Lvco wco wco* and *lvco lvco wco wco* produce pink, violet, white and white flowers, respectively. Reddy and Chopde (1977a) reported that two complementary genes, designated *Pcoa* and *Pcob*, control flower colour in chickpea with pink dominant to violet. Pawar and Patil (1979) and Rao *et al.* (1980) found digenic inheritance of flower colour giving an $F_2$ ratio of 9:3:3:1 for pink, light pink, blue and light blue-flowered plants. Ghatge and Kolhe. (1985) and Ghatge (1994a) reported that two dominant genes *Pco* and *Bco* are needed for expression of pink flower colour.

Interaction of three genes in controlling the flower colour was observed by Ayyar and Balasubramaniyan (1936). They reported that genes *C, B* and *P* are responsible for this trait. *B* and *C* are complementary and P is supplementary to *B*. All three genes in dominant condition

produce pink flowers (*C B P*) and when *B* and *C* alone are present blue flowers are produced (*C B pp*). In all other combinations (*CC bb PP, CC bb pp, cc BB PP, cc BB pp, cc bb PP*, and *cc bb pp*) flowers are white. Similarly, D'Cruz and Tendulkar (1970) used the gene symbols *Pcoa, Pcob1* and *Pcob2*; Phadnis (1976) used the gene symbols *A, B* and C; Vijayalakshmi (1998) and Kumar *et al* (2000b) used the gene symbols C, B and P for explaining the three-factor inheritance of petal colour in chickpea.

Gaur and Gour (2001) studied the inheritance of pink-veined white flower colour, which is rare in chickpea. They reported a recessive gene (*ifc*) which inhibit flower colour without affecting vein colour of the corolla in pink-veined white flowered accession ICC 4929. Results indicated that homozygous recessive condition (*ifc ifc*) changes the pink flower colour (*P P B B Ifc Ifc*) to pink-veined white colour (*P P B ifc ifc*) and of blue flower colour (*p p B Ifc Ifc*) to blue-veined white colour (*p p B ifc ifc*). Thus, indicating the presence of three factors in controlling petal colour in chickpea.

### 2.1.3. Foliage colour

The anthocyanin pigments impart purplish colour to different parts of the plant. The intensity of pigmentation is influenced by light. The *kabuli* types lack anthocyanin; their stems and foliage are green. It appears that chickpea cultivars with higher anthocyanin content are relatively more stable and yield better as often acknowledged by Indian and Ethiopian farmers (Pundir and Mengesha 1983). Anthocyanin pigmentation is a good genetic marker and low anthocyanin content is dominant over high anthocyanin and zero anthocyanin contents (Rao *et al.*, 1980). Of the chickpea germplsm available in gene bank at ICRISAT, 67.1% is low in

anthocyanin and 32.4% does not have it. The remainder (0.5%) of the collection is high in anthocyanin; leaves and stems are dark purple (Pundir *et al.*, 1985).

Monogenic inheritance of anthocyanin pigmentation was reported by Bhapkar and Patil (1962), Rao *et al.* (1980), Ghatge (1993), Tefera (1998) and Kiran Kumar (2001). However, digenic inheritance was proposed by Ghatge and Kolhe (1985), Mathur (1989) and Sandu *et al.* (1993).

Mathur (1989) reported that development of purple pigmentation on plants depends on light. The chickpea line that he studied developed purple pigmentation in the whole plant when exposed to sunlight, while no pigmentation was developed in absence of light. He further (1998) suggested that two complementary genes control light dependent purple pigmentation.

There are variable reports on dominance of pigmentation trait. Non-pigmented condition (normal green) was reported to be dominant to pigmented (purple) by Bhapkar and Patil (1962), Rao *et al.* (1980), Sandu *et al.* (1993) and Kiran Kumar (2001), whereas dominance of pigmentation to normal green condition was reported by More and D'Cruz (1976), Ghatge (1994a), Mathur (1998) and Tefera (1998).

## 2.2. Linkage

The marker-assisted selection for genes of economically important traits with easily identified markers can improve the efficiency of breeding and hasten the development of

cultivars. There are numerous reports on identification of linkages between qualitative traits and also between qualitative and quantitative traits in chickpea. The reports on the linkage relations of characters under study are summarized here.

Aziz *et al.* (1960) reported that corolla colour (*P*) was linked with seed surface (*W*) and seed coat colour (*R*) with crossover percentages of 18.2 and 13.8 between P and *W* and 18.1 and 30.4 between *P* and *R* in two crosses of chickpea. Similarly corolla colour was linked with loci for *chrysanthemum*-like leaves (*nls*) with a map distance of 28.3 cM (Reddy and Chopde 1977a) and with seed coat thickness with a map distance of 19 cM (Gil and Cubero 1993).

Corolla colour (*Lvco)* was found linked with one of the loci (*Rsa* or *Rsb*) for type of seed surface (Deshmukh et al. 1972, and Pawar and Patil 1983), with one of the loci ($Bsc_a$ or $Bsc_b$) for seed coat colour (Reddy and Chopde 1977b), with locus for number of flowers per axis (*Sfl*) (Pawar and Patil 1983 and Singh *et al.* 1988), with one of the loci controlling wilt resistance (Singh *et al.* 1988) and with the locus (*fil*) for filiform leaf trait (Davis 1991). Rao and Pundir (1983) reported a three lobed vexillum mutant in which they observed a loose linkage between double flowered peduncle and lobed vexillum.

Pleiotropic effects of gene (*Pco*) or one of the genes (*Pcoa*, $Pcob_1$ and $Pcob_2$) governing corolla colour on other qualitative traits was reported by many researchers. The reports on pleiotropic effects included stem and corolla colour (D'Cruz and Tendulkar 1970); corolla colour, vein colour, stem colour, seed coat colour, pedicel colour and seed shape

(Reddy and Nayeem 1978); and stem colour, pedicel colour, corolla colour and colours of veins on the ventral surface of the standard petal (Aher and Patil 1984). Ghatge *et al* (1985) reported the presence of a common gene (*Bco*) for corolla colour (*Pco* and *Bco*) and seed coat colour (*Bco, Gsc, Ycot, Blsc$_a$* and *Blsc$_b$*). Later Ghatge (1994b) reported that the gene (*Bco*) also shows pleiotropic effects on stem colour.

Pundir and Reddy (1997) reported a natural mutant (designated ICC 17101) that combines purplish stem (low pigmented) with white flower. This trait combination was not previously known in *Cicer*.

## 2.3. Penetrance and Expressivity

Singh *et al.* (1986) investigated the extent of proportion of double-podded nodes among advanced generations ($F_5$, $F_6$ and $F_7$) of double-podded progenies of chickpea. The percentage of double-podded nodes varied from 28.5% to 80.2% in $F_5$, 29% to 83.2% in $F_6$, and 33.6% to 77.6% in $F_7$ generation. They opined that higher proportion of double-podded nodes may add to the increase and stability in seed yield.

Sheldrake *et al.* (1978) observed that there was no significant difference in expression of double-pod character, at Hyderabad ($17^0$32'N) and at Hissar ($29^0$10'N). Though difference was not significant, percentage of double-podded nodes were higher at Hyderabad than at Hissar.

Kumar *et al.* (2000a) studied penetrance and expressivity of the gene for double podding in $F_2$ population and RILs of the cross ICCV 2 (single-podded) x JG 62 (double-podded). The penetrance of homozygous recessive (*ss*) gene responsible for production of double-flowers per peduncle showed unstable penetrance (53% in $F_2$ and 84.5% in RILs) and variable expressivity (1.1-14.8% in $F_2$ and 0.1-33% in RILs).

## 2.4. Contribution of double-pod trait to yield and its stability

There are variable reports on yield advantage of double-flower/ double-pod trait in chickpea. Sheldrake *et al.* (1978) studied effects of double-pod traits by converting double-podded nodes to single-podded by removing one flower from each node and reported that double-pod trait could confer a yield advantage of about 6-11% under conditions in which the character is well expressed, for example, terminal drought in peninsular India. Singh and van Rheenen (1989, 1994) observed an yield advantage due to double-floweredness in their study. The mean yield of double-podded plants (JG 62 – 10.2 g) was higher than that of single podded plants (MS 24 – 9.4 g). Rubio *et al.* (1998) found a positive effect of the double-pod gene on the stability of yield in chickpea. In the studies of Kumar *et al.* (2000a), the seed yield advantage of the double-pod trait over the single-pod trait was 18% in $F_2$ and 7% in the RILs. The increased number of pods and seeds contributed to the higher yield in double-podded plants. However, there was a slight decrease in seed size of the double-podded genotypes.

There are also reports about the negative effect or no significant effect of double-pod trait on yield contributing characters. Knights (1987) conducted an experiment to quantify the effects of double-pod gene by comparing single and double-podded genotypes involved in

three sets of inbred lines. Results showed that mean yield and seed mass of double- podded lines was significantly less than single podded-lines in two of the trails. Rubio *et al* (1998) found no significant difference between double and single-podded lines for seeds per plant (general mean for SP=79 and for DP=75) and yield (SP=137g/$m^2$ and DP=131g/$m^2$). They concluded that double-pod character would not decrease the seed size in chickpea, which was contrary to the report on negative effect of double-pod trait on seed size given by Knights (1987).

# Chapter III
# Materials and Methods

CHAPTER III

# MATERIALS AND METHODS

The present investigation was undertaken to study allelic relationships, penetrance and expressivity of genes controlling number of flowers per axis in chickpea (*Cicer arietinum* L.). In addition, investigation was also done to understand the association between number of flowers per axis and yield-contributing characters. The experiment was conducted during *Rabi* (post rainy) season 2004 in the field BM 14D at ICRISAT, Patancheru, A.P., which is situated at an altitude of 545m above the mean sea level. Geographically it lies at a latitude of 17°27'N and a longitude of 78°28'E. The weather data for crop growth period are given in Appendix I.

## 3.1. MATERIALS

The experimental materials for present investigation comprised of three chickpea lines (ICC 4929, IPC 99-18 and JGM 7), $F_1$s from all possible cross combinations between these lines, and three $F_2$ populations derived from one-way crosses (ICC 4929 x IPC 99-18, IPC 99-18 x JGM 7 and ICC 4929 x JGM 7). As the maternal effects were not observed in any of the cross combinations for the traits under study, $F_2$ populations from reciprocal crosses were not studied. Details of the four parents are presented in Table 3.1.

