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Inheritance and relationships of flowering time and seed size in *kabuli* chickpea

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Abstract Flowering time and seed size are the important traits for adaptation in chickpea. Early phenology (time of flowering, podding and maturity) enhance chickpea adaptation to short season environments. Along with a trait of consumer preference, seed size has also been considered as an important factor for subsequent plant growth parameters including germination, seedling vigour and seedling mass. Small seeded *kabuli* genotype ICC 16644 was crossed with four genotypes (JGK 2, KAK 2, KRIPA and ICC 17109) to study inheritance of flowering time and seed size. The relationships of phenology with seed size, grain yield and its component traits were studied. The study included parents, F₁, F₂ and F₃ of four crosses. The segregation data of F₂ indicated flowering time in chickpea was governed by two genes with duplicate recessive epistasis and lateness was dominant to earliness. Two genes were controlling 100-seed weight where small seed size was dominant over

large seed size. Early phenology had significant negative or no association (ICC 16644 × ICC 17109) with 100-seed weight. Yield per plant had significant positive association with number of seeds per plant, number of pods per plant, biological yield per plant, 100-seed weight, harvest index and plant height and hence could be considered as factors for seed yield improvement. Phenology had no correlation with yield per se (seed yield per plant) in any of the crosses studied. Thus, present study shows that in certain genetic background it might be possible to breed early flowering genotypes with large seed size in chickpea and selection of early flowering genotypes may not essentially have a yield penalty.

Keywords Adaptation · *Cicer arietinum* · Correlation · Genetics · Market trait · Phenology

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Introduction

Globally chickpea (*Cicer arietinum* L.) is the third most important food legume crop in the world after beans (*Phaseolus vulgaris* L.) and pea (*Pisum sativum* L.) in terms of annual production (FAOSTAT 2017). It is grown over an area of 14.56 million hectares with a production of 14.77 million tonnes and productivity of 1014 kg per hectare. India is the largest chickpea producing country in the world with a share of 61.4%

(9.07 million tonnes) in production and 65.5% (9.53 million hectares) in area (FAOSTAT 2017). During the past decade, chickpea production increased considerably in Russian Federation, Australia, Tanzania, Ethiopia, United States, Myanmar and India (Gaur et al. 2018). Due to its nutritive seed, which is high in protein content, its use as substitute for animal protein is increasing which is leading to expansion of chickpea area in the world. Along with the yield, phenology (time of flowering, podding and maturity) and seed size are the two important traits in chickpea which decide the choice of farmers' preferences to chickpea variety. Large genotypic variations exist for flowering time in chickpea. Phenology plays critical role in adaptation of chickpea to different environments (Berger and Turner 2004; Berger et al. 2006; Gaur et al. 2008a, b) particularly in semi-arid regions where growth is restricted by water availability and by seasonal temperature profile (Bonfil and Pinthus 1995; Subbarao et al. 1995). Chickpea often experiences short growing season because of terminal stresses (drought, extremes temperatures) which often restrict its yield potential (Khanna-Chopra and Sinha 1987). Early maturity is an important trait for increasing and stabilizing chickpea productivity by avoiding end of season drought (Subbarao et al. 1995; Kumar and Abbo 2001). Early flowering leads to prolong reproductive phase thereby increasing the yield through more efficient water use system (Kumar and Abbo 2001). Early flowering is important in temperate environments for escaping end-of-season frost (Warkentin et al. 2003; Gaur et al. 2015) In chickpea, time of flowering is variable depending upon season, date of sowing, latitude and altitude (Summerfield and Roberts 1988), it is a function of temperature and photoperiod (Roberts et al. 1985; Ellis et al. 1994) or solely photoperiod (Ellis et al. 1994). Studies suggest that flowering time is governed by one or few major genes (Gumber and Singh 1996; Or et al. 1999; Kumar and van Rheenen 2000; Anbessa et al. 2006; Hegde 2010; Gaur et al. 2015). In chickpea, four flowering genes have been identified, *eft-1* (Kumar and van Rheenen 2000), *eft-2* (Or et al. 1999), *eft-3* (Hegde 2010), and *eft-4* (Gaur et al. 2015). Early flowering genes can be introduced into promising cultivars of late flowering genetic backgrounds. However, breeding programmes with a goal of developing varieties with early phenology, other traits must also be considered. Within cultivated chickpea, two distinct

groups of cultivars are found; *desi* type (pink flowers, angular shaped and brown coloured small seeds) and *kabuli* type (white flowers, owl's head shaped and beige coloured large seeds). Large-seeded *kabuli* types are gaining importance as the market price of *kabuli* chickpea is up to twice that of *desi* chickpea (Upadhyaya et al. 2006). In *kabuli* chickpea seed size is an important trait. A wide range of genetic variability is present for seed size in chickpea. Larger seed size coupled with other desirable seed traits (e.g. light colour) commands price premiums in a market-dependant manner (Graham et al. 2001). It is an important component of yield and adaptation (Singh and Paroda 1986). It has also been considered as an important factor for subsequent plant growth parameters including germination, seedling vigour and seedling mass (Narayanan et al. 1981; Dahiya et al. 1985). Earlier studies have reported monogenic (Argikar 1956), digenic (Ghatge 1993; Upadhyaya et al. 2006; Hossain et al. 2010), oligogenic (Patil and D'Cruze 1964) and polygenic (Niknejad et al. 1971; Kumar and Singh 1995; Malhotra et al. 1997; Kumhar et al. 2013) inheritance of seed size depending on the number of genes segregating in the populations studied. According to Smithson et al. (1985) and Kumar and Singh (1995), small seed size was dominant over large one. In contrast, Niknejad et al. (1971) stated that large seed size was partially dominant over the small seed size. The study of inheritance of seed size and flowering time is important for adopting appropriate breeding strategy for developing improved cultivar of chickpea. In framework of an effort to breed early flowering genotypes with large seeds the present investigation was carried out to determine inheritance of flowering time and seed size and whether the phenology affects seed size. In addition, the relationships of phenology with grain yield and its component traits were studied.

