Photoperiod Response of Annual Wild *Cicer* Species and Cultivated Chickpea on Phenology, Growth, and Yield Traits

Shivali Sharma⋆ and Hari D. Upadhyaya

ABSTRACT

Frequent utilization of wild *Cicer* species in chickpea (*Cicer arietinum* L.) improvement programs, as well as the regeneration of these wild species for efficient conservation in genebanks, is hindered due to photoperiod and/or temperature sensitivity (vernalization). In this study, the response to four extended photoperiod treatments (15, 18, 21, and 24 h) was compared with a control (12 h) for phenology and growth in terms of reduction in number of days to first flowering, as well as for yield-related traits in cultivated chickpea and seven annual wild *Cicer* species. The study revealed that wild *Cicer* species required long photoperiods (varying from 15 to 18 h) for transition from the vegetative to reproductive phase. Optimum photoperiods also improved agronomic traits such as pod number and seed yield per plant. Of the photoperiods studied, 18 h was the most appropriate photoperiod treatment for both reducing the vegetative phase and for efficient regeneration in *C. reticulatum* Ladiz. Fifteen hours was the most appropriate photoperiod in *C. judaicum* Boiss. and *C. yamashitae* Kitamura. Both 15 and 18 h were the most appropriate photoperiods in *C. bijugum* K.H. Rech. and *C. pinnatifidum* Jaub. & Sp., depending on the objective (15 h for regeneration and 18 h for reducing vegetative phase). *Cicer chorassanicum* (Bunge) M. Pop. and *C. cuneatum* Hochst. ex A. Rich. showed a weak response to all the extended photoperiod treatments. These results contribute to enhanced utilization of wild *Cicer* species for chickpea improvement through synchronization of flowering facilitating hybridization and for efficient regeneration by using species-specific extended photoperiod treatments.

Cʜɪᴄᴋᴘᴇᴀ (*Cicer arietinum* L.) is an important grain legume crop worldwide and is cultivated for its protein rich seeds in ∼82 countries on 12.65 million ha with production of 12.09 Tg and productivity of 956 kg ha^{-1} (FAOSTAT, 2018). The crop is affected by a number of biotic (pod borer [*Helicoverpa armigera* (Hübner)], Fusarium wilt [*Fusarium oxysporum* Schlechtend.], botrytis gray mold [*Botrytis cinerea* Pers.:Fr.], dry root rot [*Macrophomina phaseolina* (Tassi) Goidanich], Ascochyta blight [*Ascochyta rabiei* (Pass.) Labr.]), and abiotic (drought, heat and cold) stresses resulting in low productivity. Chickpea has a narrow genetic base that hinders its genetic improvement by using cultivated genepools, especially for high levels of resistance against important biotic and/or abiotic stresses. In contrast, the genus *Cicer*, containing 43 wild species comprising eight annual and 35 perennial species, provides sufficient genetic variability for use in chickpea improvement programs. Although the importance of wild species for crop improvement is well known, very few attempts have been made to introgress important traits from wild *Cicer* species into cultivated backgrounds (Jaiswal et al., 1986; Singh and Ocampo, 1997; Malhotra et al., 2002; Singh et al., 2005; Knights et al., 2008; Upadhyaya, 2008).

As chickpea is an annual crop, annual wild *Cicer* species are of particular interest to breeders for introgressing useful alleles into cultivated backgrounds. However, the difference in phenology of cultivated and wild species (Summerfield et al., 1989; Robertson et al., 1997; Abbo et al., 2002; Berger et al., 2005) poses serious concern for their utilization in hybridization programs. Chickpea is adapted to diverse environments in tropical, subtropical, and

warm-temperate zones, whereas annual wild *Cicer* species are mostly found in western and central Asia above 34.5° N, as well as along the coastal eastern Mediterranean and in isolated populations adjacent to the African Red Sea coast (Berger et al., 2003), and exhibit vernalization and photoperiod response (Sharma and Upadhyaya, 2015). In subtropical regions such as southern India, the vernalization and photoperiod requirements of wild *Cicer* species are not fulfilled under natural field conditions. Under such environments, wild *Cicer* species are generally late in phenology and therefore cannot be used frequently in crossing programs.

