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Association of flowering time with phenological and productivity traits in chickpea

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Abstract Phenology is an important trait for the adaption of chickpea (*Cicer arietinum* L.) to various target environments. The aim of this study was to determine the effects of flowering time on other phenological traits and yield-related traits. F_2 and F_3 segregating populations derived from the crosses of four early-flowering lines (ICCV 96029, ICC 5810, BGD 132 and ICC 16641) with a late-flowering cultivar (CDC Frontier) were used. In all crosses, flowering time showed significant positive association with days to pod initiation, days to maturity, plant height and biomass and non-significant correlation with number of pods per plant, number of seeds per plant and grain yield per plant. Flowering time had a positive correlation with 100-seed weight in all crosses, with the exception of ICC 16641 × CDC Frontier where the correlation was non-significant. Harvest index was negatively associated with flowering time. In most of the crosses, early- and late-maturing F_3 bulks showed significant differences with respect to biomass and harvest index, while for grain yield and 100-seed weight the differences were found to be non-significant. These results indicate that flowering time could be used as a reliable selection criterion in breeding for early-maturing chickpea and that a reduction in the duration of flowering time and maturity may not necessarily have a yield penalty in these genetic backgrounds.

Keywords *Cicer arietinum* · Correlation coefficient · Early maturity · Grain yield

Introduction

Chickpea (*Cicer arietinum* L.; $2n = 2x = 16$) is the second most important food legume globally and a staple protein crop in the Indian subcontinent. It is cultivated in more than 50 countries worldwide, over an area of 12.6 million hectares, with a total production of 12.1 million tonnes and average yield of 956 kg/ha (FAOSTAT 2016). Being a highly nutritious food legume (Jukanti et al. 2012), chickpea also improves soil nutritional status by fixing atmospheric nitrogen through symbiotic nitrogen fixation. Chickpea is traditionally a low-input crop, and about 80% of the world's chickpea crop is grown in the areas relying on conserved soil moisture conditions where the crop often experiences terminal drought stress (Gaur et al. 2008b).

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Based on research undertaken in India and other countries, early phenology (time to flowering, podding and maturity) has been identified as a key trait for the adaption of chickpea to different growing environments (Kumar and Abbo 2001; Berger et al. 2006; Gaur et al. 2008a, b, 2018). Early maturity helps the crop escape end-of-season stresses, such as drought (Subbarao et al. 1995) and frost (Anbessa et al. 2006), thereby increasing and stabilizing chickpea yields in short-season environments. Consequently, breeding for early maturity has been one of the major chickpea breeding objectives in recent years. The duration of crop maturity is the end result of several phenological and morphological variables, which are interrelated and could be manipulated separately. Breeders generally have used days to flowering as a key indicator of maturity duration since this trait provides a good indication of subsequent phenological traits, such as time of podding and maturity in chickpea (Gaur et al. 2015).

To date, major genes controlling flowering time have been reported in chickpea lines ICCV 96029 (Kumar and van Rheenen 2000), ICC 5810 (Or et al. 1999), BGD 132 (Hegde 2010) and ICC 16641 (Gaur et al. 2015), and the corresponding genomic regions have been recently identified (Mallikarjuna et al. 2017). This simple genetic basis of major flowering time genes facilitates introgression into any popular late-flowering genetic background by simple backcrossing. However, breeding programs aimed at early maturity should also consider other important agronomic traits to exploit additional gains (Hovav et al. 2003). Also, it would be of interest to know the association of these early-flowering genes with other phenological traits as well as with other component traits of productivity. Such information will be useful to breeders in developing early maturing varieties with other desired traits. Therefore, the aim of this study was to examine the relationships of flowering time with other phenological and yield-related traits in four chickpea crosses.