Materials and methods

Five genotypes of chickpea, which included two landraces (ICC 16644 and ICC 17109) and three cultivars (KAK 2, KRIPA and JGK 2) were used as parents for four crosses in the study. All the genotypes were *kabuli* type. The descriptions of parental lines are given in Table 1. To study the genetics and segregation patterns of seed size and flowering time, parents

Table 1 Origin, pedigree and key traits of the parental genotypes

Genotype	Origin/pedigree	Key traits
ICC 16644	A land race from Punjab province of Pakistan	<i>Kabuli</i> type, semi-spreading growth habit, super early and small seed size
JGK 2	(ICC12339 × ICC4967) × [(ICC982 × ICC4973) × ICC15980] × ICC12975]	<i>Kabuli</i> type, semi-spreading growth habit, medium duration and medium seed size
KAK 2	(ICCV2 × Surutato 77) × ICC 7344	<i>Kabuli</i> type, semi-spreading growth habit, medium duration and medium seed size
KRIPA	Also called Phule G 0517, a selection from local germplasm	<i>Kabuli</i> type, semi-spreading growth habit, medium duration and large seed size
ICC 17109	A line from Mexico	<i>Kabuli</i> type, semi-spreading growth habit, late and large seed size

differing in both the traits were selected for crossing. ICC 16644 was an early maturing line (early flowering) with small seed size. The remaining genotypes had medium maturity and large (JGK 2 and KAK 2) to extra-large (ICC 17109 and KRIPA) seed size. Four crosses were made by crossing early flowering and small seeded genotype 16644 with the remaining four genotypes KAK 2, JGK 2, KRIPA and ICC 17109. The crosses ICC 16644 × JGK 2, ICC 16644 × KAK 2, ICC 16644 × KRIPA and ICC 16644 × ICC 17109 were designated as C₁, C₂, C₃ and C₄ respectively. The F₁, F₂ and F₃ along with the respective parents of each cross were sown in the field in November, 2013 (post-rainy season) at International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) Patancheru, India (17°52' N 78°24' E). Seeds were treated before sowing with a mixture of 2 g thiram and 1 g of carbendazim per kg of seed. The seeds were sown at a wider spacing of 60 cm × 20 cm with single seed per hill in the rows of 4 m. Care was taken to sow the seeds at uniform depth (5 cm). The plots of various generations contained different number of rows i.e., two rows of parents, one row of F₁, and six rows each of F₂ and F₃. All recommended agronomical practices (Gaur et al. 2010) and necessary plant protection measures were followed to raise a healthy crop. The minimum and maximum temperature ranged between 8.31–18.34 °C and 26.54–32.22 °C respectively during the experimental period. One intercultural operation was done to control the weeds and three sprays of Indoxacarb (@20 mL/ha in 300 L water) were done to

manage pod borer (*Helicoverpa armigera*). Observations were recorded on individual plants (20 plants in parents and F₁, 210 plants each in F₂ and F₃ per cross) for days to first flower, days to first pod formation, days to maturity, plant height (cm), number of pods per plant, number of seeds per plant, number of seeds per pod, grain yield per plant (g), biological yield per plant (g) and 100-seed weight (g). The day first flower fully opened was recorded as days to first flower. The weight of 100 randomly selected seeds from each plant was recorded as 100-seed weight. For those plants which had less than 100-seeds, the weight of 100-seed in grams was calculated by the following formula:

$$100\text{-seed weight (HSW)} = \frac{\text{Weight of total seed of the plant (g)}}{\text{Total number of seeds of the plant}} \times 100$$

Data were subjected to mean, variance, range and standard error estimation. Based on distribution pattern in F₂ and F₃ population, the quantitative data of days to first flower and 100-seed weight collected from individual plants were converted into qualitative data using different class intervals. The qualitative data was analyzed using χ^2 test for Mendelian ratio. In addition, standard statistical procedure, *t* test, regression and simple correlations were used to analyze the data using GENSTAT (version 18.0).

Table 2 Days to first flower, maturity and 100-seed weight of parents, F₁, F₂, and F₃ of four crosses in chickpea