Studies have shown the response of wild *Cicer* species to vernalization (Abbo et al., 2002; Berger et al., 2005; Sharma and Upadhyaya, 2015) and extended photoperiod (Sethi et al., 1981; Sharma and Upadhyaya, 2015). Our recent studies revealed that annual wild *Cicer* species are responsive both to vernalization and 24-h extended photoperiod treatments. However, compared with vernalization, the response of most of these species to 24-h photoperiod treatment was greater (Sharma and Upadhyaya, 2015). In a similar study, long-day (16/8 h day/night) and short-day (10/14 h day/night) photoperiods were used to study the flowering response in cultivated chickpea accessions (Daba et al., 2016). Nevertheless, none of the studies was aimed at determining the critical daylength required by the wild *Cicer* species for flowering. The present investigation is the first attempt to study the response of annual wild *Cicer* species and cultivated chickpea to different photoperiod treatments on days to first flowering and yield-contributing traits with an aim to appraise critical daylength for desired days to first flowering.

MATERIALS AND METHODS
Plant Material
Germplasm accessions of seven annual wild *Cicer* species [*C. reticulatum* Ladiz., *C. judaicum* Boiss., *C. bijugum* K.H. Rech., *C. pinnatifidum* Jaub. & Sp., *C. chorassanicum* (Bunge) M. Pop., *C. cuneatum* Hochst. ex A. Rich., and *C. yamashitae* Kitamura] and cultivated *C. arietinum* were used in this investigation. These germplasm accessions were collected or originated from Afghanistan, Ethiopia, Syria, and Turkey (Table 1). *Cicer arietinum* was represented by late-maturing desi-type chickpea variety G 130, cultivated in India (Singh, 1987) (Table 1).

Methodology
The study was performed under controlled greenhouse conditions maintained at 25°C in 2012 and 2013. In both years, all eight *Cicer* species were evaluated under five photoperiods by extending the natural daylength artificially to 15, 18, 21, and 24 h. Natural daylength of 11 to 12 h of exposure at 25°C was the control.

To initiate germination, seeds of wild *Cicer* species were scarified by incising the hard seed coat. Scarified seeds of wild *Cicer* accessions and nonscarified seeds of *C. arietinum* 'G 130' were placed on wet filter paper in Petri dishes for germination at

Table 1. List of annual wild and cultivated *Cicer* species used in study at ICRISAT, Patancheru, India.

Species	Accession identity	Country of origin or pedigree
C. reticulatum	ICC 17123	Turkey
C. judaicum	ICC 17188	Syria
C. pinnatifidum	ICC 17126	Turkey
C. bijugum	ICC 17289	Turkey
C. yamashitae	ICC 17117	Afghanistan
C. chorassanicum	ICC 17141	Afghanistan
C. cuneatum	ICC 20176	Ethiopia
C. arietinum	G 130 (Desi late)	India/708 × C 235

room temperature for 3 d. After 3 d, the germinated seedlings were transplanted in pots (one seedling per pot) containing a 2:1:1 mixture of black soil, farmyard manure, and sand. Each light exposure treatment was performed in separate chambers in the greenhouse maintained at 25°C. In each chamber, the natural daylength was extended by using 60-W incandescent lights 15 d after transplanting through maturity (Sethi et al., 1981). Each of the eight species were randomly allotted to three pots, with one plant per pot, and kept under each chamber. These 24 pots were randomized within each chamber. For the control treatment, seedlings were transplanted in pots and maintained under natural daylength (11–12 h) at 25°C in the greenhouse. In both years, data were recorded for each plant on number of days to first flowering starting from the day of transplanting, and yield-contributing traits such as plant height, canopy width, number of pods per plant, number of seeds per plant, seed weight per plant, and 100-seed weight.