Materials and methods

Experimental material

The study described herein was conducted at the International Crops Research Institute for Semi-Arid Tropics (ICRISAT), Patancheru, India. The experimental material was developed by crossing four early-flowering lines, namely, ICCV 96029, ICC 5810, BGD 132 and ICC 16641 (for detailed characteristics, see Gaur et al. 2015) with a late-flowering cultivar, CDC Frontier (Warkentin et al. 2005). A total of 190 F_2 plants in each of the three crosses, ICCV 96029 × CDC Frontier, ICC 5810 × CDC Frontier and BGD 132 × CDC Frontier, and 146 F_2 plants in the cross ICC 16641 × CDC Frontier were evaluated during the post-rainy season of 2013–2014 along with their parents and F_1s. Observations were recorded on each plant on flowering time, days to pod initiation, days to maturity, plant height, number of pods per plant, number of seeds per plant, grain yield per plant, 100-seed weight, biomass per plant and harvest index. Simple correlation coefficients between flowering time and other traits were calculated using Microsoft Excel 2013 (Microsoft Corp., Redmond, WA, USA).

During the crop season 2014–2015, a total of 164, 174, 182 and 102 F_3 progenies from the crosses ICCV 96029 × CDC Frontier, ICC 5810 × CDC Frontier, BGD132 × CDC Frontier and ICC 16641 × CDC Frontier, respectively, were raised. Each F_3 progeny row was observed for flowering time at regular intervals and classified as non-segregating (uniform early or late flowering) and segregating types. Observations such as flowering time, days to maturity, grain yield, biomass, 100-seed weight and harvest index (calculated) were recorded on 1-m continuous rows of uniform early- and late-flowering segregants. Statistical procedures, including descriptive statistics and student *t* test, were performed to compare the means of early- and late-flowering groups using Microsoft Excel 2013.

Results and discussion

Mean performance of parental lines for flowering time and other important traits

The female parents, i.e. ICCV 96029, ICC 5810, BGD 132 and ICC 16641, took 27, 28, 29 and 29 days to flowering, 33, 38, 35 and 35 days to pod initiation and 76, 77, 79 and 79 days to maturity after sowing, respectively (Table 1). In comparison, the male parent CDC Frontier was very late in days to flowering (67 days), pod initiation (72 days) and maturity (108 days). The mean plant height among the early-flowering lines

Table 1 Mean performance of parental lines during post rainy season of 2013–2014

Chickpea line	Flowering time	Days to pod initiation	Days to maturity	Plant height (cm)	No. of pods per plant	No. of seeds per plant	Grain yield per plant (g)	Biomass per plant (g)	100 seed weight (g)	Harvest index (%)
ICCV 96029										
Mean ± SE	27 ± 0.21	33 ± 0.42	76 ± 0.30	43.13 ± 0.54	113 ± 1.46	131 ± 2.2	15.55 ± 0.49	26 ± 0.91	12.02 ± 0.2	60.17 ± 0.8
Range	25–28	30–38	75–81	37–49	100–129	116–155	12.04–23.43	17.43–40.6	9.86–14.86	50.9–69.88
ICC 5810										
Mean ± SE	28 ± 0.15	38 ± 0.19	77 ± 0.16	52.83 ± 0.58	97 ± 1.67	126 ± 1.87	16.62 ± 0.51	31.01 ± 0.98	13.25 ± 0.27	53.75 ± 0.78
Range	28–30	36–41	77–80	48–61	85–120	112–157	12.9–22.2	24–44.82	11.43–17.76	43.48–62.5
BGD 132										
Mean ± SE	29 ± 0.11	35 ± 0.09	79 ± 0.23	43.1 ± 0.86	61 ± 1.66	66 ± 1.63	18.1 ± 0.62	29.08 ± 1.12	27.56 ± 0.52	62.63 ± 0.7
Range	28–29	34–35	78–83	37–55	43–76	51–80	13.75–25.76	21.1–42.2	21.91–32.92	56.03–68.26
ICC 16641										
Mean ± SE	29 ± 0.00	35 ± 0.18	79 ± 0.00	48.63 ± 0.89	75 ± 1.82	96 ± 1.55	22.26 ± 1.17	34.4 ± 1.83	22.92 ± 0.39	64.88 ± 0.73
Range	29–29	34–38	79–79	40–58	60–97	84–114	15.16–38.22	22.6–64.3	17.01–26.09	52.17–68.63
CDC Frontier										
Mean ± SE	67 ± 0.25	72 ± 0.74	108 ± 0.59	63.97 ± 0.71	38 ± 1.33	41 ± 1.82	13.31 ± 1.36	45.51 ± 3.33	31.62 ± 0.4	29.15 ± 1.7
Range	65–68	67–79	104–112	56–71	24–55	22–65	4.76–27.3	14.1–85.85	26.89–34.6	11.08–46.2