S. no.	Parent/generation	Days to first flower		Days to maturity		100-seed weight (g)	
		Mean ± SE	Range	Mean ± SE	Range	Mean ± SE	Range
1	ICC 16644	28.0 ± 0.17	27–30	80.5 ± 0.12	80–82	24.6 ± 0.07	22.9–24.8
2	JGK 2	36.7 ± 0.17	34–38	87.5 ± 0.16	85–88	36.8 ± 0.58	32.5–35.8
3	KAK 2	35.8 ± 0.13	35–39	87.5 ± 0.14	86–89	42.5 ± 0.54	39.5–43.4
4	KRIPA	40.8 ± 0.16	37–41	91.2 ± 0.12	88–95	51.8 ± 1.98	49.5–53.1
5	ICC 17109	40.2 ± 0.14	38–42	93.5 ± 0.17	89–94	60.6 ± 1.17	57.8–62.0
6	F ₁ (ICC 16644 × JGK 2)	48.5 ± 0.26	48–51	92.4 ± 0.26	90–95	25.3 ± 0.70	23.7–25.9
7	F ₁ (ICC 16644 × KAK 2)	49.4 ± 0.25	49–54	91.8 ± 0.25	90–96	28.6 ± 0.59	26.6–28.7
8	F ₁ (ICC 16644 × KRIPA)	51.1 ± 0.24	50–54	94.6 ± 0.63	92–100	33.2 ± 0.96	32.7–34.0
9	F ₁ (ICC 16644 × ICC 17109)	51.4 ± 0.14	51–54	96.6 ± 0.38	94–101	33.7 ± 0.81	32.3–35.6
10	F ₂ (ICC 16644 × JGK 2)	40.1 ± 0.63	27–58	87.1 ± 0.63	78–111	26.4 ± 0.69	18.3–51.7
11	F ₂ (ICC 16644 × KAK 2)	47.3 ± 0.76	27–64	93.2 ± 0.74	78–110	30.3 ± 0.94	15.8–57.7
12	F ₂ (ICC 16644 × KRIPA)	42.3 ± 0.53	27–63	90.2 ± 0.53	77–110	36.6 ± 0.88	14.5–54.8
13	F ₂ (ICC 16644 × ICC 17109)	42.5 ± 0.71	27–62	91.2 ± 0.76	76–112	35.8 ± 0.79	21.9–61.9
14	F ₃ (ICC 16644 × JGK 2)	43.4 ± 0.56	29–61	87.1 ± 0.12	78–110	30.8 ± 0.50	11.7–63.7
13	F ₃ (ICC 16644 × KAK 2)	47.6 ± 0.73	28–75	93.3 ± 0.26	79–107	28.7 ± 0.45	14.3–54.2
16	F ₃ (ICC 16644 × KRIPA)	45.5 ± 0.79	28–73	94.6 ± 0.61	78–111	34.3 ± 0.48	18.3–55.9
17	F ₃ (ICC 16644 × ICC 17109)	46.0 ± 0.63	29–71	95.4 ± 0.36	79–111	35.5 ± 0.54	21.3–62.9

Results and discussion

Inheritance of flowering time

The flowering time of parental lines varied from 27 to 42 days (Table 2). The F₁s of all the crosses were late to flower with mean flowering time of 48.5, 49.4, 51.1 and 51.4 days in C₁, C₂, C₃ and C₄ respectively, indicating dominance of lateness in all the four crosses. In chickpea, late flowering is known to be dominant over early flowering (Gumber and Singh 1996; Or et al. 1999; Kumar and van Rheenen 2000; Anbessa et al. 2006; Hegde 2010; Gaur et al. 2015). F₂ and F₃ populations of all crosses had wide variation for flowering time. The frequency distributions of flowering timing in F₂ of each cross was skewed towards late parent and also number of plants with late flowering was much higher than the number of plants with early flowering, indicating late flowering is dominant over early flowering. F₂ segregation for days to first flower in C₁, C₂, C₃ and C₄ had a range of 27–58 days, 27–64 days, 27–63 days, and 27–62 days respectively. All F₂ populations had transgressive segregants in both directions for flowering time. The

individuals were grown during post-rainy season and they flowered during the period when temperatures were not very high [28.1 °C (average maximum temperature) and 13.1 °C average minimum temperature)] and days were long with mean bright sunshine hours of 9.7 (28–36 days after sowing), 8.1 (36–56 days after sowing) and 7.0 (57–70 days after sowing). Physiological study revealed that flowering time is a function of temperature and photoperiod in chickpea (Roberts et al. 1985). Three factors, response to photoperiod, response to temperature and “earliness per se genes” have been reported to determine time of flowering in wheat (Snape et al. 2001). Transgressive segregation in the study may be the results of new genetic combinations related to photo-thermal response and earliness per se genes.

The quantitative data for flowering time in each F₂ was converted to qualitative data and individuals were classified into two groups i.e. early and late flowering depending upon natural break points in the distribution frequency within each population (Fig. 1). Classification of F₂ individual into groups varied among crosses, it may be due to individual effect of genes present in the parent selected for the crosses. The F₂ populations