Statistical Methods
Response of wild and cultivated *Cicer* species to different treatments was based on a reduction in number of days to first flowering under different treatments compared to the control. To assess variation due to accessions and interactions between accession and extended photoperiods and years, ANOVA was performed on the data where accession effects were modeled under the completely randomized design and combined over the photoperiods, incorporating the interaction between accession and photoperiod. The means for accessions and their combinations with the photoperiods were obtained for each response variable, along with their SEs. Significant differences between species were detected by comparing means with respective LSDs. The patterns of differential responses of accessions to treatments were estimated by partitioning accession × treatment interaction in polynomial components in exposure length. For each species, the relationship of days to first flowering with length of photoperiod was evaluated by fitting linear, quadratic, and cubic regressions, selecting the best of the three regressions that accounted for the highest percentage variance, and estimating regression coefficients for the best regression. There are other types of nonlinear curves that could support days to first flowering and photoperiod relationships and are available in Genstat software (VSN International, 2015, p. 312–321). Left exponential, exponential, and linear divided by linear curves were tested and the one that accounted for the highest percentage variance was chosen. Although many of these forms can be used for estimating days to first flowering for a given photoperiod, some,

such as an exponential curve, could be more suited for estimation of daylength required to achieve a given number of days to first flowering. The exponential form, expressed as days to first flowering = $A + BR^h$, was used for estimating days to first flowering for a given photoperiod (h). The parameter R ($R < 1$) is an indicator of nonlinear rate of decline in h, whereas B is the slope or linear decline with the nonlinear variable R^h. The parameter A measures the potential as an asymptotic value of days to first flowering that can be achieved after an infinite or practically the longest possible exposure. Thus, days to first flowering would vary between A and $A + B$. Such an equation was fitted using the FITCURVE directive in Genstat software. The exposure times required to achieve a low value of days to first flowering expressed as $10\% > A$ were also estimated. Standard errors of estimated hours were determined using the results for inverse estimation for a general nonlinear function given in Singh et al. (1992). Genstat statistical software (VSN International, 2015) was used for all the calculations.

RESULTS

Year–wise and pooled analysis showed significant differences among species and photoperiod treatment × species interaction ($p \leq 0.001$) for all the traits. Pooled analysis (Table 2) indicated that different photoperiod treatments significantly affected days to first flowering and 100-seed weight ($p \leq 0.001$) with significant linear and quadratic trends in treatments; plant height ($p \leq 0.05$) with a significant linear trend; number of pods and seeds per plant ($p \leq 0.001$) with significant quadratic and cubic trends and deviations; and seed weight per plant ($p \leq 0.05$) with significant quadratic and cubic trends. Further, partitioning of the total sum of squares showed that different photoperiod treatments explained ~58% of the variation in days to first flowering, with the linear trend being much stronger (78% variation) than the quadratic trend (21% variation) (Table 2). However, in treatment × species interaction, the linear trend was much stronger (51% variation) than the quadratic trend (24%) for days to first flowering. Similarly, the linear trend was most important in explaining variation (~66%) for plant height, linear (40%), cubic (28%) and quadratic (22%) trends for canopy width, cubic (~40%) and linear (24%) trends for number of pods and seeds per plant, linear (37%) and cubic (30%) trends for seed weight per plant, and linear (49%) and quadratic (31%) trends for 100-seed weight (Table 2). Although deviations from cubic polynomials were found to be statistically significant, the cubic polynomials in daylength explained a large part of variability, exceeding 89% for days to first flowering, plant height, 100-seed weight, and canopy width, and exceeding 80% for number of pods and seeds per plant, implying that response to the exposure time can be well described by cubic polynomials.

Response to Treatments

All the treatments reduced the number of days to first flowering compared with the control (12 d to first flowering,

Table 2. Pooled ANOVA for days to first flowering and other yield-related traits in annual wild and cultivated *Cicer* species under different treatments at ICRISAT, Patancheru, India.