SE Standard error

ranged from 43.1 (ICCV 96029) to 52.8 cm (ICC 5810), whereas mean recorded plant height of CDC Frontier was 64.0 cm. Among the early-flowering lines, the highest number of pods per plant (113.3) and highest number of seeds per plant (130.6) were recorded for ICCV 96029, while BGD 132 was found to have the lowest number of pods per plant (60.9) and lowest number of seeds per plant (66.4). For CDC Frontier, the mean number of pods per plant was 37.9 and the mean number of seeds per plant was 41.8. Among the early-flowering lines, ICC 16641 was found to have the highest grain yield per plant (22.3 g) and highest biomass per plant (34.4 g), while ICCV 96029 had the lowest grain yield per plant (15.6 g) and lowest biomass per plant (26 g). The late-flowering line CDC Frontier recorded grain yield per plant of 13.3 g with 45.5 g biomass per plant. Hundred seed weight among the early-flowering lines ranged from 12.0 (ICCV 96029) to 27.6 g (BGD 132), while that of the late-flowering line CDC Frontier was 31.62 g. The harvest index of the early-flowering lines ranged from 53.8 (ICC 5810) to 64.9% (BGD 132), while that of CDC Frontier was 29.2%. These results clearly indicate that there is significant difference between the early- and late-flowering lines with respect to phenology and other traits.

Association between flowering time and other agronomic traits

The data collected on individual F_2 plants were used to estimate correlation coefficients between flowering time and other agronomic traits in all the crosses (Table 2). Flowering time exhibited a positive significant correlation with days to pod initiation in all crosses (ICCV 96029 × CDC Frontier, $r = 0.99$; ICC 5810 × CDC Frontier, $r = 0.99$; BGD 132 × CDC Frontier, $r = 0.99$; ICC 16641 × CDC Frontier, $r = 0.99$), suggesting that early flowering leads to early podding in these crosses. Also in all crosses, flowering time and days to maturity exhibited highly significant positive correlation ($r = 0.88$, $r = 0.89$, $r = 0.93$ and $r = 0.95$, respectively). These results indicate that in general early-flowering lines also mature early. It appears that flowering time influenced maturity duration in chickpea mainly through its effect on days to pod initiation. Therefore, flowering time would appear to be the more precise trait to record than days to maturity to discriminate between early and late genotypes under conditions where the recording of maturity is influenced by environmental factors such as available soil moisture and temperature. However, effective manipulation of final maturity duration would best be achieved by selecting for more than one component of crop duration (Anbessa et al. 2006).

Flowering time showed significant positive correlation with plant height in all crosses (ICCV 96029 × CDC Frontier, $r = 0.51$; CC 5810 × CDC Frontier, $r = 0.19$; BGD 132 × CDC Frontier, $r = 0.45$; ICC 16641 × CDC Frontier, $r = 0.47$) and biomass ($r = 0.26$, $r = 0.18$, $r = 0.33$ and $r = 0.28$). A similar type of correlation has been

Table 2 Correlation coefficients between flowering time and other important traits observed in different F_2 populations