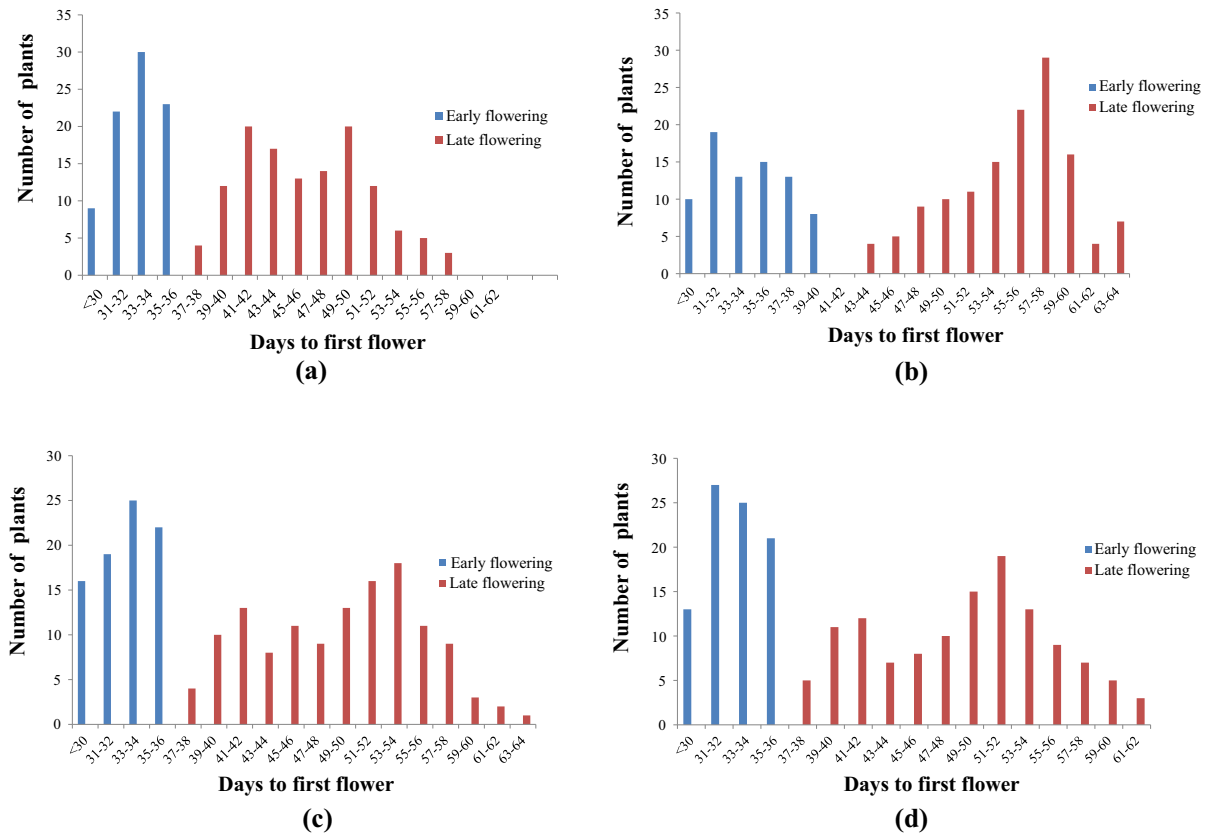


Fig. 1 **a** Distribution of flowering time in F_2 populations of the cross ICC 16644 \times JGK 2. **b** Distribution of flowering time in F_2 populations of the cross ICC 16644 \times KAK 2. **c** Distribution

of flowering time in F_2 populations of the cross ICC 16644 \times KRIPA. **d** Distribution of flowering time in F_2 populations of the cross ICC 16644 \times ICC 17109

of all the crosses gave good fit to a ratio of late to early flowering of 9:7 with non-significant χ^2 values (Table 3). This indicates that the flowering time was governed primarily by two genes with duplicate recessive epistasis between them. In chickpea, Anbessa et al. (2006) and Gaur et al. (2015) reported two major genes with duplicate recessive epistasis controlling flowering time. In a different study using F_2 population, it was reported that flowering time in chickpea was governed by duplicate dominant genes (good fit to a 9:6:1 ratio) with cumulative but unequal effect (Hegde 2010). Segregation pattern may differ in different studies depending upon variable effects of major and minor genes present for the flowering time and the classification of individuals in different classes. Genes responsible for flowering time are identified in many legumes. One gene in each was reported in common bean (Coyné and Mattson 1964)

and lentil (Sarker et al. 1999), two in pigeon pea (Koebner et al. 1991; Craufurd et al. 2001), six in pea (Murfet 1985 and eight in soybean (Bernard 1971; Buzzell 1971; Buzzell and Voldeng 1980; McBlain and Bernard 1987; Ray et al. 1995; Bonato and Vello 1999; Cober and Voldeng 2001). Late flowering was dominant to early flowering in all the above studies except for pigeon pea (Saxena and Sharma 1990) and common bean (Coyné and Mattson 1964) where earliness was dominant to lateness. The genetic basis of flowering time genes observed in the present study reveals that early-flowering trait in chickpea can be easily incorporated into high-yielding cultivars by backcross breeding or by selection of desired type individual in F_2 and subsequent generations.

Table 3 Goodness-of-fit (χ^2 -test) for a 9:7 ratio for late and early flowering; and small and large seed size plants observed in F₂ of four crosses in chickpea

S. no.	Trait/cross	F ₂ observed phenotype		Expected ratio	χ^2 (ns)	P-value
		Late flowering	Early flowering			
	Flowering time					
1	ICC 16644 × JGK 2	126	84	9:7	1.19	0.27
2	ICC 16644 × KAK 2	132	78	9:7	3.71	0.06
3	ICC 16644 × KRIPA	128	82	9:7	1.87	0.18
4	ICC 16644 × ICC 17109	124	86	9:7	0.66	0.42
	Seed size	Small seeded	Large seeded			
5	ICC 16644 × JGK 2	116	94	9:7	0.09	0.76
6	ICC 16644 × KAK 2	120	90	9:7	0.06	0.79
7	ICC 16644 × KRIPA	125	85	9:7	0.91	0.34
8	ICC 16644 × ICC 17109	118	92	9:7	0.01	0.98

ns non-significant, P-value probability value

Inheritance of seed size

100-seed weight of JGK 2, KAK 2, KRIPA and ICC 17109 were on average 49.8%, 72.8%, 110.5% and 136.6% heavier than that of the small seeded parent (ICC 16644). The 100-seed weight of ICC 16644 was 24.6 g with a range of 22.9–24.8 g. The 100-seed weight of F₁s were 25.3 g, 28.6 g, 33.2 g and 33.7 g in C₁, C₂, C₃ and C₄ respectively. The 100-seed weight of F₁s was lower than that of the mid parental value in all the crosses indicating, small seed size is dominant over large seed size. Majority of previous studies indicated dominance of small seed size over large seed size (Smithson et al. 1985; Kumar and Singh 1995; Malhotra et al. 1997; Upadhyaya et al. 2006; Hossain et al. 2010; Upadhyaya et al. 2011) except Niknejad et al. (1971) who stated that large seed size was partially dominant over small seed size. Seed size in legume crops is generally attributed to the cell number and cell size of cotyledons (Lemontey et al. 2000). The range and variation in 100-seed weight in segregating generations (F₂ and F₃) were high in all the crosses. The mean 100-seed weight of parents, F₁, F₂ and F₃ are given in Table 2. Despite of continuous variation exhibited by seed size of F₂ in all crosses, frequency distribution of seed size displayed definite segregating patterns. The quantitative data on 100-seed weight were converted into two different phenotypic classes (small seeded and large seeded) based on natural breakpoints in the distribution frequency. For example, in frequency distribution of 100-seed weight of