Source of variation	df	Mean squares†														
		DTFF	F pr	PLHT	F pr	CAWD	F pr	PPP	F pr	SPP	F pr	SWPP	F pr	HSW	F pr
Year × treatment interaction															
Year	1	1.2	0.965	12,745.8	0.002	1.4	0.936	3,666.0	0.246	1,505.0	0.568	25.5	0.045	28.7	0.002
Treatments	4	44,326.9	<0.001	2,659.0	0.019	537.1	0.173	122,545.0	<0.001	185,007.0	0.001	115.8	0.002	25.0	0.001
Linear	1	138,686.0	<0.001	9,056.7	<0.001	1,582.3	0.003	2,896.0	0.294	224.0	0.822	0.3	0.779	60.6	<0.001
Quadratic	1	37,068.6	0.001	611.4	0.001	353.7	0.046	183,249.0	<0.001	295,848.0	<0.001	252.3	<0.001	33.9	0.001
Cubic	1	1,026.7	0.243	866.7	0.001	31.5	0.247	241,114.0	<0.001	376,096.0	<0.001	204.1	<0.001	3.8	0.059
Deviations	1	526.5	0.383	101.2	0.547	181.1	0.127	62,920.0	0.005	67,860.0	0.014	6.7	0.213	1.7	0.155
Residual	4	549.3		234.6		193.1		1,993.0		3,904.0		3.1		0.6	
Year × treatment × replication × species interaction															
Species	7	5,159.9	<0.001	49,262.9	<0.001	122.0	<0.001	169,354.0	<0.001	405,140.0	<0.001	230.4	<0.001	958.2	<0.001
Year × species	7	532.6	<0.001	5,166.1	<0.001	56.7	<0.001	30,791.0	<0.001	25,976.0	<0.001	18.1	<0.001	5.4	<0.001
Treatment × species	28	2,669.0	<0.001	1,106.0	<0.001	41.3	<0.001	45,985.0	<0.001	95,084.0	<0.001	41.8	<0.001	6.4	<0.001
Linear × species	7	5,394.6	<0.001	2,911.0	<0.001	66.6	<0.001	44,717.0	<0.001	96,436.0	<0.001	61.3	<0.001	12.5	<0.001
Quadratic × species	7	2,532.4	<0.001	628.3	<0.001	35.8	0.002	29,357.0	0.002	54,336.0	<0.001	16.7	<0.001	7.9	<0.001
Cubic × species	7	1,748.8	<0.001	626.4	<0.001	46.8	<0.001	75,790.0	<0.001	153,035.0	<0.001	50.7	<0.001	2.4	0.006
Deviations	7	1,000.1	<0.001	258.4	<0.001	16.1	0.159	34,078.0	<0.001	76,529.0	<0.001	38.3	<0.001	2.8	0.002
Residual	188	77.6		74.4		10.5		2,308.0		3,489.0		2.6		0.8	
Total	239														

† DTFF, days to first flowering; F pr, F probability; PLHT, plant height; CAWD, canopy width; PPP, pods per plant; SPP, seeds per plant; SWPP, seed weight per plant; HSW, 100-seed weight.

Fig. 1). Overall, the three highest exposure treatments, 24, 21, and 18 h were similar and significantly better than 15 h at reducing the number of days to first flowering in wild and cultivated *Cicer* species. On average, the number of days to first flowering varied from 41 to 45 d under these three treatments, 70 d under 15 h and 111 d for the control. Therefore, the number of days to first flowering was reduced by 60 to 63% under three treatments (24, 21, and 18 h), followed by a 37% reduction under 15 h compared with the control. Similar patterns were observed in two groups separately: in wild *Cicer* species, where days to first flowering were reduced by ~61 to 65% under 24, 21, and 18 h, followed by a ~38% reduction under 15 h, and in cultivated chickpea cultivar G 130, where days to first flowering was reduced by ~44% under 24 h, ~42% under 21 h, ~39% under 18 h, and 28% under 15 h. Further, the effect of these treatments on days to first flowering varied significantly between *Cicer* species, and different responses were observed in different species (Fig. 1).