## 3.2. METHODS

### 3.2.1. Field experiment

The seeds from the parents, $F_1$s and $F_2$s were sown in an unreplicated block at a wider spacing of 60 x 20 cm with single seed per hill. The experimental materials were sown at two locations to asses effects of environment on expressivity of traits. The locations were ICRISAT, Patancheru in southern India and Indian Institute of Pulses Research (IIPR), Kanpur in northern India. The experiment at IIPR had to be abandoned, as there was severe plant mortality due to fusarium wilt. The sowings at ICRISAT were undertaken in vertisols on 9$^{th}$ October 2004. All recommended package of practices, including basal application of fertilizer (DAP 100 kg.ha$^{-1}$), one intercultural operation to control weeds and three sprays of pesticide to manage pod borer, were followed to raise a healthy crop. One light irrigation was given during early flowering stage to overcome moisture stress conditions

Table 3.1: Description of parental lines used in the study

| **Character** | **ICC 4929** | **IPC 99-18** | **JGM 7** |
|---|---|---|---|
| Origin | Punjab, India | IIPR Kanpur | JNKVV Jabalpur |
| Varietal status | Land race | Mutant line | Mutant line |
| Growth habit | Semi erect | Semi erect | Semi spread |
| Flower colour | Pink-veined white | Pink | Pink |
| Foliage colour | Normal green | Light green | Bronze or purple |
| Flowering type | Double-flowered | Triple-flowered | Multi-flowered |
| Seed type | *Desi* | *Desi* | *Desi* |
| Seed coat colour | Yellow brown | Yellow brown | Yellow brown |
| Reaction to Fusarium wilt | Susceptible | Resistant | Susceptible |
| Maturity | Late | Late | Late |

## 3.3. Recording of observations:

The observations were recorded on all individual plants in each $F_2$ population (around 330 plants in ICC 4929 x IPC 99-18, 360 plants in IPC 99-18 x JGM 7, and 360 plants in ICC 4929 x JGM 7) and around 40 random plants in each parental line for the following traits:

### 3.3.1. Type of flowering

Each $F_2$ plant was classified as single-flowered, double-flowered, triple-flowered, multi-flowered or double multi-flowered depending on the expression of trait on at least 20% of flowering nodes at maximum flowering stage.

### 3.3.2. Number of flowers or pods per node:

In chickpea, the flowering and podding stages overlap and continue for over a month due to indeterminate growth habit. Thus, each flowering node needs to be tagged and the plants need to be visited several times for recording observations on number of flowers per axis on the whole plant. This is very difficult for the study of large $F_2$ populations. Thus, two random branches were selected from each plant and observations were recorded on number of flowers per axis i.e. one, two, three or multi (more than three) on five consecutive nodes at the time of maximum flowering stage. These five nodes in each branch were tagged for further recording podding data during crop maturity.

**3.3.3. Number of pods per node:**

Podding data was recorded on the nodes, which were used for recording flowering data, i.e. one, two, three, and multi (more than three pods), at crop maturity.

**3.3.4. Flower colour**

Colour of the freshly opened flower was recorded as pink or pink-veined white flower.

**3.3.5. Foliage colour**

Colour of the foliage was recorded based on the visual observation on intensity of pigmentation. The classes observed were light green, normal green and purple.

**3.3.6. Number of filled pods per plant**

The total number of filled pods in each plant were counted at maturity.

**3.3.7. Number of unfilled pods per plant**

The total number of pods without seed in each plant were counted at maturity.

**3.3.8. Number of seeds per plant**

The total number of seeds were counted in each plant after threshing the dried filled pods.

**3.3.9. Number of Seeds per pod**

Number of seeds per pod was calculated by using the formula:

$$\frac{\text{Number of seeds per plant}}{\text{Number of filled pods per plant}} \cdot X\ 100$$

**3.3.10. Seed yield per plant (g)**

Weight of the total seed of a single plant was taken in grams.

**3.3.11. 100 seed weight (g)**

Weight of 100 seed in grams for individual plants was calculated by using the formula:

$$\frac{\text{Seed yield per plant (g)}}{\text{Number of seeds per plant}} \cdot X\ 100$$

## 3.4. Statistical analysis

The data was subjected to the following statistical analyses using the standard statistical procedures

3.4.1. Goodness-of-fit chi-square test and linkage analysis

3.4.2. Penetrance

3.4.3. Expressivity

3.4.4. T-test for significance of differences between means

**3.4.1. Goodness-of-fit chi-square test and linkage analysis**

The inheritance of various qualitative traits was studied using the computer programme "LINKAGE-1" (Suiter et al., 1983), which tests goodness-of-fit to expected

ratios by chi-square analysis, detects the presence of linkage using a contingency chi-square test and calculates recombination fraction (r) and its standard error (SE) using maximum likelihood formulae. For further confirmation of results, data was also subjected to another statistical package "JOINMAP" which detects linkages and make linkage groups.

### 3.4.2. Penetrance

Penetrance is defined as the frequency (expressed as percentage) with which individuals of a given genotype manifest at least some degree of a specific mutant phenotype associated with a trait. i.e. - the % of individuals expressing a trait. The level of penetrance can be calculated as the proportion of individuals with a given genotype who exhibit a particular phenotype. When all individuals of a particular genotype have the same phenotype, the gene shows complete penetrance and the level of penetrance is defined as 1.0. The presence of incomplete penetrance at a gene may cause a phenotype to skip a generation in a pedigree.

Penetrance of gene for double-flower (DF), triple-flower (TF) and multy-flower (MF) traits was calculated by using formula:

$$\frac{\text{Number of observed DF or TF or MF plants}}{\text{Number of expected DF or TF or MF plants}} \times 100$$

To know the probability limits at 1 degree of freedom, chi-square test was performed while explaining penetrance.

### 3.4.3. Expressivity :

The expressivity is defined as the degree to which a trait is expressed in an individual. It can vary from "hardly noticeable" to severe.

#### 3.4.3.1. Expressivity of double-flower trait:

Expressivity of genes for double-flower trait was calculated by using the formula:

$$\frac{\text{Number of double-flowered nodes}}{\text{Number of flowering nodes}} \cdot X\ 100$$

#### 3.4.3.2. Expressivity of triple-flower trait:

Expressivity of genes for triple-flower trait was calculated by using the formula:

$$\frac{\text{Number of triple-flowered nodes}}{\text{Number of flowering nodes}} X\ 100$$

#### 3.4.3.3. Expressivity of multi-flower trait:

Expressivity of genes for multi-flower trait was calculated by using the formula:

$$\frac{\text{Number of multi-flowered (3 or more flowers) nodes}}{\text{Number of flowering nodes}} \times 100$$

**3.4.3.4. Expressivity of double-pod trait:**

$$\frac{\text{Number of double-podded nodes}}{\text{Number of podding nodes}} \times 100$$

**3.4.3.5. Expressivity of triple-pod trait:**

$$\frac{\text{Number of triple-podded nodes}}{\text{Number of podding nodes}} \times 100$$

**3.4.3.6. Expressivity of multi-pod trait:**

$$\frac{\text{Number of multi-podded (3 or more pods) nodes}}{\text{Number of podding nodes}} \times 100$$

NOTE: In the above formulae for calculating the expressivity of flowering and podding traits, the observations were recorded on 10 nodes per plant as described earlier.

### 3.4.4. Test of significance of differences between means

Test of significance is a procedure for distinguishing whether the observed difference connotes any real difference among groups or can be ascribed to mere sampling fluctuations. It helps in making due allowance for the sampling fluctuation affecting the results of experiments or observations. 't' and 'Z' tests (based on the sample size) were used to test the significance of differences between two means for yield contributing characters and seed yield between parents and between crosses, respectively (Kapur and Saxena, 1969). When the p-value associated with t was low (< 0.05), the null hypothesis was rejected and it was concluded that there is significant difference between means under comparision. The Windows-based statistical package "GENSTAT" was used for conducting these tests.

# Chapter IV
# Results

CHAPTER IV

# RESULTS

An experiment was conducted in the *Rabi* (post rainy season) 2004/05 to study the allelic relationships, penetrance and expressivity of genes controlling number of flowers per axis in chickpea (*Cicer arietinum* L.). The effects of various types for number of flowers per axis (single-flower, double-flower, triple-flower and multi-flower) on grain yield and its components were also assessed. The experimental materials included three chickpea lines, ICC 4929 (double-flowered), IPC 99-18 (triple-flowered) and JGM 7 (multi-flowered); $F_1$s from all possible crosses (including reciprocals) between three lines; and three $F_2$ populations from one way crosses (ICC 4929 x IPC 99-18, IPC 99-18 x JGM 7 and ICC 4929 x JGM 7).

The data were recorded on all $F_2$ plants (around 340) in each cross and around 40 random plants in each parental line for three qualitative (flowering type, flower colour and foliage colour) and eight quantitative attributes (number of flowers per axis, number of pods per axis, number of pods per plant, number of seeds per plant, grain yield per plant, 100-seed weight, number of seeds per pod and number of empty pods per plant). The results are presented in this chapter under the following heads.

4.1. Penetrance of flowering types

4.2. Expressivity of flowering/podding types

4.3. Allelic relationships of genes controlling different flowering types

4.4. Inheritance of flower colour

4 5 Inheritance of foliage colour

4 6 Genetic linkages

4 7 Genetic variability among flowering types for various characters

## 4.1. Penetrance of flowering types

The penetrance of the genes for double, triple and multi-flower traits was calculated from the parental lines ICC 4929, IPC 99-18 and JGM 7, respectively

Penetrance was found to be 100% for each of these traits in respective parental line

## 4.2. Expressivity of flowering /podding types

### 4.2.1. Expressivity of flowering types

Expressivity of the genes for double, triple and multi-flower traits was calculated in respective parental line

#### 4.2.1.1. Double-flower

ICC 4929 (double-flowered parent) showed mean expressivity of 96 34% for double-flower trait The range was between 60% to 100% The mean number of flowers per node was 1 96 and ranged between 1 7 to 2 0 (Table 4 1) (Plate No 4 1)

#### 4.2.1.2. Triple-flower

IPC 99-18 (triple-flowered parent) showed mean expressivity of 81 15% for triple-flower trait The range was between 20% to 100% The number of flowers per node ranged between 1 9 to 3 0 with an average of 2 8 (Table 4 1)

#### 4.2.1.3. Multi-flower

JGM 7 (multi-flowered parent) exhibited mean expressivity of 51 33% for multi-flower trait The range was between 0% to 80% The average number of flowers per node was found to be 2 83 and ranged between 1 2 to 4 1 (Table 4 1) (Plate No 4 2)

### 4.2.2. Expressivity of podding types

Expressivity of double-pod, triple-pod and multi-pod was studied in double, triple and multi-flowered parents, respectively (Plate No 4 3)

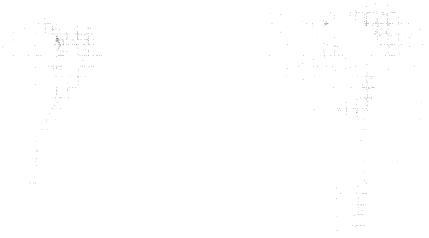
#### 4.2.2.1. Double-pods

ICC 4929 (double-flowered parent) showed mean expressivity of 76 36% for double-pod trait and the range was between 20% to 90% The mean number of pods per node was found to be 1 77 and ranged between 1 8 to 1 9 The number of nodes without pods accounted for 6 83% (Table 4 1)