individual plants of F₂ in C₁, two peaks at 29 g and 39 g and valley (break point) at 35 g were observed (Fig. 2). So, the individual plants with 100-seed weight up to 35 g were grouped into small seeded and those having 100-seed weight more than 35 g were grouped into large seeded. Similarly, natural breakpoints of 100-seed weight were observed at 31 g (C₂) and 39 g (C₃ and C₄) and F₂ individuals of each cross were divided into two groups. All the four crosses exhibited skewness of data on 100-seed weight towards smaller seed weight. The numbers of individuals with small seed size and large seed size in F₂ populations of each cross fitted well to the expected ratio of 9:7 suggesting 100-seed weight in all crosses were governed by two genes with complementary gene action (Table 3). These results were consistent with di-genic inheritance with duplicate recessive epistasis. In a previous study of a cross between two *kabuli* chickpea the number of plants in the three groups in F₂ fitted well to an expected ratio of 5:6:5 and in backcross generations to an expected ratio 1:2:1 which suggested that seed size in the two parents is controlled by two genes exhibiting additive effects with each parent having one pair of alleles with increasing effect at one locus in homozygous form (Upadhyaya et al. 2011). The F₂ plants of different study fitted well to the expected ratio of 12:3:1 (Upadhyaya et al. 2006) suggesting that seed size in chickpea is controlled by two genes exhibiting dominance epistasis with dominance of normal seed size over small seed size. Di-genic mode of inheritance for

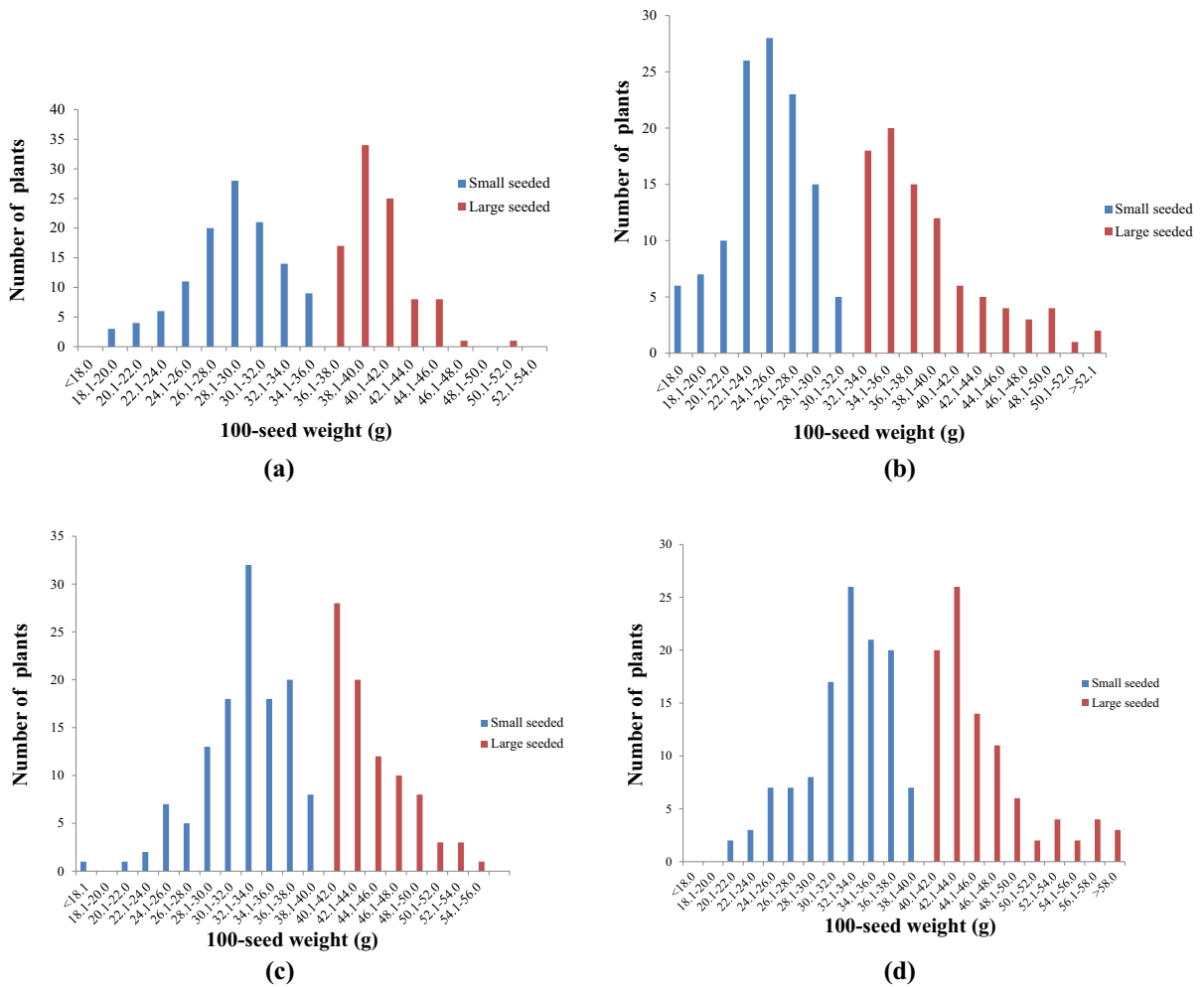


Fig. 2 **a** Distribution of 100-seed weight in F_2 populations of the cross ICC 16644 \times JGK 2. **b** Distribution of 100-seed weight in F_2 populations of the cross ICC 16644 \times KAK 2.

c Distribution of 100-seed weight in F_2 populations of the cross ICC 16644 \times KRIPA. **d** Distribution of 100-seed weight in F_2 populations of the cross ICC 16644 \times ICC 17109

seed size in chickpea has been reported earlier (Ghatge 1993; Upadhyaya et al. 2006; Hossain et al. 2010). In some studies, it was considered monogenic (Argikar 1956), oligogenic (Balasubrahmanyam 1950; Patil and D’Cruze 1964) and polygenic (Niknejad et al. 1971; Kumar and Singh 1995; Malhotra et al. 1997; Kumhar et al. 2013) depending on the number of genes segregating in the populations.