Species Response

All four treatments were similarly effective in reducing the number of days to first flowering in tertiary genepool species, *C. judaicum* and *C. yamashitae* (~70–77% reduction under 21, 24, 18, and 15 h) and cultivated chickpea compared with the control (Fig. 1). The reduction in number of days to first flowering observed at 18, 21, and 24 h was similar but significantly greater than the reduction observed in the 15-h treatment in the primary gene pool species *C. reticulatum* and tertiary gene pool species *C. pinnatifidum* and *C. bijugum* (Fig. 1). Two treatments, 24 and 21 h, were similar and significantly better than other treatments in reducing the number of days to first flowering in tertiary genepool species *C. cuneatum* (~38–39% reduction under 21 and 24 h, ~23% under 18 h, and ~16% under 15

h) and *C. chorassanicum* (~22–27% reduction under 21 and 24 h, ~16% under 18 h, and ~10% under 15 h) (Fig. 1).

The regression models for days to first flowering in terms of photoperiods are given in Table 3. The best model that accounted for the highest percent variance was quadratic for three species (*C. reticulatum*, *C. pinnatifidum*, and *C. cuneatum*) and cubic for the other species. Not only did the relationship of days to first flowering and photoperiod vary with the species in terms of the coefficients, but also in its form. These equations (Table 3) can be used to estimate days to first flowering for a given photoperiod.

Although the polynomial equations described the response well (Table 3), these can also be used to predict the response within the observed photoperiod. However, estimation of the daylength from polynomials may result in invalid or multiple estimates. Examination of the days to first flowering and photoperiod graphs supported the fitting of an exponential curve (days to first flowering $= A + BR^h$; Fig. 2), and the estimates of A, B, and R parameters are given in Table 4, along with estimate of exposure $h = \log[(\text{days to first flowering} - A)/B]/\log(R)$ when days to first flowering $= 1.1A$ (10% above the lower limit). The relationship between days to first flowering and exposure hours varied with species (Fig. 2, Table 4). The exponential decline rate (R) in days to first flowering with exposure was slowest for *C. yamashitate* followed by *C. judaicum*. The lowest potential flowering earliness that can be reached among the species studied among the species studied is in *C. reticulatum* ($A = 6.2$ d), followed by *C. pinnatifidum* ($A = 10.3$ d). The higher side of potential earliness lies in *C. cuneatum* ($A = 44.9$ d). The exposure hours for days to first flowering to reach within 10% of the potential was found to go beyond the experimental period of 24 h for the four species *C. reticulatum*, *C. chorassanicum*, *C. pinnatifidum*, and *C. cuneatum* (exceeding 36.6 h), where
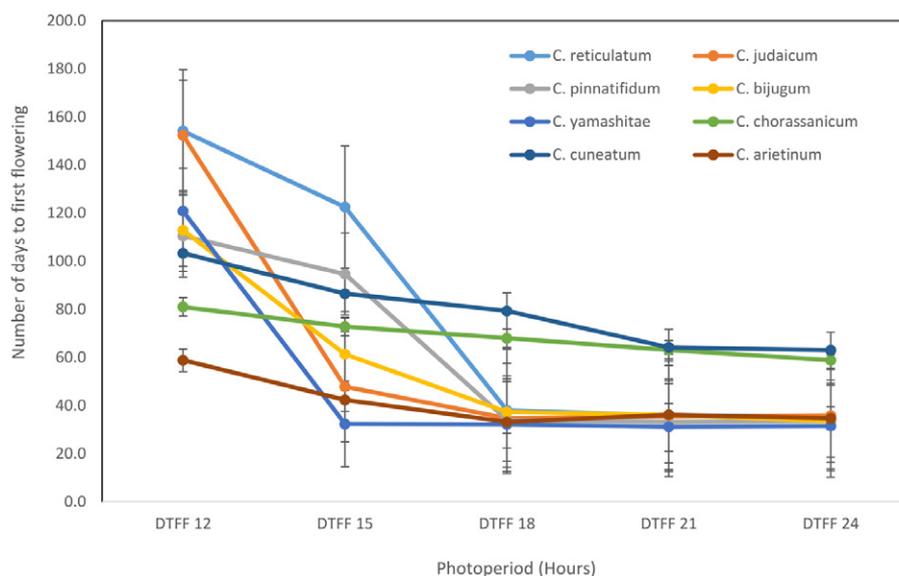

Fig. 1. Response of wild and cultivated *Cicer* species to different photoperiod treatments at ICRISAT, Patancheru, India (days to first flowering [DTFF]).

for the remaining species, such a potential was seen to be observed within 14 to 21 h.