#### 4.2.2.2. Triple-pods

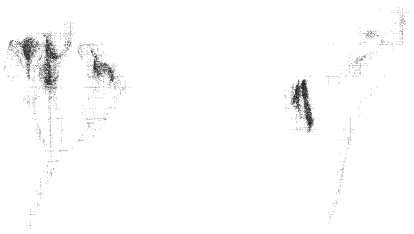
IPC 99-18 (triple-flowered parent) exhibited mean expressivity of 0 00% for triple-pod trait, i e no single node was found with three pods in this parent The average number of pods per node was 1 44 and it ranged between 0 7 to 1 9 In this parent, the proportion of double-podded nodes (55 3%) was more than single-podded nodes (44 7%) (Table 4 1)

**PLATE 4.1: Different flowering types**



**Single**

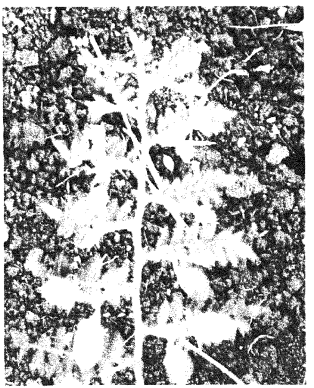
**Double**



**Triple**

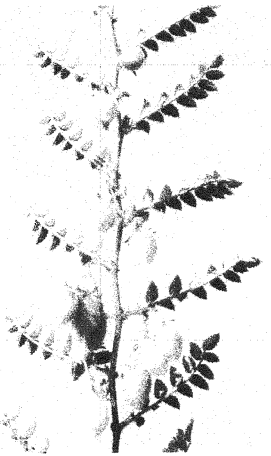
**PLATE 4.2: Expression of different flowering nodes in the multi-flowered line JGM 7**

**PLATE 4.3: Podding behaviour in parents**



ICC 4929

IPC 99-18



JGM 7

**4.2.2.3. Multi-pods**

JGM 7 multi-flowered parent recorded mean expressivity of 24.70% for multi-pod trait. The range was between 0.00% to 66.70%. The average number of pods per node was 2.16 and ranged between 0.88 to 4.17. In this parent, 37.27% of flowering nodes did not set pods (Table 4.1).

Table 4.1: Expressivity of flowers and pods in different parental lines

| CHARACTER | EXPRESSION % (Range) |
|---|---|
| **ICC 4929** | |
| Expression of double-flowers | **96.34% (60-100%)** |
| Expression of double-pods | **76.36% (20-90%)** |
| Flowering nodes without pods | **6.83% (0-40%)** |
| Number of flowers per node | **1.96 (1.7-2.0)** |
| Number of pods per node | **1.77 (1.7-1.9)** |
| **IPC 99-18** | |
| Expression of triple-flower | **81.15% (20-100%)** |
| Expression of triple-pod | **0.00%** |
| Expression of double-pod | **55.3% (0.00-77.80%)** |
| Expression of single-pod | **44.70% (10-100%)** |
| Flowering nodes without pods | **26.34% (0-50%)** |
| Number of flowers per node | **2.8 (1.9-3.0)** |
| Number of pods per node | **1.44 (0.7-1.9)** |
| **JGM 7** | |
| Expression of multi-flower | **51.33% (0-80%)** |
| Expression of multi-pod | **24.7% (0.00-66.7%)** |
| Flowering nodes without pods | **37.27% (10-50%)** |
| Number of flowers per node | **2.83 (1.22-4.10)** |
| Number of pods per node | **2.16 (0.88-4.17)** |

Table 4.2: Expression level of different flowering and podding nodes in multi-flowered parent **JGM 7:**

| NUMBER OF FLOWERS PER NODE | **EXPRESSION % (Range)** |
|---|---|
| One flower | 31.31% (0.00-55.60%) |
| Two flowers | 17.37% (0.00-42.90%) |
| Three flowers | 23.50% (10.00-44.40%) |
| Four flowers | 9.90% (0.00-25.00%) |
| Five flowers | 11.40% (0.00-40.00%) |
| Six flowers | 4.03% (0.00-20.00%) |
| Seven flowers | 2.50% (0.00-20.00%) |
| NUMBER OF PODS PER NODE | |
| One pod | 42.16% (16.70-71.40%) |
| Two pods | 32.95 % (0.00-83.30 %) |
| Three pods | 17.55% (0.00-66.70%) |
| Four pods | 7.14% (0.00-40.00%) |

## 4.3 Allelic relationships of genes controlling different flowering types

### 4.3.1. Inheritance of number of flowers per node in the cross ICC 4929 x IPC 99-18:

All $F_1$s (direct and reciprocal) from the cross between double-flowered (ICC 4929) and the triple-flowered parents (IPC 99-18) were double-flowered (Table 4 3) indicating that double-flower trait is dominant over triple-flower trait There were 254 double-flowered and 78 triple-flowered plants in $F_2$ These gave a good fit to the monogenic 3 1 ratio ($\chi 2$=0 401, P=0 53) (Table 4 4) The possible genotypes for parents and $F_2$ phenotypic classes are suggested in Table 4 5 The inheritance pattern is explained in Fig 4 1

### 4.3.2. Inheritance of number of flowers per node in the cross IPC 99-18 x JGM 7

When triple-flowered line (IPC 99-18) was crossed with multi-flowered line (JGM 7), all $F_1$s were found to be single-flowered (Table 4 3) In the $F_2$, 199 single-flowered, 74 triple-flowered, and 87 multi-flowered plants were observed which gave a good fit to the expected 9 3 4 ratio ($\chi 2$=0 133, P=0 71) (Table 4 4), confirming the two-factor inheritance for number of flowers per axis It was not possible to distinguish the double recessive class (triple-multiflowered) from multi-flowered, so these two classes were combined This led to an expected ratio of 9 3 4 in place of 9 3 3 1 The suggested genotypes of parents and different phenotypic classes of $F_2$ are given in Table 4 5 The inheritance pattern is explained in Fig 4 2

### 4.3.3. Inheritance of number of flowers per node in the cross ICC 4929 x JGM 7

When double-flowered line (ICC 4929) was crossed with multi-flowered line (JGM 7), all $F_1$s were found to be single-flowered (Table 4 3) In the $F_2$, 195 single-flowered, 66

double-flowered, 72 multi-flowered, and 27 double-multiflowered plants were observed which gave a good fit to the expected 9:3:3:1 ratio ($\chi 2$=1.51; P=0.68) (Table 4.4), confirming the two-factor inheritance for number of flowers per axis. The suggested genotypes of parents and different phenotypic classes of $F_2$ are given in Table 4.5. The inheritance pattern is explained in Fig. 4.3.

The results clearly established that two loci control number of flowers per axis in these genotypes. The double-flowered and triple-flowered are controlled by a single locus (*Sfl*) and the allele for double-flowered trait ($sfl_d$) is dominant over the allele for triple-flowered trait ($sfl_t$). The multi-flowered trait in JGM 7 is controlled by a different gene (*cym*) and the single-flowered plants have dominant allele at the both loci (*Sfl_ Cym* ).

Table 4.3: Flowering types of $F_1$s in crosses between different flowering types:

| ♀ / ♂ | ICC 4929 (Double-flowered) | IPC 99-18 (Triple-flowered) | JGM 7 (Multi-flowered) |
|---|---|---|---|
| ICC 4929 (Double-flowered) | | Double-flowered | Single-flowered |
| IPC 99-18 (Triple-flowered) | Double-flowered | | Single-flowered |
| JGM 7 (Multi-flowered) | Single-flowered | Single-flowered | |

Table 4.4: Goodness-of-fit $\chi 2$ test for observed frequencies of different flowering types in $F_2$ of three crosses.

| Cross | Flowering type | Observed | Expected | Expected genetic ratio | $\chi 2$ | P |
|---|---|---|---|---|---|---|
| ICC 4929 x IPC 99-18 | Double-flowered (DF) | 254 | 249 | 3 | 0.401 | 0.53 |
| | Triple-flowered (TF) | 78 | 83 | 1 | | |
| IPC 99-18 x JGM 7 | Single-flowered (SF) | 199 | 202.5 | 9 | 0.133 | 0.71 |
| | Triple-flowered (TF) | 74 | 67.5 | 3 | | |
| | Multi-flowered (MF) & Triple-multi-flowered (TM) | 87 | 90 | 4 | | |
| ICC 4929 x JGM7 | Single-flowered (SF) | 195 | 202.5 | 9 | 1.510 | 0.68 |
| | Double-flowered (DF) | 66 | 67.5 | 3 | | |
| | Multi-flowered (MF | 72 | 67.5 | 3 | | |
| | Double-multi-flowered (DF) | 27 | 22.5 | 1 | | |

Table 4.5: Suggested possible genotypes for different flowering types.

| Flowering type | Possible genotype |
|---|---|
| Single-flowered (SF) | *Sfl _ Cym _* |
| Double-flowered (DF) | *$sfl_d$ $sfl_d$ Cym_ or $sfl_d$ $sfl_t$ Cym _* |
| Triple-flowered (TF) | *$sfl_t$ $sfl_t$ Cym_* |
| Multi-flowered (MF) | *Sfl _ cym cym* |
| Double-multiflowered (DM) | *$sfl_d$ $sfl_d$ cym cym or $sfl_d$ $sfl_t$ cym cym* |
| Triple-multiflowered (TM) | *$sfl_t$ $sfl_t$ cym cym* |

**Fig. 4.1:** Schematic representation of the inheritance of number of flowers per axis in the cross **ICC 4929** (DF) x **IPC 99-18** (TF) :

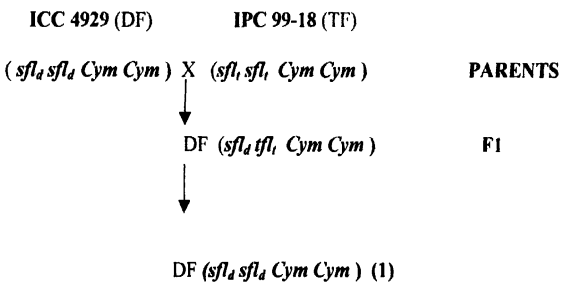
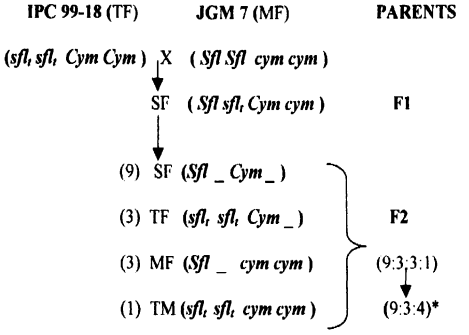
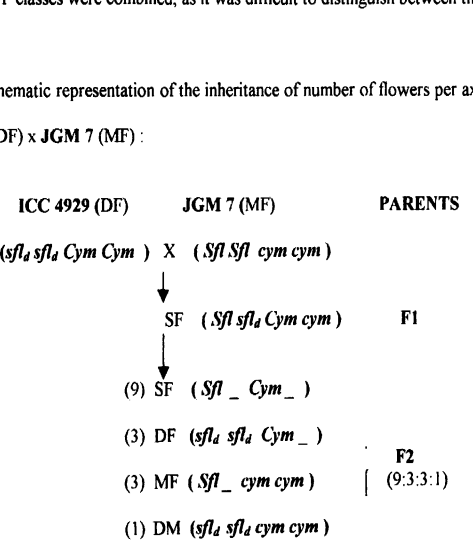


**Fig. 4.2**: Schematic representation of the inheritance of number of flowers per axis in the cross **IPC 99-18** (TF) x **JGM 7** (MF) :




[* MF and TF classes were combined, as it was difficult to distinguish between these classes]

**Fig. 4.3**: Schematic representation of the inheritance of number of flowers per axis in the cross **ICC 4929** (DF) x **JGM 7** (MF) :



## 4.4. Inheritance of flower colour

Inheritance of flower colour (pink vs. pink-veined white flower) was studied in two crosses that involved ICC 4929 (pink-veined white flower) as one of the parents, i.e. ICC 4929 x IPC 99-18 and ICC 4929 x JGM 7.