Comparisons of means of different groups based on days to flowering and 100-seed weight

The mean of 100-seed weight of two groups i.e. early flowering and late flowering were compared with each other in each cross using *t*-test to find out whether the flowering individuals had more 100-seed weight and vice versa (Table 4). Significant differences for 100-seed weight were observed between early flowering group and late flowering group in C_1 (*t* value: 4.08, $P < 0.001$) and C_2 (*t* value 4.51, $P < 0.001$) indicating early flowering individuals could assimilate more photosynthates as compared to late flowering

Table 4 Differences in mean seed size (100-seed weight) between early and late flowering groups of F₂ plants and differences between mean days to first flower between small-seeded and large-seeded groups of F₂ plants

Cross	No. of plants	100-seed weight (mean ± SE) of early flowering plants of F ₂	No. of plants	100-seed weight (mean ± SE) of late flowering plants of F ₂	<i>t</i> -test	<i>P</i> -value
ICC 16644 × JGK 2	84	35.6 ± 0.91	126	31.6 ± 0.75	4.87*	< 0.001
ICC 16644 × KAK 2	78	33.0 ± 0.84	132	28.4 ± 0.78	4.51*	< 0.001
ICC 16644 × KRIPA	82	38.1 ± 0.78	128	37.7 ± 0.79	0.44	0.33
ICC 16644 × ICC 17109	86	39.4 ± 0.82	124	39.2 ± 0.84	0.45	0.11
Cross	No. of plants	Days to first flower (mean ± SE) of small seeded plants of F ₂	No. of plants	Days to first flower (mean ± SE) of large seeded plants	<i>t</i> -test	<i>P</i> -value
ICC 16644 × JGK 2	116	43.2 ± 0.58	94	37.2 ± 0.57	5.22*	< 0.001
ICC 16644 × KAK 2	120	50.6 ± 0.54	90	43.2 ± 0.64	5.06*	< 0.001
ICC 16644 × KRIPA	125	42.7 ± 0.64	85	41.7 ± 0.68	0.80	2.11
ICC 16644 × ICC 17109	118	42.9 ± 0.46	82	41.8 ± 0.51	0.91	0.18

*Significant difference at $P < 0.001$

individuals in these crosses. In contrast, means of 100-seed weight of early and late flowering groups of C₃ and C₄ were at par, indicating late and early flowering group do not differ significantly in their mean 100-seed weight. Likewise, the mean of days to first flower for two groups of seed size i.e. small seeded group and large seeded group were compared. Significant differences for days to first flower in C₁ and C₂ were observed. In C₁ there were a difference of 10.6 g (100-seed weight) and 6 days (days to first flower) between the groups. In C₂, groups had difference of 12 g (100-seed weight) and 7.4 days (days to first flower). There were non-significant differences between the mean days to first flower of two groups for seed size in C₃ and C₄. These findings are in agreement with the findings from correlation studies.

Association among phenological traits

The data observed on F₂ individuals were used to calculate correlation coefficients between flowering time and other phenological traits and, morphological

and yield traits (Table 5). The association analysis revealed that phenological traits i.e. days to first flower, days to first pod formation and days to maturity were significantly positively correlated among each other in all the four crosses, suggesting early flower initiation leads to early pod setting which further leads to early maturity of genotype. Also, observations on flowering time can be recorded with more precision than on days to maturity (Gaur et al. 2015) particularly in long growing season environments thus flowering time can be used to select for early maturity. Gaur et al. (2015) suggested that, in general, the early flowering genotypes also mature early and the early flowering does not result in extending of reproductive period under residual soil moisture condition. However, in early flowering genotypes the duration of reproductive period may get extended due to indeterminate growth habit of chickpea (Subbarao et al. 1995). In the study, there was no supplementary irrigation or precipitation, these conditions might result early flowering lines to mature early, without extending the duration of reproductive phase. Several studies reported significant positive association among days to flowering and

Table 5 Phenotypic correlation coefficients among studied traits in F₂ population of four crosses in chickpea