Effect on Yield-Contributing Traits

Different daylength treatments had different effects on yield–contributing traits in different wild and cultivated *Cicer* species. At the species level, there was no significant effect of extended photoperiod treatments on plant height in *C. reticulatum*, *C. bijugum*, and *C. arietinum* (Fig. 3). In *C. judaicum*, treatments ≥18 h resulted in significant reduction in plant height (63.3 cm under 18 h, 47.2 cm under 21 h, and 34.7 cm under 24 h) compared with the control (97.0 cm) and 15 h (100.7 cm). In *C. pinnatifidum*, plant height was significantly increased under 15 h (80.7 cm) compared with the control (63.3 cm) but was significantly reduced under treatments ≥18 h (44.3 cm under 18 h, 34.8 cm under 21 h, and 27.7 cm under 24 h). Treatments ≥15 h were similar and resulted in 44 to 55% reduction in plant height compared with the control (50.5 cm) in *C. yamashitae* (23.8 cm under 15 h, 23.0 cm under 18 h, 28.3 cm under 21 h, and 26.2 cm under 24 h), whereas treatments ≥15 h were similar and resulted in 23 to 37% increases in plant height compared with the control (49.8 cm) in *C. chorassanicum* (62.8 cm under 15 h, 68.0 cm under 18 h, 61.5 cm under 21 h, and 64.3 cm under 24 h). In *C. cuneatum*, 15- and 18-h treatments were similar and both resulted in 18% increase in plant height, whereas 21- (145.3 cm) and
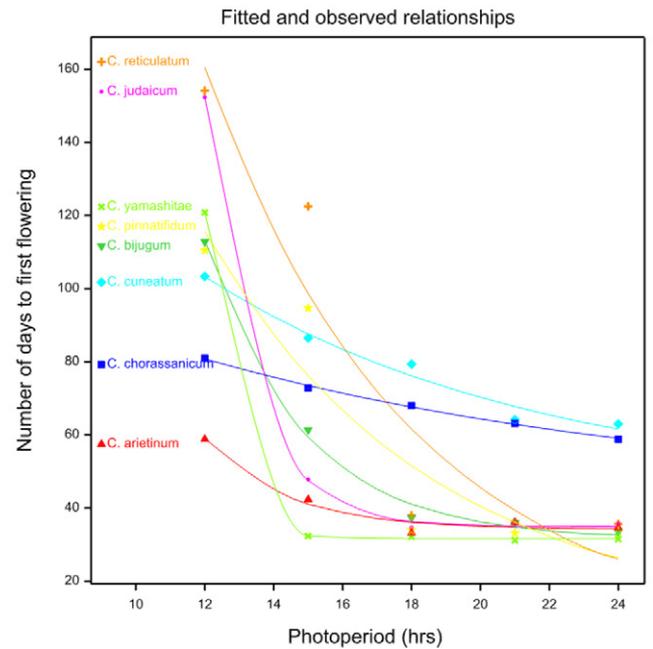

Fig. 2. Observed (points) and fitted exponential curves for days to first flowering in terms of exposure hours for eight wild and cultivated *Cicer* species.

24-h (140.3 cm) treatments had no effect on plant height compared with the control (141.8 cm) (Fig. 3).

Treatments effects on canopy width were not consistent across species. Overall, as well as in individual

Table 3. Regression relationships between days to first flowering and photoperiod.