### 4.4.1. Cross ICC 4929 x IPC 99-18

The cross between ICC 4929 (pink-veined white flower) and IPC 99-18 (pink flower) produced pink flowered $F_1$s and the observed ratio of 260 pink: 72 pink-veined white flowered plants in $F_2$ that corresponded to the expected 3:1 ratio ($\chi 2 = 1.94$; $P = 0.16$) (Table 4.6). The results suggested that a single gene control difference in the flower colour between the parents of this cross and pink flower colour is dominant over pink-veined white flower colour.

### 4.4.2. Cross ICC 4929 x JGM 7

The cross between ICC 4929 (pink-veined white flower) and JGM 7 (pink flower) produced pink flowered $F_1$s. The $F_2$ data (n =360 plants) fitted well to the ratio of 3 pink : 1 white flowered plants ($\chi 2 = 3.79$ : $P = 0.05$). The results were similar to those from the cross ICC 4929 x IPC 99-18.

Table 4.6: Segregation for **Flower colour** in the $F_2$ of two crosses of chickpea

| Cross | Parent/ generation | Flower colour | Expected ratio | No. of plants | | $\chi 2$ | P |
|---|---|---|---|---|---|---|---|
| | | | | Observed | Expected | | |
| ICC 4929 x IPC 99-18 | ICC 4929 | Pink-veined white | | | | | |
| | IPC 99-18 | Pink | | | | | |
| | $F_1$ | Pink | | | | | |
| | $F_2$ | Pink | 3 | 260 | 270 | 1.94 | 0.163 |
| | | Pink-veined white | 1 | 72 | 90 | | |
| ICC 4929 x JGM 7 | ICC 4929 | Pink-veined white | | | | | |
| | JGM 7 | Pink | | | | | |
| | $F_1$ | Pink | | | | | |
| | $F_2$ | Pink | 3 | 286 | 270 | 3.79 | 0.051 |
| | | Pink-veined white | 1 | 74 | 90 | | |

## 4.5. Inheritance of foliage colour

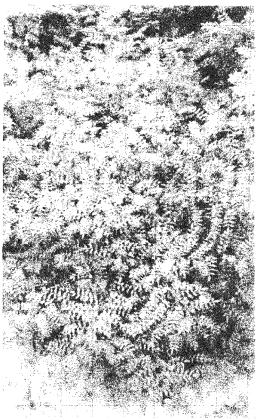
Inheritance of foliage colour (normal green, light green and purple) was studied (Plate No 4.4). The $F_2$ population (n=360 plants) of a cross between IPC 99-18 (light green) and JGM 7 (purple) segregated for light green and purple-foliage plants in a ratio of 3:1 confirming the monogenic inheritance of the character ($\chi 2 = 1.48$; $P = 0.22$) (Table 4.7).

The $F_2$ population (n=360 plants) of a cross between ICC 4929 (normal green) and JGM 7 (purple) also showed monogenic inheritance of foliage colour where normal green foliage was dominant to purple ($\chi 2 = 3.33$; $P = 0.07$) (Table 4.7).

**PLATE 4.4: Parents showing variation in foliage pigmentation**

**ICC 4929 (Normal green)**

**IPC 99-18 (Light green)**



**JGM 7 (Purple)**

Table 4.7: Segregation for **Foliage colour** in the $F_2$ of three crosses of chickpea

| Cross | Parent/ generation | Foliage colour | Expected ratio | No. of plants | | $\chi^2$ | P |
|---|---|---|---|---|---|---|---|
| | | | | Observed | Expected | | |
| IPC 99-18 x JGM 7 | IPC 99-18 | Light green | | | | | |
| | JGM 7 | Purple | | | | | |
| | $F_1$ | Light green | | | | | |
| | $F_2$ | Light green | 3 | 260 | 270 | 1.48 | 0.22 |
| | | Purple | 1 | 100 | 90 | | |
| ICC 4929 x JGM 7 | ICC 4929 | Normal green | | | | | |
| | JGM 7 | Purple | | | | | |
| | $F_1$ | Normal green | | | | | |
| | $F_2$ | Normal green | 3 | 255 | 270 | 3.33 | 0.07 |
| | | Purple | 1 | 105 | 90 | | |

**4.6. Genetic linkages:**

Linkage relationships of loci controlling number of flowers per axis, flower colour and foliage colour were studied in $F_2$ populations of three crosses, ICC 4929 x IPC 99-18, IPC 99-18 x JGM 7, and ICC 4929 x JGM 7. Loci controlling foliage colour (normal green vs. light green) was found linked with flower colour (pink vs. pink-veined white), and flowering type (DF vs. TF) in the cross ICC 4929 (DF) x IPC 99-18 (TF) with recombination frequencies 0.386 (P= <0.01) and 0.447 (P= 0.03), respectively (Table 4 8).

Table 4 8: Joint segregation of characters in different $F_2$ populations

| Cross | | Segregation ratio | | | | | | | | $\chi^2$ | P |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | *X1Y1* | *X1Y2* | *X2Y1* | *X2Y2* | *X3Y1* | *X3Y2* | *X4Y1* | *X4Y2* | | |
| Flowering type and flower colour | | | | | | | | | | | |
| ICC 4929 x JGM 7 | Appr. ratio | 27 | 9 | 9 | 3 | 9 | 3 | 3 | 1 | | |
| | Obse. | 153 | 42 | 53 | 13 | 58 | 14 | 23 | 4 | 6.71 | 0.46 |
| | Exp. | 151.9 | 50.62 | 50.62 | 16.87 | 50.62 | 16.87 | 16.87 | 5.62 | | |
| Flowering type and foliage colour | | | | | | | | | | | |
| ICC 4929 x JGM 7 | Appr ratio | 27 | 9 | 9 | 3 | 9 | 3 | 3 | 1 | | |
| | Obse | 142 | 53 | 50 | 16 | 47 | 25 | 17 | 10 | 8.35 | 0.31 |
| | Exp. | 151.8 | 50.62 | 50.62 | 16.87 | 50.62 | 16.87 | 16.87 | 5.62 | | |
| Flower and foliage colours | | | | | | | | | | | |
| ICC 4929 x JGM 7 | Appr ratio | 9 | 3 | 3 | 1 | | | | | | |
| | Obsc. | 184 | 104 | 71 | Nil | | | | | | |
| | Exp. | 202.5 | 67.5 | 67.5 | 22.5 | | | | | | |
| Flowering type and foliage colour | | | | | | | | | | | |
| IPC 99-18 x JGM7 | Appr ratio | 27 | 9 | 9 | 3 | 9+3 | 3+1 | | | | |
| | Obsc. | 140 | 59 | 58 | 16 | 62 | 25 | | | 4.16 | 0.76 |
| | Exp. | 151.9 | 50.62 | 50.62 | 16.87 | 50.62 + 16.9 | 16.87 + 5.62 | | | | |
| Flowering type and flower colour | | | | | | | | | | | |
| ICC 4929 x IPC 99-18 | Appr ratio | 9 | 3 | 3 | 1 | | | | | | |
| | Obse. | 199 | 55 | 61 | 17 | | | | | 2.33 | 0.50 |
| | Exp. | 186.7 | 62.25 | 62.25 | 20.75 | | | | | | |

| Cross | | *X1Y1* | *X1Y2* | *X2Y1* | *X2Y2* | $\chi^2$ | P | r ± SE |
|---|---|---|---|---|---|---|---|---|
| Flowering type and foliage colour | | | | | | | | |
| ICC 4929 x IPC 99-18 | Appr ratio | 9 | 3 | 3 | 1 | | | |
| | Obse | 203 | 68 | 51 | 10 | 9.55 | 0.03* | 0.447±0.043 |
| | Exp. | 186.7 | 62.25 | 62.25 | 20.75 | | | |
| Flower and foliage colours | | | | | | | | |
| ICC 4929 x IPC 99-18 | Appr ratio | 9 | 3 | 3 | 1 | | | |
| | Obse | 220 | 52 | 41 | 20 | 14.88 | <0.01* | 0.386±0.046 |
| | Exp. | 186.7 | 62.25 | 62.25 | 20.75 | | | |

**NOTE:** *X1, X2, X3* and *X4* representing different flowering types (SF, DF or TF, MF and DM) and two flower colours (pink and pink-veined white). *Y1* and *Y2* representing the foliage colours (normal green and light green or normal green and dark green or light and dark green) in some combinations and flower colours in others.

Joint segregation analysis indicated that genes for flower colour and number of flowers per axis are independent from each other. The $F_2$ of the cross ICC 4929 x JGM 7 segregated for flower colour (pink vs. pink-veined white) and foliage colour (normal green vs. purple). No segregant was observed in this cross that had purple foliage with pink-veined white flower, indicating that either the two genes responsible for these characters are closely linked or one gene has pleiotropic effects on both the characters (Table 4.8).

**4.7. Genetic variability among flowering types for various characters**

### 4.7.1. Number of flowers per axis (at 10 flowering nodes)

The mean values were calculated and ranges were recorded for number of flowers per axis (average of 10 flowering nodes) for each phenotypic class (flowering type) in $F_2$ population of each cross. **SF** phenotypic class was observed in the $F_2$ of crosses (ICC 4929 x JGM and IPC 99-18 x JGM 7). None of the **SF** plants showed more than one flower at any of the flowering node. **DF** types exhibited mean values of 1.80 (range=1.10 to 2.00) and 1.94 (range=1.40 to 2.00) flowers per flowering node in crosses ICC 4929 x JGM 7 and ICC 4929 x IPC 99-18, respectively. Mean values of 1.98 (range=1.30 to 2.60) and 2.20 (ranged from 1.40 to 2.70) flowers/node were observed for **TF** types in $F_2$s of IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, respectively. **MF** types recorded mean values of 1.38 (range=1.00 to 2.70) and 1.30 (range=0.80 to 2.80) flowers per node in $F_2$ of IPC 99-18 x JGM 7 and ICC 4929 x JGM 7 crosses, respectively. The **DM** types showed a mean value 2.05 and the range from 1.40 to 3.40 flowers/node. Results indicated that only TF and DM types showed mean values of more than two flowers per axis (Tables 4.9 to 4.13).