Cross	Days to pod initiation	Days to maturity	Plant height	Pods per plant	Seeds per plant	Biomass per plant (g)	Grain yield per plant (g)	100 Seed weight (g)	Harvest index
ICCV 96029 × CDC Frontier	0.99**	0.88**	0.51**	− 0.03	− 0.05	0.26**	0.05	0.28**	− 0.43**
ICC 5810 × CDC Frontier	0.99**	0.89**	0.19**	− 0.09	− 0.13	0.18*	− 0.01	0.31**	− 0.44**
BGD 132 × CDC Frontier	0.99**	0.93**	0.45**	0.13	0.09	0.33**	0.13	0.24**	− 0.49**
ICC 16641 × CDC Frontier	0.99**	0.95**	0.47**	− 0.05	− 0.09	0.28*	− 0.09	− 0.06	− 0.62**

*Correlation is significant at the 0.05 level; **correlation is significant at the 0.01 level reported earlier in chickpea (Bonfil et al. 2006), pea and lentil (Slinkard and Sindhu 1988; Erskine et al. 2000). The nature of this association would appear to be that late-flowering genotypes had more time for vegetative growth (growing taller and accumulating higher biomass) compared to extra-early and early plants.

A non-significant correlation was found in all crosses for flowering time with number of pods per plant, number of seeds per plant and grain yield per plant. Several causes for the lack of a simple relationship between these traits are possible. One underlying cause could be that the common male parent in all crosses, CDC Frontier, is a very late-maturing line but a poor yielder under the growing conditions of Patancheru compared to all of the extra-early female lines (Table 1). These results suggest that selection for early flowering (to a certain extent) need not necessarily involve a severe yield penalty in early-maturing genotypes as the possibility of combining components of earliness with yield-promoting alleles has been demonstrated in *desi* chickpea (Siddique and Khan 1996).

A weak positive correlation was observed between flowering time and 100-seed weight in the crosses ICCV 96029 × CDC Frontier ($r = 0.28$), ICC 5810 × CDC Frontier ($r = 0.31$) and BGD 132 × CDC Frontier ($r = 0.24$), whereas, a non-significant correlation was observed in ICC 16641 × CDC Frontier. A significant and positive correlation between time to flowering and seed weight was also reported by Hovav et al. (2003), suggesting that in certain genetic backgrounds it might be difficult to breed early-flowering cultivars without compromising seed size. On the other hand, Gaur et al. (2015) observed a non-significant correlation between flowering time and mean seed weight in early-flowering segregants in the crosses where ICCV 96029 (*efl-1*), ICC 5810 (*efl-2*), BGD 132 (*efl-3*) and ICC 16641 (*efl-4*) were used as parents. These findings highlight the notion that under certain genetic backgrounds, there is scope for combining earliness with large seed size in chickpea.

Flowering time exhibited a significant negative correlation with harvest index in all the crosses (ICCV 96029 × CDC Frontier, $r = -0.43$; ICC 5810 × CDC Frontier, $r = -0.44$; BGD 132 × CDC Frontier, $r = -0.49$; ICC 16641 × CDC Frontier, $r = -0.62$). This negative correlation indicates that the extra-early and early genotypes were more efficient in partitioning their yield and accumulating the biomass necessary to ensure optimum seed yield within a shorter duration, possibly through a higher crop growth rate and growth vigor. The genomic regions identified for plant vigor was found to be the quantitative trait locus hotspot contributing drought tolerance in chickpea (Sivasakthi et al. 2018). These characteristics would be useful for stabilizing yield under short-season environments. The results are consistent with the widely believed hypothesis that early maturity is associated with a high harvest index (Wallace et al. 1993; Anbessa et al. 2007). A high harvest index and drought escape through early flowering and early maturity are considered to be important attributes of adaptation in chickpea under environments prone to drought stress (Berger et al. 2004).