No.	Species	Best of the three models	Adjusted R^2	Coefficients of the regression model†			
				a	*b*	*c*	*d*
			%				
1	C. reticulatum	Quadratic	85.1	624	−52.3	1.153	
2	C. judaicum	Cubic	97.8	2249	−338.9	17.08	−0.2834
3	C. pinnatifidum	Quadratic	79.7	414	−33.5	0.731	
4	C. bijugum	Cubic	99.5	927	−123.9	5.74	−0.0885
5	C. yamashitae	Cubic	92.5	2005	−309	15.9	−0.269
6	C. chorassanicum	Cubic	99.7	166	−12.22	0.533	−0.00874
7	C. cuneatum	Quadratic	95.7	197.7	−10.1	0.185	
8	C. arietinum	Cubic	94.1	369	−48.2	2.31	−0.0365

† Quadratic regression: DTFF = $a + bh + ch^2$. Cubic regression: DTFF = $a + bh + ch^2 + dh^3$. DTFF is days to first flowering; h is daylength exposure in hours.

Table 4. Estimates of parameters of exponential curve fitted to days to first flowering in exposure (in hours) and estimated exposure hours to reach 10% above the potential value of days to first flowering.

No.	Species	Adjusted R^2	Estimates of parameters†			DTFF observed at 24 h	DTFF1 (10% above *A*)	Estimated photoperiod for DTFF1
			R	*B*	*A*			
		%				d		h
1	C. reticulatum	80.4	0.8431	1,196	6.2	35.7	6.8	44.3
2	C. judaicum	99.9	0.475	892,149	34.93	35.8	38.4	16.7
3	C. pinnatifidum	74.9	0.8554	686	10.3	33.5	11.3	41.6
4	C. bijugum	99.1	0.698	6,055	31.6	33.5	34.8	21.0
5	C. yamashitae	100	0.2	1.92×10^{10}	31.61	31.5	34.8	14.0
6	C. chorassanicum	99.5	0.931	89	43	58.8	47.3	42.4
7	C. cuneatum	95.3	0.901	203	44.9	63.0	49.4	36.6
8	C. arietinum	94.4	0.653	4,123	34.16	34.7	37.6	16.6
	When fitted jointly over all the species	89.5						

† Exponential curve: DTFF = $A + BR^h$. DTFF is days to first flowering; *h* is daylength exposure in hours.
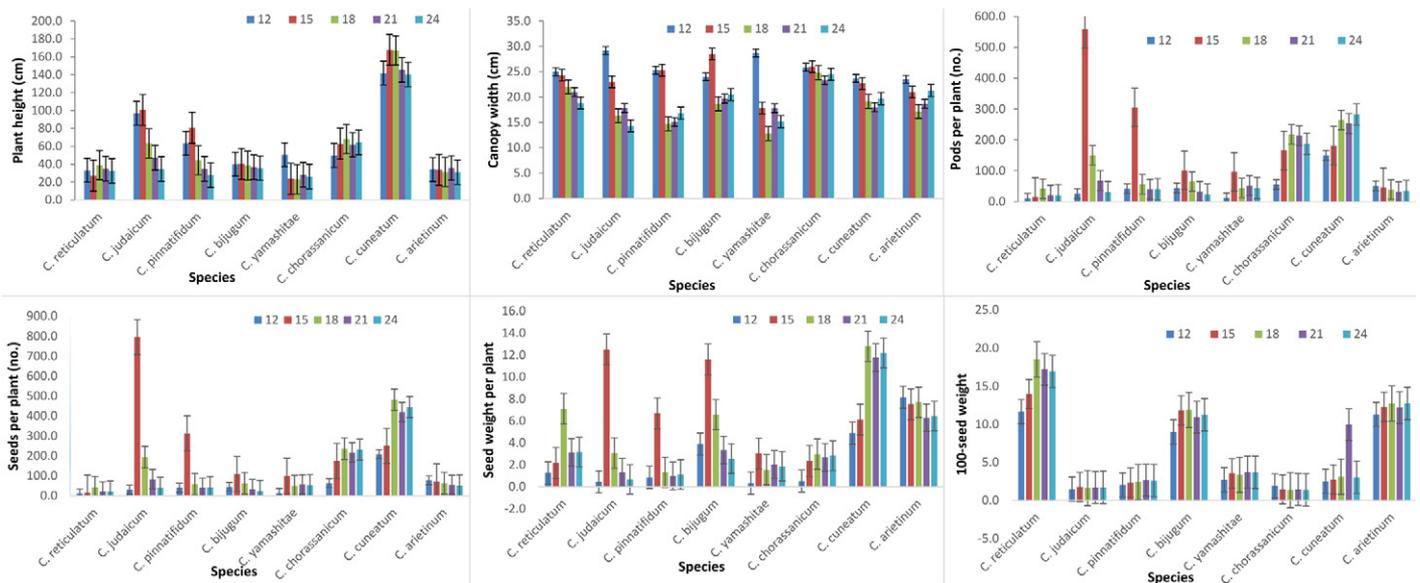
Fig. 3. Effect of different extended photoperiod treatments on yield-contributing traits at ICRISAT, Patancheru, India.