### 4.7.2. Number of pods per axis (at 10 flowering nodes)

Number of pods per axis in **SF** plant showed means of 0.94 and 0.86, and ranges from 0.40 to 1.00 and from 0.30 to 1.00 in the crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM 7, respectively. Mean values of 1.40 (range=0.80 to 2.00) and 1.26 (range=0.50 to 2.00) for **DF** types were observed in crosses ICC 4929 x JGM 7 and ICC 4929 x IPC 99-18, respectively. In **TF** types, means were found to be 1.57 with a range of 0.80 to 2.50, and 1.25 with a range of 0.40 to 2.00 in two crosses IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, respectively. While means of 0.95 (range=0.40 to1.80) and 0.80 (range=0.30 to 1.00) were found for **MF**

types in crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM, respectively. In the cross ICC 4929 x JGM 7, **DM** types showed a mean value of 1.10 with a maximum range between 0.70 and 2.30. Results showed that TF and DF types produced more number of pods per axis than SF, MF and DM types (Tables 4.9 to 4.13).

**4.7.3. Number of pods per plant**

The average number of pods per plant was 159 (range=5 to 553) and 200 (range=8 to 778) in **SF** types, and 149 (range=25 to 375) and 192 (range=3 to 444) in **MF** types in the crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM 7, respectively. **TF** types had a mean value of 144 (range=22 to 480), and 158 (range=25 to 410) in the crosses IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, respectively. **DF** types showed mean values of 188 (range=1 to 546) and 176 (range=10 to 557) in crosses ICC 4929 x JGM and ICC 4929 x IPC 99-18, respectively. Among all flowering types, **DM** types exhibited, more number of pods per plant with a mean value 207 (range=18 to 365). However, there was no significant difference for this character among different flowering types in any of the crosses (Tables 4.9 to 4.13).

**4.7.4. Number of seeds per plant**

The number of seeds per plant had mean values of 208 (with a range of 6 to 808) and 249 (with a range of 9 to 945) in **SF** type and 199 (with a range of 26 to 560) and 236 (with a range of 4 to 558) in **MF** types in crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM 7, respectively. A mean of 188 with a range of 26 to 555 and a mean of 199 with a range of 26 to 496 were found in **TF** type in crosses IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, respectively. The **DF** types gave mean values of 224 (range= 11 to 565) and 209 (range= 16 to

636) in crosses ICC 4929 x JGM 7 and ICC 4929 x IPC 99-18, respectively A mean value of 258 with a range of 24 to 411 was found in **DM** in the cross ICC 4929 x JGM 7 No significant difference was observed among different flowering types for this character in three crosses, but DM type produced more number of seeds than other flowering types (Tables 4 9 to 4 13)

**4.7.5. Yield per plant**

Seed yield per plant was found to have mean values of 28 64g (range=0 40 to 110g) and 32 41g (range=0 60 to 131g) in **SF** plants, and mean values of 28 80g (range =3 70 to 80g) and 33 30g (range=0 50 to 77 5g) in **MF** types in crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM 7, respectively Mean values of 27 00g (with a range of 2 to 64 50) and 28 10g (with a range of 0 40 to 81 20) in **DF** types were recorded in crosses ICC 4929 x JGM 7 and ICC 4929 x IPC 99-18, respectively The mean value of 34 00g (range=1 70 to 62 40g) was observed in **DM** type in cross ICC 4929 x JGM 7 **TF** types showed mean values of 24 7g (range=2 to 64 50g) and 27 6g (range=2 30 to 70 00g) in the crosses IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, respectively No significant difference was found among different flowering types for seed yield per plant (Tables 4 9 to 4 13)

**4.7.6. 100 seed weight**

In **SF** type, 100-seed weight showed mean values of 13 61g (with a range of 6 67 to 20 30g) and 12 91g (with a range of 7 28 to 22 00g), while it showed means of 14 45g (range= 7 00 to 28 70g) and 14 30g (10 00-19 00g) in **MF** type in crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM 7, respectively In **DF** type, mean values of 11 90g (range= 7 30 to 14 70g) and

13 32 (range=6 25 to19 00g) were obtained in crosses ICC 4929 x JGM 7 and ICC 4929 x IPC 99-18, respectively Mean values of 13 10g (range=7 40 to 18 30g) and 13 77g (range=8 20 to 17 30g) were found in **TF** types in crosses IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, respectively The **DM** types recorded a mean of 13 00g (range=7 10-16 10g) in the cross ICC 4929 x JGM 7 Most of the comparisions with the MF and DM types recorded significant differences and it is clear that MF types produced more seed size than other flowering types (Tables 4 9 to 4 13)

**4.7.7. Number of seeds per pod**

The number of seeds per pod in **SF** plants ranged from 1 to 3 05 and 1 to 2 86 with mean values of 1 32 and 1 27 and in **MF** plants it ranged from 1 00 to 1 70 and 1 00 to 1 90 with mean values of 1 33 and 1 27 in two crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM 7, respectively In **DF** type, it ranged from 1 00 to 1 86 and 1 00 to 2 90 with mean values of 1 22 and 1 21 in crosses ICC 4929 x JGM 7 and ICC 4929 x IPC 99-18, respectively It varied from 1 00 to 2 12 and 1 00 to 2 90 with mean values of 1 32 and 1 27 in **TF** type in crosses IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, respectively **DM** types showed range from 1 00 to 1 95 with a mean value of 1 27 in the cross ICC 4929 x JGM 7 No significant differences were observed among different flowering types for this trait (Tables 4 9 to 4 13)

**4.7.8. Number of empty pods per plant**

This character recorded a range of 0 to 103 with a mean of 8 17 and a range of 0 to 55 with a mean of 8 0 in **SF** type, and a range of 0 to 50 with a mean of 6 53 and a range of 0 to 23 with a mean of 6 18 in **MF** type in crosses IPC 99-18 x JGM 7 and ICC 4929 x JGM 7,

respectively. A range of 0 to 40 with a mean of 7.90 and a range of 0 to 35 with a mean of 9.34 was recorded in **DF** plants of crosses ICC 4929 x JGM 7 and ICC 4929 x IPC 99-18, respectively. In crosses IPC 99-18 x JGM 7 and ICC 4929 x IPC 99-18, **TF** type exhibited ranges of 0 to 60 and 0 to 54 with mean values 7.80 and 10.30 respectively. In the cross ICC 4929 x JGM 7, **DM** types showed a range from 0 to 64 with a mean value of 7.63. No significant differences were found among flowering types for number of empty pods per plant, but the extreme values of ranges were low in MF type than other flowering types (Tables 4.9 to 4.13).

These results suggest that the different flowering types (SF, DF, TF, MF and DM) had no significant effects on number of pods per plant, number of seeds per plant, number of seeds per pod and number of empty pods. The significant differences observed for number of flowers per axis, number of pods per axis, 100-seed weight and yield per plant.

Table 4.9: Comparision of different characters among Single, Triple and Multi-flowered plants in $F_2$ population of the cross **IPC 99-18(TF) x JGM 7(MF):**

| VARIABLE | | SF | TF | MF |
|---|---|---|---|---|
| Number of flowers per axis (average of 10 flowering nodes) | MEAN±SE | 0.99±0.03 | 1.98±0.36 | 1.38±0.52 |
| | RANGE | 0.80-1.00 | 1.30-2.60 | 1.00-2.70 |
| Number of pods per axis (average of 10 flowering nodes) | MEAN±SE | 0.94±0.07 | 1.57±0.42 | 0.95±0.34 |
| | RANGE | 0.40-1.00 | 0.80-2.50 | 0.40-1.80 |
| Number of pods per plant | MEAN±SE | 159.10±6.92 | 143.50±9.78 | 148.80±9.16 |
| | RANGE | 5-553 | 22-480 | 25-375 |
| Number of seeds per plant | MEAN±SE | 207.90±8.94 | 187.70±12.43 | 199.10±13.10 |
| | RANGE | 6-808 | 26-555 | 26-560 |
| Yield per plant | MEAN±SE | 28.64±1.29 | 24.75±1.73 | 28.8±1.88 |
| | RANGE | 0.40-110 | 2-64.5 | 3.70-79.70 |
| 100 seed weight | MEAN±SE | 13.61±0.16 | 13.10±0.27 | 14.45±0.29 |
| | RANGE | 6.67-20.30 | 7.40-18.30 | 7-28.70 |
| Number of seeds per pod | MEAN±SE | 1.32±0.02 | 1.32±0.03 | 1.330±0.21 |
| | RANGE | 1-3.05 | 1-2.12 | 1-1.75 |
| Number of empty pods per plant | MEAN±SE | 8.17±0.72 | 7.79±0.99 | 6.53±0.82 |
| | RANGE | 0-103 | 0-60 | 0-50 |

Table 4.10: Pair wise comparisions (test of significance) among different flowering types in $F_2$ of IPC **99-18(TF) x JGM 7(MF)** :

| VARIABLE | (SF X TF) | | (SF X MF) | | (TF X MF) | |
|---|---|---|---|---|---|---|
| | t-value | Probability value | t-value | Probability value | t-value | Probability value |
| Number of flowers per axis (at 10 flowering nodes) | -27.02 | <0.001** | 7.47 | <0.001** | -9.4 | <0.001** |
| Number of pods per axis (at 10 flowering nodes) | -15.08 | <0.001** | 0.29 | 0.773 | -11.85 | <0.001** |
| Number of pods per plant | 1.22 | 0.225 | -0.85 | 0.395 | 0.40 | 0.691 |
| Number of seeds per plant | 1.23 | 0.22 | -0.58 | 0.579 | 0.62 | 0.536 |
| Yield per plant | 1.65 | 0.1 | 0.07 | 0.943 | 1.58 | 0.121 |
| 100 seed weight | 1.6 | 0.11 | 2.67 | 0.008* | 3.36 | <0.001** |
| Number of seeds per pod | 0.01 | 0.99 | 0.23 | 0.819 | 0.2 | 0.84 |
| Number of empty pods per plant | 0.29 | 0.77 | -1.5 | 0.135 | -0.98 | 0.33 |