Effect of flowering time on productivity traits

To quantify the effect of flowering time on maturity, grain yield, biomass, harvest index and seed size, we performed *t* tests between the early- and late-flowering F_3 progenies of all the crosses (Table 3; Fig. 1). In the crosses ICCV 96029 × CDC Frontier, ICC 5810 × CDC Frontier, BGD 132 × CDC Frontier and ICC 16641 × CDC Frontier, the mean flowering time of early-flowering progenies (36, 41, 31 and 33 days, respectively) differed significantly with those of the late-flowering progenies (56, 60, 56 and 65 days, respectively). Similarly, F_3 progenies of these crosses also differed significantly for mean days to maturity, with 87, 90, 81 and 83 days for the respectively crosses among the early bulks and 107, 109, 103 and 109 days for the respectively crosses among the late bulks. These results provide evidence that flowering time had a positive effect on maturity, suggesting that selection for the early-flowering trait will be appropriate for developing early-maturing lines. Significant differences were found between the two bulks with respect to biomass in all of the crosses except ICC 5810 × CDC Frontier, where the difference was statistically non-significant, with the mean biomass of early- and late-flowering progenies being 391 and 443 g, respectively. The difference in grain yield of the early-flowering progenies was statistically non-significant from that of the late-flowering bulks in all crosses except BGD 132 × CDC Frontier, where the

Table 3 Comparison of early-and late-flowering F_3 progenies for phenology and agronomical traits in different crosses

Cross	Phenotypic class	Flowering time	Days to maturity	Biomass (g)	Grain yield (g)	Harvest index (%)	100 Seed weight (g)
ICCV 96029 × CDC Frontier	Early						
	Mean	36	87	394.36	241.3	61	21.85
	Range	30–40	80–96	311–530	194.48–309.93	50–67	14.87–29.86
	Late						
	Mean	56	107	587.51	270.62	46	21.29
	Range	48–66	96–116	466.25–801.42	198.38–334.67	37–57	14.2–31.78
	Probability (*t* test)	< 0.005*	< 0.005*	< 0.005*	0.02 ns	< 0.005*	0.65 ns
ICC 5810 × CDC Frontier	Early						
	Mean	41	90	391.02	209.12	53	21.77
	Range	34–44	80–96	260–502.5	132.58–288.36	44–59	16.15–30.76
	Late						
	Mean	60	109	443.28	220.50	43	25.13
	Range	54–64	101–115	322.5–564.28	161.13–284.7	34–54	20.03–32.48
	Probability (*t* test)	< 0.005*	< 0.005*	0.17 ns	0.46 ns	< 0.005*	0.13 ns
BGD 132 × CDC Frontier	Early						
	Mean	31	81	350.38	182.11	52	34.24
	Range	30–35	78–94	187.5–466.66	102.91–286.28	25–64	28.62–38.87
	Late						
	Mean	56	103	483.5	229.45	48	32.27
	Range	42–66	93–116	318–596	151.56–287.95	31–64	25.23–39.1
	Probability (*t* test)	< 0.005*	< 0.005*	< 0.005*	< 0.005*	0.17 ns	0.04 ns
ICC 16641 × CDC Frontier	Early						
	Mean	33	83	275.69	170.46	61	29.48
	Range	31–35	82–88	177.5–367.77	110.87–234.13	52–66	20.66–36.42
	Late						
	Mean	65	109	485.54	190.98	37	30.56
	Range	41–73	101–119	302–580	107.25–319.35	21–55	23.24–37.5
	Probability (*t* test)	< 0.005*	< 0.005*	< 0.005*	.018 ns	< 0.005*	0.44 ns

*Significant difference between phenotypic class at the 0.05 level; ns, not significant

mean grain yield in early- and late-flowering bulks was 182.11 and 229.45 g, respectively, however the maximum range of grain yield produced by early- and late-flowering bulks was 286.28 and 287.95 g, respectively. These results indicate that given the duration of the crop, early-flowering bulks were more efficient in their yield partitioning potential, which is expressed by their higher harvest indices compared to those of the late bulks of all the crosses (Table 3). This result suggests that selection for early flowering in these genetic backgrounds need not necessarily involve a severe reduction in grain yield compared to late flowering. This trait is particularly important under short season environments where growing early varieties can stabilize yield when the crop is more prone to suffer terminal stresses. A non-significant difference between 100-seed weight of early- and late-flowering bulks was observed in all of the crosses, indicating that selection for early flowering does not pose any negative effect on seed size.