Trait	Cross	Days to pod initiation	Days to maturity	Plant height	No. of seeds per plant	No. of seeds per pod	Biological yield per plant	Grain yield per plant	Harvest index	100-seed weight
Days to first flower	ICC 16644 × JGK 2	0.99**	0.82**	0.24**	0.25**	- 0.28**	0.16*	- 0.05	- 0.24**	- 0.35**
	ICC 16644 × KAK 2	0.99**	0.89**	0.14*	0.08	- 0.37**	0.14*	- 0.05	- 0.44**	- 0.30**
	ICC 16644 × KRIPA	0.99**	0.89**	0.19**	0.03	- 0.34**	0.09	- 0.08	- 0.47**	- 0.14*
Days to first pod initiation	ICC 16644 × ICC 17109	0.99**	0.88**	0.21**	0.05	- 0.23**	0.03	- 0.09	- 0.40**	- 0.09
	ICC 16644 × JGK 2		0.82**	0.14*	0.24**	- 0.26**	0.15*	- 0.05	- 0.23**	- 0.35**
	ICC 16644 × KAK 2		0.89**	0.14*	0.08	- 0.36**	0.15*	- 0.05	- 0.44**	- 0.30**
Days to maturity	ICC 16644 × KRIPA		0.88**	0.18**	0.03	- 0.32**	0.09	- 0.07	- 0.44**	- 0.14*
	ICC 16644 × ICC 17109		0.88**	0.20**	0.05	- 0.22**	0.03	- 0.08	- 0.38**	- 0.08
	ICC 16644 × JGK 2		0.14*	0.14*	0.22**	- 0.19**	0.18*	- 0.10	- 0.21**	- 0.21**
Plant height	ICC 16644 × KAK 2		0.14*	0.16*	0.14*	- 0.36**	0.16*	- 0.03	- 0.42**	- 0.28**
	ICC 16644 × KRIPA		0.20**	0.04	0.04	- 0.26**	0.09	- 0.09	- 0.45**	- 0.14*
	ICC 16644 × ICC 17109		0.26**	0.04	0.26**	0.06	- 0.03	- 0.03	- 0.36**	- 0.09
No. of seeds per plant	ICC 16644 × JGK 2		0.09	0.09	0.09	0.09	0.30**	0.33**	0.02	0.19**
	ICC 16644 × KAK 2		0.09	0.09	0.09	- 0.01	0.23**	0.18**	- 0.03	0.23**
	ICC 16644 × KRIPA		0.12	0.12	0.23**	- 0.18**	0.30**	0.26**	- 0.02	0.14*
No. of seeds per pod	ICC 16644 × ICC 17109		0.05	0.05	0.05	- 0.05	0.23**	0.18**	- 0.11	0.14*
	ICC 16644 × JGK 2		0.09	0.09	0.09	- 0.25**	0.89**	0.86**	0.32**	- 0.23**
	ICC 16644 × KAK 2		0.01	0.01	0.15*	0.15*	0.85**	0.89**	0.39**	- 0.30**
Biological yield per plant	ICC 16644 × KRIPA		0.09	0.09	0.09	0.01	0.89**	0.91**	0.23**	- 0.18**
	ICC 16644 × ICC 17109		0.05	0.05	0.05	0.09	0.92**	0.93**	0.14*	- 0.18**
	ICC 16644 × JGK 2		0.17*	0.17*	0.26**	0.17*	0.26**	- 0.21**	0.14*	0.05
Grain yield per plant	ICC 16644 × KAK 2		0.15**	0.15**	0.15**	0.15**	0.15**	0.05	0.39**	- 0.21**
	ICC 16644 × KRIPA		0.09	0.09	0.09	0.09	0.15**	- 0.06	0.26**	- 0.12
	ICC 16644 × ICC 17109		0.03	0.03	0.03	0.09	0.03	0.03	0.18**	- 0.19**
Grain yield per plant	ICC 16644 × JGK 2		0.12	0.12	0.12	0.12	0.96**	0.96**	0.28**	0.12
	ICC 16644 × KAK 2		0.06	0.06	0.06	0.06	0.92**	0.92**	0.16**	0.06
	ICC 16644 × KRIPA		0.17*	0.17*	0.17*	0.17*	0.92**	0.92**	0.14*	0.25**
Grain yield per plant	ICC 16644 × ICC 17109		0.04	0.04	0.04	0.04	0.95**	0.95**	0.04	0.17*
	ICC 16644 × JGK 2		0.32**	0.32**	0.32**	0.32**	0.32**	0.32**	0.32**	0.27**
	ICC 16644 × KAK 2		0.51**	0.51**	0.51**	0.51**	0.51**	0.51**	0.51**	0.14*
Grain yield per plant	ICC 16644 × KRIPA		0.39**	0.39**	0.39**	0.39**	0.39**	0.39**	0.39**	0.21**
	ICC 16644 × ICC 17109		0.26**	0.26**	0.26**	0.26**	0.26**	0.26**	0.26**	0.16*
	ICC 16644 × JGK 2		0.26**	0.26**	0.26**	0.26**	0.26**	0.26**	0.26**	0.16*

Table 5 continued

Trait	Cross	Days to pod initiation	Days to maturity	Plant height	No. of seeds per plant	No. of seeds per pod	Biological yield per plant	Grain yield per plant	Harvest index	100-seed weight
Harvest index	ICC 16644 × JGK 2									0.49**
	ICC 16644 × KAK 2									0.18**
	ICC 16644 × KRIPA									0.38**
	ICC 16644 × ICC 17109									0.31**

*,**Significant at 5% and 1% level of significance respectively

days to maturity in chickpea (Malik et al. 1988; Atta et al. 2008; Sidramappa et al. 2010; Naveed et al. 2012; Jivani et al. 2013; Monpara and Dhameliya 2013; Gaur et al. 2015).

Association of phenology with 100-seed weight

The efficiency of selection for phenology and seed size mainly depends upon the direction and magnitude of association between these traits. This is particularly important for *kabuli* chickpea, where seed size is an important yield component and a significant yield determinant. Days to first flower and days to first pod formation and days to maturity exhibited significant negative association with 100-seed weight in C₁, C₂ and C₃, while for C₄ association was non-significant. While, Hovav et al. (2003) on the basis of association studies between time of flowering and 100-seed weight which were positively associated, suggested that in certain genetic backgrounds it might be difficult to breed early-flowering cultivars without compromising seed weight. Values of correlation coefficients were low revealing that early phenology might be a component of larger seed size. Seed size depends upon duration of reproductive phase, sink transfer, soil moistures condition during pod filling stage and gene involved. In earlier studies, either significant negative (Gaur et al. 2015) or no correlation (Ali et al. 2010; Jivani et al. 2013; Gaur et al. 2015) between days to flower initiation and 100-seed weight has been reported in chickpea. Thus, present and earlier studies show that in certain crosses there is scope of combining large seed size with earliness in chickpea. This is also supported by the fact that there are many large seeded *kabuli* type varieties with early maturity (Gaur et al. 2007).