**SF** = Single flowered type

**TF** = Triple flowered type

**MF** = Multiflowered type

Table 4.11: Comparision of characters under study among Single, Double, Multi and Double-multi-flowered plants in $F_2$ population of the cross **ICC 4929(DF) x JGM 7(MF)**

| VARIABLE | | SF | DF | MF | DM |
|---|---|---|---|---|---|
| Number of flowers per axis (average of 10 flowering nodes) | MEAN±SE | 0.984±0.039 | 1.8±0.337 | 1.3±0.414 | 2.05±0.93 |
| | RANGE | 7-10 | 11-20 | 8-28 | 14-34 |
| Number of pods per axis (average of 10 flowering nodes) | MEAN±SE | 0.863±0.14 | 1.4±0.46 | 0.8±0.372 | 1.1±0.707 |
| | RANGE | 3-10 | 8-20 | 3-10 | 7-23 |
| Number of pods per plant | MEAN±SE | 199.7±8.83 | 187.9±14.27 | 192.3±10.96 | 206.6±16.76 |
| | RANGE | 8-778 | 10-546 | 3-444 | 18-365 |
| Number of seeds per plant | MEAN±SE | 249.3±10.78 | 223.8±15.92 | 236.3±13.73 | 257.9±20.24 |
| | RANGE | 9-945 | 11-565 | 4-558 | 24-411 |
| Yield per plant | MEAN±SE | 32.41±1.47 | 27.03±2.053 | 33.3±2.103 | 33.99±2.84 |
| | RANGE | 0.6-131 | 3-72.3 | 0.5-77.5 | 1.7-62.4 |
| 100 seed weight | MEAN±SE | 12.91±0.436 | 11.89±0.213 | 14.29±0.34 | 12.99±0.173 |
| | RANGE | 7.28-22 | 7.3-14.7 | 10-18.98 | 7.1-16.1 |
| Number of seeds per pod | MEAN±SE | 1.274±0.028 | 1.221±0.025 | 1.231±0.0236 | 1.271±0.05 |
| | RANGE | 1-2.86 | 1-1.86 | 1-1.72 | 1.02-1.95 |
| Number of empty pods per plant | MEAN±SE | 8.016±0.73 | 7.903±1.105 | 6.176±0.804 | 7.63±2.43 |
| | RANGE | 0-55 | 0-40 | 0-23 | 0-64 |

**SF** = Single flowered type

**DF** = Double flowered type

**MF** = Multiflowered type

**DM** = Double multiflowered type

Table 4.12: Pair wise comparisions (test of significance) among different plant types in $F_2$ population of a cross **ICC 4929 (DF) x JGM 7(MF)**

| Variable | | (SF X DF) | (SF X MF) | (SF X DM) | (DF X MF) | (DF X DM) | (MF X DM) |
|---|---|---|---|---|---|---|---|
| Number of flowers per axis (at 10 flowering nodes) | t-value | 23.98 | 7.45 | 11.42 | 9.41 | 2.53 | 7.39 |
| | Prob. | <0.001** | <0.001** | <0.001** | <0.001** | 0.017* | <0.001** |
| Number of pods per axis (at 10 flowering nodes) | t-value | 11.06 | -1.81 | 3.13 | 10.30 | -3.66 | 4.03 |
| | Prob. | <0.001** | 0.070 | 0.004* | <0.001** | <0.001** | <0.001** |
| Number of pods per plant | t-value | -0.68 | -0.53 | 0.29 | -0.25 | 0.77 | 0.70 |
| | Prob. | 0.500 | 0.600 | 0.770 | 0.800 | 0.442 | 0.480 |
| Number of seeds per plant | t-value | -1.23 | -0.75 | 0.37 | -0.6 | 1.24 | 0.86 |
| | Prob. | 0.220 | 0.450 | 0.700 | 0.550 | 0.220 | 0.400 |
| Yield per plant | t-value | -1.92 | 0.32 | 0.4 | -2.13 | 1.92 | 0.18 |
| | Prob. | 0.050 | 0.750 | 0.700 | 0.030* | 0.060 | 0.850 |
| 100 seed weight | t-value | -3.72 | 3.92 | 0.17 | -5.97 | 2.55 | -2.14 |
| | Prob. | <0.001** | <0.001** | 0.870 | <0.001** | 0.010* | 0.030* |
| Number of seeds per pod | t-value | -1.41 | -1.19 | -0.06 | 0.28 | 1.01 | 0.85 |
| | Prob. | 0.160 | 0.240 | 0.950 | 0.780 | 0.320 | 0.40 |
| Number of empty pods per plant | t-value | -0.08 | -1.69 | -0.18 | 1.26 | -0.10 | 0.57 |
| | Prob. | 0.930 | 0.090 | 0.860 | 0.210 | 0.920 | 0.570 |

Table 4.13: Comparison of characters under study between double and triple-flowered plants in $F_2$ population of cross **ICC 4929 (DF) X IPC 99-18 (TF):**

| VARIABLE | MEAN±SE | | RANGE | | DF x TF | |
|---|---|---|---|---|---|---|
| | DF | TF | DF | TF | t-value | Prob |
| Number of flowers per axis (average of 10 flowering nodes) | 1.94±0.085 | 2.2±0.270 | 14-20 | 14-27 | -9.36 | 0.001** |
| Number of pods per axis (average of 10 flowering nodes) | 1.26±0.230 | 1.25±0.430 | 5-20 | 4-20 | 0.11 | 0.91 |
| Number of pods per plant | 175.9±7.080 | 158±9.590 | 10-575 | 25-410 | 1.5 | 0.136 |
| Number of seeds per plant | 208.9±8.140 | 198.8±12.02 | 16-636 | 26-496 | 0.69 | 0.49 |
| Yield per plant | 28.14±1.110 | 27.58±1.745 | 0.4-81.2 | 2.3-70 | 0.25 | 0.80 |
| 100 seed weight | 13.32±0.126 | 13.77±0.250 | 6.25-19 | 8.2-17.3 | -1.75 | 0.09 |
| Number of seeds per pod | 1.21±0.014 | 1.274±0.035 | 1-2.3 | 1-2.89 | -1.74 | 0.08 |
| Number of empty pods per plant | 9.34±0.475 | 10.32±1.130 | 0-35 | 0-54 | -0.8 | 0.42 |

**DF** = Double flowered type
**TF** = Triple flowered type

# Chapter V
# Discussion

CHAPTER V

# DICUSSION

The investigation was aimed at studying inheritance, penetrance and expressivity of factor(s) controlling number of flowers per axis in chickpea (*Cicer arietinum* L.). A double-flowered line (ICC 4929), a triple-flowered line (IPC 99-18) and a multi-flowered line (JGM 7) were crossed in all possible combinations. Data were recorded on parental lines (ICC 4929, IPC 99-18 and JGM 7), $F_1$s and $F_2$s. Reciprocal crosses were excluded in $F_2$, as $F_1$s from direct and reciprocal crosses did not reveal maternal effects for any of the characters under study. Thus, three crosses (ICC 4929 x IPC 99-18, IPC 99-18 x JGM 7 and ICC 4929 x JGM 7) were studied in $F_2$. Efforts were also made to assess the effect of the trait controlling number of flowers per axis on grain yield and its components. The results of the present investigation are discussed under the following headings:

5.1. Penetrance

5.2. Expressivity

5.3. Inheritance of factors controlling number of flowers per axis

5.4. Inheritance of flower colour

5.5. Inheritance of foliage colour

5.6. Genetic linkages

5.7. Genetic variability among flowering types

**5.1. Penetrance**

Apart from the genes for quantitative traits, some genes controlling qualitative traits are highly influenced by the environment and cause variable phenotypic effects. The phenotypic effects produced by such genes may not be noticed in some individuals of a given population. Level of penetrance can be calculated as the proportion of individuals with a given genotype, which exhibit a particular phenotype.

The parental genotypes ICC 4929, IPC 99-18 and JGM 7 exhibited 100% penetrance for double-flower (DF), triple-flower (TF), and multi-flower (MF) traits, respectively. Kumar et al. (2000a) found unstable penetrance of 53% [$\chi 2$=8.71; 0.01>P>0.0001] in $F_2$ and 84.5% [$\chi 2$=1.52; 0.25>P>0.20] in RILs for double-podded gene (*ss*). We considered only flowering data (number of flowers per axis) of the fixed lines (parental lines) for determining penetrance of genes. This is because the conversion of flowers to pods is highly affected by the external environment (temperature, soil moisture and sunlight, etc.). We preferred to use fixed lines, as we are sure about the genotype of the plants. We believe that the penetrance of double-flower trait would have been higher in the study of Kumar et al. (2000a) if it was calculated from the flowering data on parental lines. The crop was provided one light irrigation during flower initiation, to avoid moisture stress and to get maximum expression of the trait.

**5.2. Expressivity**

The double-flowered line ICC 4929 showed mean expressivity of 96.34% (range between 60% to 100%) for the double-flower trait, the triple-flowered line IPC 99-18 showed mean expressivity of 81.15% (range between 20% to 100%) for triple-flower trait, and the

multi-flowered line JGM 7 showed mean expressivity of 51.33% (range between 0% to 80%) for the multi-flower trait. Thus, the highest expression was found for the double-flower trait. This variation in expressivity of different flowering types could possibly be due to the positive influence of environment on double-flower gene than on triple and multi-flower genes, or less adaptive value of triple and multi-flower genes as they were evolved recently.

On an average, 79.26% of the flowering nodes produced pods in the double-podded line ICC 4929 and the double-pod trait showed mean expressivity of 76.36% with the range between 20% and 90%. Kumar et al (2000a) observed variable expression of double-pod trait in $F_2$ (1.1-14.8%) and in RILs (0.1-33%). For the same trait he observed higher expression in late sown (17.1-68.7%) than in early sown double-podded parent. The difference in the expressivity for double-pod trait between the study of Kumar et al (2000a) and this study would be due to differences in the genetic background of lines used. The triple-flower trait showed 0.00% expressivity in triple-flowered parent IPC 99-18. Thus, one flower at each flowering node always failed to set pod. It was distinctly different in appearance than other two flowers (Plate 4.1). So, the triple-flowered line IPC 99-18 did not have any triple-podded node. It only had double-podding nodes (55.3%) and single-podding nodes (44.7%). In multi-flowered parent, 48.15% of multi-flowering nodes converted into podding nodes. The multi-pod trait showed mean expressivity of 24.7% with range from 0.00% to 66.7% in multi-flowered parent JGM 7. The expressivity of this trait was very low as compared to other flowering types.

The penetrance and expressivity of the gene(s) controlling number of flowers per axis can be increased and stabilized by placing it in appropriate genetic backgrounds and then this character can be efficiently tapped to get higher yield in scarce soil moisture conditions under which chickpea is traditionally grown in India and elsewhere.

The difference between expressivity and penetrance is that with the former, the trait is expressed to a variable extent, while with the latter it may or may not be expressed even though the genetic makeup of the genotype suggests that it should be. It is often impossible to determine causation of penetrance and expressivity of a gene but both environmental factors and other modifier genes (genetic suppression and position effect) are known to influence them.