Flowering time is considered to be an important adaptive trait to various target environments. The

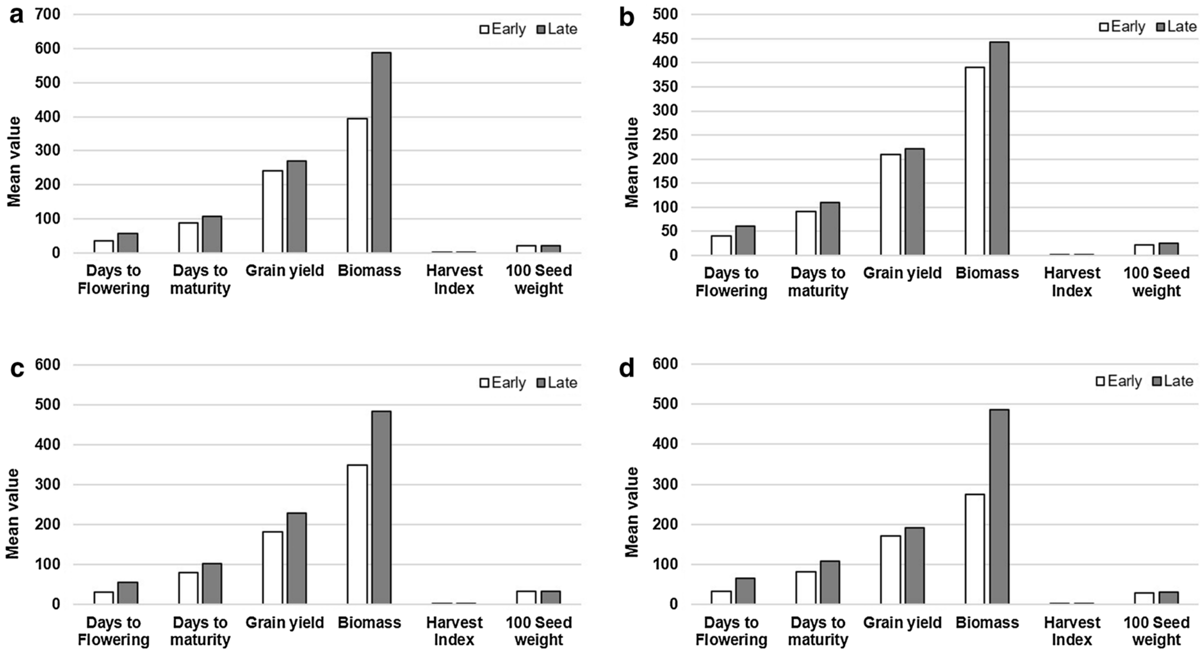

Fig. 1 Comparison between early-and late-flowering F_3 segregants for phenological and yield-related traits in different crosses. **a** ICCV 96029 × CDC Frontier, **b** ICC 5810 × CDC Frontier, **c** BGD 132 × CDC Frontier, **d** ICC 16641 × CDC Frontier

results of this study show that the early-flowering trait in suitable genetic backgrounds can produce high-yielding genotypes similar to that of late-flowering or maturing genotypes under terminal drought stress conditions. This knowledge will help breeders to develop early-maturing varieties with a high yield potential for short season environments.

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Author's contributions PMG coordinated this project. PMG, KPV, SS guided BPM in planning and designing this study. BPM, PMG and SS were involved in developing the mapping populations, designing field experimentation, phenotyping and data analysis. BPM, PMG, and SS drafted the manuscript, and all the authors reviewed and approved the manuscript.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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