Association of phenology with other traits

Phenology had no correlation with yield per se (seed yield per plant) in any of the crosses studied. These findings were corroborative with the findings of Arshad et al. (2004), Atta et al. (2008), Ali et al. (2010), Sidramappa et al. (2010), and Monpara and Dhameliya (2013). Thus there is no constraint in combining early phenology with higher grain yield in chickpea thereby allowing simultaneous selection for both traits. In general, it is difficult to improve both the yield as well as phenological traits simultaneously

through selection. According to Or et al. (1999) genotypes with early flowering alleles, may have longer reproductive period which further enhance seed yield in chickpea by allowing formation of a relatively large numbers of pods and through longer grain filling duration. Phenological traits exhibited positive and significant correlation with plant height, indicating early flowering results into shorter plant height. If onset of reproductive phase i.e. flowering is early vegetative growth is reduced which further stop the growth of branches resulting in less plant height. Phenology showed either non-significant (C_3 or C_4) or positive significant (C_1 and C_2) correlation with biological yield per plant. Non-significant association of number of seeds per plant with phenology was observed in all the crosses except C_1 where association was significant positive. These results indicate that early plants of these F_2 populations matured early and could not accumulate enough biomass (had less plant height and biomass), had lesser number of pods and seeds per plant than the late maturing plants. Results are in accordance with the findings of Gaur et al. (2015). Number of pods per plant was also studied, it had high significant positive (C_1 -0.98**, C_2 -0.97**, C_3 -0.98**, C_4 -0.97**) association with number of seeds per plant so, only correlation coefficients of number of seeds per plant with other traits are included in the Table 5. Singh et al. (1990) reported that days to flowering and days to maturity contribute to seed yield mainly via biological yield and harvest index in chickpea. Thus, reducing the growth period after a threshold level may have a penalty on grain yield. Phenology showed significant negative association with harvest index in all the crosses. These results indicate that early genotypes are more efficient in their yield partitioning and accumulated biomass necessary to ensure optimum seed yield within shorter duration possibly through a higher crop growth rate. These results encourage combining earliness with high harvest index in these crosses. High harvest index and drought escape through early flowering and early maturity are considered as important attributes of adaptation in chickpea under drought stressed environments (Berger and Turner 2004).

Association of yield and other traits

Yield per plant was significantly positively associated with plant height, number of seeds per plant, number

of pods per plant, biological yield per plant, 100-seed weight and harvest index and hence could be considered as factors for seed yield improvement. Results indicate that for higher yield, selection of genotypes with taller plant height, higher harvest index, a greater number of pods, more biological yield per plant and seeds per plant with larger seed size traits would be beneficial in these crosses. Generally, biological yield and harvest index are accepted as the most important traits for improving grain yield. Such positive inter-relationship between these attributes had also been reported in chickpea. (Arshad et al. 2004; Vaghela et al. 2009; Jivani et al. 2013). 100-seed weight had positive correlation with grain yield per plant. Mathur and Mathur (1996) and Ali et al. (2010) had similar results, while Lal et al. (1993) reported a negative correlation between seed yield and 100-seed weight. Number of pods per plant, number of seeds per plant and biological yield per plant were highly interrelated among each other. These results get support with the findings of Ali et al. (2010). Number of seeds per pod had significant negative association (C_2 and C_4) or no association with 100-seed weight indicating either a pod has a greater number of smaller seeds or a lesser number of larger seeds. In the study 100-seed weight was positively associated with harvest index and plant height but, 100-seed weight had significant negative association with number of pods and seeds per plant indicating plant with larger seed size had less pods and seeds and vice versa. So, variety with higher yield will have either a greater number of pods and seeds or will have larger seeds. Simultaneous selection for both a greater number of pods and seeds and larger seed size may not be possible. Some difficulties might be encountered in breeding larger seed cultivar without compromising number of pods and seeds per plant.

Conclusions

Early phenology is an important trait for adaptation of chickpea to different environments. In *kabuli* chickpea, the seed size is an important trait for marketing. The genetic control of seed size in *kabuli* chickpea is under two major genes exhibiting complementary epistasis and small size is dominant over large. Furthermore, two major genes with duplicate recessive epistasis control flowering time in *kabuli* chickpea where lateness is dominant over earliness. The

results of association studies suggest that phenology had significant negative association with seed size in some crosses and no association in other crosses. Thus, in certain genetic background, it would be possible to breed early flowering cultivars with large seed size by allowing simultaneous selection for both the traits. These findings will be useful to plant breeders in designing strategies to develop early maturing varieties of chickpea with large seed size.

The early maturing parent ICC 16641 used in this study has been reported to carry the early flowering gene *eft-4* (Gaur et al. 2015). Studies on allelic relationships of *eft-4* with other early flowering genes, *eft-1* (present in ICCV 2), *eft-2* (present in ICC 5810) and *eft-3* (present in BGD 132) indicated that these early flowering genes are non-allelic (Gaur et al. 2015). Availability of four different early flowering genes with similar effects provide options for choosing a specific early flowering gene based on the desired background and linkage relationships of the flowering time genes with other traits (Gaur et al. 2015). Major QTLs corresponding to flowering time genes *eft-1*, *eft-3* and *eft-4* have been mapped on CaLG04, CaLG08 and CaLG06, respectively (Mallikarjuna et al. 2017). Thus, markers identified linked to these QTLs can be used in marker-assisted breeding for developing early maturing varieties and combining early maturity trait with other desired traits, such as seed size.

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