**5.3. Inheritance of factors controlling number of flowers per axis**

The results from the study of inheritance and allelic relationships of genes for different flowering types indicated that the double-flower trait and the triple-flower trait are controlled by a single locus (*Sfl*). The allele for double-flower trait ($sfl_d$) was dominant over the allele for triple-flower trait ($sfl_t$). The multi-flower trait is controlled by another locus (*cym*). The homozygous condition of recessive alleles at *cym* locus leads to expression of multi-flower trait. There are several reports on inheritance of double-flower/double-pod trait in chickpea. All these reports suggest that single-flower/single-pod trait is dominant over double-flower/double-pod trait and controlled by a single locus (Khan and Akhtar 1934; Ahmad 1964; Singh 1965; Patil 1966; D'Cruz *et al.*, 1970; More et al.,1976; ICRISAT annual report 1977; Yadav *et al.*, 1978; Pawar *et al.*, 1983 Rao *et al.*, 1983; Singh *et al.*, 1989; Kumar *et al.*,

2000a; Gaur and Gour 2002). This locus was designated '*S*' by Khan and Akhtar (1934), and '*Sfl*' by D'Cruz and Tendulkar (1970). The only report on inheritance of triple-flower trait is by Singh and Chaturvedi (1998). They demonstrated that triple-flower trait in chickpea is controlled by single recessive gene '*rtf*'. The allelic relationship of genes for double-flower trait and the triple-flower trait has been studied for the first time in the present study. As the results clearly demonstrate that the genes for these traits are allelic, there is a need to change the gene symbols '$sfl_d$' for the allele for double-flower trait and '$sfl_t$' for the allele for triple-flower trait.

The inheritance of multi-flower trait has been earlier studied by Gaur and Gour (2002) who identified the multi-flower mutant and developed the line JGM 7 used in this study. They also studied the allelic relationships of genes for double-flower and multi-flower traits and reported that two independent loci (*sfl* and *cym*) control number of flowers per axis in chickpea. Results from the present study confirm their findings. The allelic relationship of genes controlling triple-flower trait and multi-flower trait has been studied for the first time in the present investigation. The results clearly demonstrated that the genes for these two traits are not allelic and two different loci control these traits. These results are expected as the gene for triple-flower trait is allelic to the gene for double-flower trait.

### 5.4. Inheritance of flower colour

Flower colour has profound effect on other morphological and physiological patterns. The inheritance of flower colour (pink and pink-veined white flower) was studied in two crosses ICC 4929 x IPC 99-18 and ICC 4929 x JGM 7, where ICC 4929 had pink-veined white flower and the other parent had pink flower. In both the crosses, the $F_1$s were pink

flowered and $F_2$s segregated in a 3:1 ratio for pink and pink-veined white flower plants, suggesting monogenic control of these variants of flower colour and dominance of pink flower colour over pink-veined white flower colour. The results obtained by Pimplikar (1943), Khan *et al.* (1950), Argikar (1955), Argikar and D'Cruz (1963), Bhapkar and Patil (1963), More (1964), Patil (1964), Athwal and Brar (1967), Patil (1967), Khosh-Khui and Niknejad (1971), Jagtap *et al.* (1973), More and D'Cruz (1976b), Nayeem *et al.* (1977), Reddy and Nayeem (1978), Yadav *et al.* (1978), Pawar and Patil (1982 and 1983), Kidambi *et al.* (1988), Singh *et al.* (1988), Gil and Cubero (1993) Pundir and Reddy (1997), Tefera (1998), Sabaghpour (2000) and Kiran Kumar (2001) showed that pink is differentiated from white/blue/light velvet colours by a single factor.

However, there are studies that suggest that two genes (Khan and Akhtar,1934; Pal, 1934; Kadam *et al.*, 1941; More and D'Cruz, 1970, 1976a; Deshmukh *et al.*, 1972; Patil and Deshmukh, 1975; Reddy and Chopde, 1977a; Pawar and Patil, 1979; Rao *et al.*, 1980; Ghatge *et al.*, 1985; Davis 1991; Ghatge 1994a; and Kumar, 1997) or three genes (Ayyar and Balasubramaniyan, 1936; Balasubramaniyan, 1951; and Kumar *et al.*, 2000b) are involved in controlling flower colour.

The inheritance of different flowering colours can only be explained when we consider three-gene hypothesis for control of flower colour in chickpea. Depending on the genotypes of the parents, one gene, two genes or three genes can segregate in a given $F_2$ population. Ayyar and Balasubramaniyan (1936), Balasubramaniyan (1951) and Kumar *et al* (2000b) who studied genetics of white, blue and pink flower colours concluded that three independent

genes *C*, *B* and *P* were involved in the production of these colours. All the three factors together in dominant condition produced pink colour. Blue colour is produced when *B* is present in combination with *C*, and white flower colour is produced when either *B* or *C* is in homozygous recessive condition.

The inheritance of pink-veined white flower colour was first reported by Gaur and Gour (2001). They studied the pink-veined white flowered line ICC 4929, which has also been used in this study. From their study it is clear that a single factor '***ifc***' is responsible for the conversion of flower colour from pink to pink-veined white, and pink flower colour is dominant over pink-veined white. The homozygous recessive (***ifc ifc***) condition at the '***Ifc***' locus will inhibit the flower colour without modifying the vein colour Thus, pink flower colour (*P? BB Ifc Ifc*) changes to pink-veined white (*P? BB ifc ifc*). The genotypes for pink and pink-veined white flowered parents used in this study are similar to genotypes suggested by Gaur and Gour (2001).

**5.5. Inheritance of foliage colour**

Variations in foliage colour of chickpea occur mainly due to intensity of anthocyanin pigmentation. This can be used as a good genetic marker for studying the evolutionary trends and varietal identification in seed production programmes. Genetics of this character is not well understood.

In the present study low pigmentation (normal green foliage) was found to be dominant over high pigmentation (purple foliage) and followed monogenic inheritance in two

crosses ICC 492 x JGM 7 and IPC 99-18 x JGM 7. Similar results were obtained by Bhapkar and Patil (1962), Rao *et al.* (1980), and Kiran Kumar (2001) Monogenic inheritance of this trait has also been reported by More and D'Cruz (1976a), Ghatge (1994a), Mathur (1998) and Tefera (1998) but in their studies high pigmentation was found dominant over low pigmentation which is contrary to the findings of present study, which shows dominance of high pigmentation. There are also reports suggesting digenic inheritance of this trait Sandu *et al.* (1993) found 13:3 ratio for green and purple plants, indicating dominance of normal green over purple pigmentation, which was contrary to the findings of Ghatge and Kolhe (1985), who found that purple was dominant over pale green (9:7). Influence of light on the intensity of pigmentation was suggested by Mathur (1989) who reported two complementary genes for light dependent purple pigmentation.

The differences in dominance recessive relationship and gene interactions for the same trait in different studies might be due to differences in genotypes and mutations that might have taken place in both the directions.

**5.6. Genetic linkages**

A linkage was detected between two morphological characters flower and foliage colours in the cross ICC 4929 x IPC 99-18.

Joint segregation studies of flowering type and flower colour revealed that none of the two genes for flowering types was linked to the gene(s) for flower colour in two crosses (ICC 4929 x IPC 99-18 and ICC 4929 x JGM 7) studied. Pawar and Patil (1983), and Singh *et al* (1988) reported that corolla colour (*I.vco)* is linked with loci (*Sfl*) controlling number of

flowers per axis. We could not detect this linkage, as the only flower colour locus segregating in the two crosses was '*Ifc*'.

Joint segregation of flower colour and foliage colour in $F_2$ of ICC 4929 x JGM 7 showed the absence of recombinant phenotype having dark foliage and white flower colour. This indicates the possibility that a single gene has pleiotropic effects on flower colour and foliage colour. Ayyar and Balasubramanyam (1936), Argikar (1955) and D'Cruz and Tendulkar (1970) also suggested pleiotropic effects of a gene on expression of stem pigmentation and flower colour.

### 5.7. Genetic variability among flowering types for various traits

The plant breeders often need to deal with oligogenic and polygenic traits showing variable expression under different environments where in careful selection must be exercised in the early generations to get the desirable final product. More diverse the parents, the greater are the chances of recovering desirable combination and obtaining higher level of expression of that trait. Thus, crop improvement depends on the magnitude of genetic variability and number of loci controlling a trait in the base population. Chickpea is a self-pollinated crop and demand extensive studies to investigate and exploit the existing variability. Therefore, the present investigation was taken up to find out significant differences for various characters between different flowering types.

Significant differences were found between flowering types for various characters studied indicating variability in flowering types. There were no references before this study regarding variability between flowering types for comparison of these results.

The characters number of flowers per axis, number of pods per axis, yield per plant and 100-seed weight were found significantly different between flowering types. No significant differences were observed for number of pods per plant, number of seeds per plant, number of seeds per pod and number of empty pods. MF types recorded more seed size and yield per plant than other types in two crosses ICC 4929 x JGM 7 and IPC 99-18 x JGM 7; however the range was higher for SF types than MF types in both crosses. The significant differences observed for seed size among different flowering types need further examination. However, the results suggest possibility of selection of proper genetic background for exploitation of loci controlling yield contributing traits. More combinations of diverse genotypes would be needed to evaluate the effects of gene(s) controlling number of flowers per axis on yield.

# Chapter VI
# Summary

CHAPTER VI

## SUMMARY

The study was aimed at determining allelic relationships, penetrance and expressivity of genes controlling number of flowers per axis and to know the effect of these traits on yield and yield contributing characters in chickpea (*Cicer arietinum* L.). Inheritance was also studied for two morphological traits, flower colour and pigmentation. These studies were conducted at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) Patancheru, near Hyderabad, A.P. during 2004-05 post-rainy season. The material for investigation comprised of a double-flowered line (ICC 4929), a triple-flowered line (IPC 99-18) and a multi-flowered (JGM 7), six $F_1$s derived from crosses between these lines (included both direct and reciprocal) and three $F_2$s (ICC 4929 x IPC 99-18, IPC 99-18 x JGM 7 and ICC 4929 x JGM 7) excluding reciprocals.

Expression of flowering in chickpea is highly variable. The variability at expression level was studied for double-pod trait by few researchers. There are no reports on penetrance and expressivity of triple-flower and multi-flower traits. Hence, the present investigation was carried out to study the penetrance and expressivity of gene(s) controlling various flowering types.

In the parental lines, gene(s) for double, triple and multi-flower traits showed 100% penetrance. But the expressivity was variable for flowering and podding trait. In double-flowered line ICC 4929, 79.26% of flowering nodes (96.34%) converted into podding nodes

(76.36%). In triple-flowered line IPC 99-18, none of the triple-flowered nodes (81.15%) converted into triple-podded nodes indicating 0.00% expressivity for triple-pod trait. This line had more double-podded nodes (55.30%) than single-podded nodes (44.70%). The multi-podded line JGM 7 showed mean expressivity of 51.33% for multi-flower trait and 24.70% for multi-pod trait, indicating that 48.15% of multi-flowered nodes converted into multi-podded nodes. Results indicated that double-flower and double-pod traits had higher expressivity than other flowering and podding types.

Average number of flowers per axis was higher in multi-flowered (2.83) and triple-flowered (2.80) lines than in double-flowered (1.96) line. Multi-flowered line JGM 7 (2.16) exhibited more number of pods per axis than double-flowered line ICC 4929 (1.77) and triple-flowered line IPC 99-18 (1.44), indicating that multi-flower trait could contribute higher yield through more number of pods per axis.

Allelic relationships were worked out for genes controlling different flowering types. $F_2$ segregation in the cross ICC 4929 (DF) x IPC 99-18 (TF) indicated that double and triple-flower traits are controlled by a single gene (***Sfl***), and double-flower trait (***sfl$_d$***) is dominant to triple-flower trait (***sfl$_t$***). The $F_1$s of the crosses of multi-flowered line JGM 7 with double-flowered line ICC 4929 and the triple-flowered line IPC 99-18 were single-flowered. The $F_2$s of the cross IPC 99-18 (TF) x JGN 7 (MF) produced different flowering types that gave a good fit to a ratio 9 (SF) : 3 (TF) : 4 (MF+TM). The $F_2$ of the cross ICC 4929 (DF) x JGM 7 (MF) gave a good fit to 9 (SF) : 3 (DF) : 3 (MF) : 1 (DM) ratio. These results indicate that two loci (***Sfl_ Cym_***) control the number of flowers per axis in chickpea. Therefore, the following

genetic constitutions are suggested for different flowering types observed in parents, $F_1$s and $F_2$s :

**ICC 4929** (DF) ( ***sfl$_d$ sfl$_d$ Cym Cym*** ), **IPC 99-18** (TF) (***sfl$_t$ sfl$_t$ Cym Cym*** ), $F_1$ (DF) (***sfl$_d$ tfl$_t$ Cym Cym*** ), $F_2$ (DF) (***sfl$_d$ sfl$_d$ Cym Cym*** **and** ***sfl$_d$ tfl$_t$ Cym Cym*** ) and (TF) (***sfl$_t$ sfl$_t$ Cym Cym*** ).

**IPC 99-18** (TF) (***sfl$_t$ sfl$_t$ Cym Cym*** ), **JGM 7** (MF) ( ***Sfl Sfl cym cym*** ), $F_1$ (SF) ( ***Sfl sfl$_t$ Cym cym*** ), $F_2$ (SF) (***Sfl _ Cym _*** ), TF (***sfl$_t$ sfl$_t$ Cym _*** ), MF (***Sfl _ cym cym*** ) and TM (***sfl$_t$ sfl$_t$ cym cym*** ).

**ICC 4929** (DF) (***sfl$_d$ sfl$_d$ Cym Cym*** ), **JGM 7** (MF) ( ***Sfl Sfl cym cym*** ), $F_1$ (SF) ( ***Sfl sfl$_d$ Cym cym*** ), $F_2$ (SF) ( ***Sfl _ Cym _*** ), DF (***sfl$_d$ sfl$_d$ Cym _*** ), MF ( ***Sfl _ cym cym*** ) and DM (***sfl$_d$ sfl$_d$ cym cym*** ).

Monogenic inheritance was confirmed for two morphological traits, pink vs. pink-veined white flower, and green foliage vs. purple foliage, where pink flower colour and green foliage colour were dominant to their contrasting traits. Joint segregation analysis showed independent assortment of genes controlling flowering type and flower colour traits in the crosses ICC 4929 x IPC 99-18 and ICC 4929 x JGM 7. Foliage colour showed a weak linkage with flowering type with crossing over percentage 38.6% ($P= <0.01$) in the cross ICC 4929 x IPC 99-18. In the cross ICC 4929 x JGM 7, no single segregant was found to have purple

foliage with pink-veined white flower indicating the pleiotropic effect of a gene over both the traits.

Significant differences among flowering types were observed in segregating populations for number of flowers per axis, number of pods per axis, yield per plant and 100-seed weight. Among flowering types, TF and DM produced more than two flowers per axis. TF plants produced more number of pods per axis, while MF and DM types had higher yield per plant and 100-seed weight. In spite of no significant difference among flowering types, DM types recorded higher mean values for characters number of pods per plant and number of seeds per plant, and MF types recorded less number of empty pods per plant.

**Conclusions**

Based on the results of the present investigation the following conclusions are drawn:

1. Two loci (*Sfl_ Cym_*) govern number of flowers per axis in chickpea. The gene for double-flower trait (*$sfl_d$*) and the gene for triple-flower trait (*$sfl_t$*) are allelic and belong to the locus '*Sfl*'. The allele '*$sfl_d$*' is dominant over the allele '*$sfl_t$*'. The double recessive at the '*Cym*' locus (*cym cym*) produces multi-flower trait.

2. The following genotypes are proposed for different flowering types

| | |
|---|---|
| Single-flowered (SF) | = *Sfl_ Cym_* |
| Double-flowered (DF) | = *$sfl_d$ $sfl_d$ Cym_ or $sfl_d$ $sfl_t$ Cym_* |
| Triple-flowered (TF) | = *$sfl_t$ $sfl_t$ Cym_* |

Multi-flowered (MF) = ***Sfl_ cym cym***

Double-multiflowered (DM) = ***$sfl_d$ $sfl_d$ cym cym or $sfl_d$ $sfl_t$ cym cym***

Triple-multiflowered (TM) = ***$sfl_t$ $sfl_t$ cym cym***

3. The genes controlling number of flowers per axis exhibited 100% penetrance in the parental lines.

4. Variable expressivity was found for genes controlling number of flowers/pods per axis in parental lines. Double-flower trait showed higher level of expression than triple-flower and multi-flower traits. Similarly, double-pod trait showed higher level of expression than multi-pod trait. Triple-pod trait exhibited zero% of expression.

5. Monogenic inheritance was observed for flower colour (pink flower vs. pink-veined white flower). The allele '***ifc***' in homozygous condition inhibits flower colour without affecting the vein colour i.e. pink flower colour changes to pink-veined white flower colour.

6. Pigmentation on foliage was found to be controlled by a single gene with normal green dominant to purple

7. The $F_2$ segregants with purple foliage always had pink flowers, suggesting that a gene has a pleiotropic effects on flower colour and foliage colour.

8. Multi-flowered line showed more number of flowers and pods per axis than double-flowered and triple-flowered lines.

9. Variability was observed among flowering types for number of flowers per axis, number of pods per axis, yield per plant and 100-seed weight. Multi and double-multiflowered types exhibited higher yield per plant and seed size than other flowering types.

The results of this study are preliminary and further investigations are needed to assess the advantages of double, triple or multi-flower traits in chickpea. These traits should be first transferred to suitable agronomic backgrounds and then fixed lines should be developed after continuous selection of the plants with higher expression of these traits. These lines should then be evaluated in replicated trails in different agroclimatic conditions

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* Originals not seen

## APPENDIX I

### Weather data during the crop growth period (October 8 – March 4, 2004/05).

| Year | Std Week | Rain (in mm) | Evap (in mm) | Max Temp (in°C) | Min Temp (in°C) | Rel Humidity1 at 07:17 (in%) | Rel Humidity2 at 14:17 (in%) | Wind Velocity (in Kmph) | Solar Radiation (in mj/ $m^2$) | Bright Sunshine (in Hrs) |
|---|---|---|---|---|---|---|---|---|---|---|
| 2004 | 41 | 11.4 | 27.3 | 31.57 | 21.19 | 96.00 | 57.00 | 3.34 | 16.62 | 6.44 |
| 2004 | 42 | 14.69 | 31.29 | 29.45 | 15.62 | 90.28 | 43.71 | 2.08 | 20.54 | 9.20 |
| 2004 | 43 | 1.39 | 25.6 | 29.68 | 19.37 | 94.42 | 56.14 | 4.34 | 14.92 | 6.71 |
| 2004 | 44 | 0 | 34.6 | 29.04 | 18.14 | 86.85 | 51.71 | 8.05 | 14.94 | 5.65 |
| 2004 | 45 | 0 | 31.89 | 29 | 17.97 | 85.57 | 42.85 | 5.70 | 14.82 | 6.31 |
| 2004 | 46 | 0 | 29.6 | 30.85 | 17.44 | 93.00 | 44.57 | 5.04 | 15.31 | 8.12 |
| 2004 | 47 | 0 | 38.1 | 29.92 | 12.11 | 87.57 | 29.14 | 4.85 | 18.02 | 10.24 |
| 2004 | 48 | 0 | 38 | 28.91 | 11.51 | 83.57 | 28.00 | 5.20 | 17.44 | 10.18 |
| 2004 | 49 | 0 | 31.39 | 28.68 | 10 | 88.71 | 31.57 | 2.11 | 17.08 | 10.08 |
| 2004 | 50 | 0 | 29.4 | 28.92 | 11.11 | 95.42 | 35.71 | 1.51 | 16.22 | 9.71 |
| 2004 | 51 | 0 | 29.69 | 30.65 | 11.21 | 92.28 | 31.42 | 2.17 | 16.50 | 10.05 |
| 2004 | 52 | 0 | 34.69 | 28.8 | 12.69 | 96.37 | 43.87 | 4.60 | 15.61 | 9.40 |
| 2005 | 1 | 0 | 34.89 | 29.61 | 15.54 | 94.28 | 45.28 | 6.62 | 14.85 | 8.37 |
| 2005 | 2 | 0 | 30.8 | 30.21 | 12.19 | 90.71 | 41.85 | 3.52 | 16.95 | 9.87 |
| 2005 | 3 | 0 | 35.89 | 29.74 | 14.11 | 86.71 | 45.28 | 5.78 | 17.13 | 9.78 |
| 2005 | 4 | 2 | 34.89 | 30.41 | 19 | 95.00 | 44.42 | 8.91 | 14.25 | 7.48 |
| 2005 | 5 | 31 | 33.19 | 27.92 | 16.34 | 92.71 | 54.42 | 8.17 | 14.84 | 6.94 |
| 2005 | 6 | 0 | 43.9 | 31.11 | 15.31 | 88.85 | 32.28 | 7.12 | 18.45 | 9.64 |
| 2005 | 7 | 0 | 44.9 | 34.85 | 14.64 | 73.42 | 20.57 | 4.38 | 20.62 | 10.67 |
| 2005 | 8 | 0 | 50.3 | 34.24 | 17.71 | 81.42 | 23.57 | 6.81 | 19.3 | 9.97 |
| 2005 | 9 | 0 | 62.2 | 34.94 | 17.6 | 69.00 | 21.71 | 6.75 | 19.71 | 9.98 |