species *C. reticulatum*, *C. chorassanicum*, *C. cuneatum*, and *C. arietinum*, there was no effect of extended photoperiod treatments on canopy width compared with the control. Canopy width was minimal and similar under treatments ≥18 h compared with 15 h and the control in *C. judaicum*, *C. pinnatifidum*, and *C. bijugum*. In *C. yamashitae*, canopy width was reduced significantly under all treatments compared with the control (Fig. 3).

All treatments had similar effects on number of pods per plant and seeds per plant (Fig. 3). Overall, 15-h treatment resulted in the highest number of pods and seeds per plant compared with the control, followed by 18-, 21-, and 24-h treatments. However, different species showed distinct responses. No treatment had a significant effect on number of pods and seed per plant in *C. reticulatum* and *C. arietinum*. The 15-h treatment was the most effective at producing the highest number of pods and seeds per plant compared with the control and other treatments in *C. judaicum*, *C. bijugum*, *C. pinnatifidum*, and *C. yamashitae*. The 18-h treatment resulted in the highest number of pods and seeds per plant in *C. chorassanicum* and *C. cuneatum* and was similar to the 21- and 24-h treatments.

Overall, the 15-h treatment resulted in the greatest seed weight per plant, followed by the 18-, 21-, and 24-h treatments, compared with the control in wild and cultivated *Cicer* species (Fig. 3). However, differential responses were observed across species. In *C. reticulatum*, 18 h resulted in the greatest seed weight per plant (7.1 g), followed by 21 (3.1 g) and 24 h (3.2 g), then 15 h (2.2 g), and finally the control (1.3 g). The 15-h treatment resulted in the greatest seed weight per plant in *C. judaicum* (12.5 g), *C. pinnatifidum* (6.7 g), *C. bijugum* (11.6 g), and *C. yamashitae* (3.0 g). Daylengths ≥18 h resulted in the highest and similar seed weight per plant in *C. chorassanicum* and *C. cuneatum*. All treatments

resulted in similar seed weight per plant, which was similar to the control in *C. arietinum* (Fig. 3).

For 100-seed weight, the 18-h treatment resulted in the highest 100-seed weight in *C. reticulatum*, whereas these treatments were similar to the control in *C. judaicum*, *C. pinnatifidum*, *C. yamashitae*, *C. chorassanicum*, and *C. cuneatum*. Daylengths ≥15 h were similar to and better than the control in yielding high 100-seed weight in *C. bijugum* and *C. arietinum* (Fig. 3).

DISCUSSION

The narrow genetic base of cultivated chickpea poses a serious threat for its improvement. Further, new biotic and abiotic stresses such as dry root rot and heat stress are emerging due to climate change and are causing huge yield losses in chickpea (Savary et al., 2011). Using the genetic variation present in wild species will facilitate the development of climate-resilient chickpea cultivars. Wild *Cicer* species harbor many useful genes and provide enormous genetic variation for use in chickpea improvement programs. Previous studies have reported enormous variability for morphological traits in wild annual *Cicer* species, with the largest variability in *C. reticulatum*, *C. echinospermum* P.H. Davis, and *C. bijugum* (Robertson et al., 1997; Talip et al., 2018). However, different phenology of wild *Cicer* species and cultivated chickpea (Summerfield and Roberts, 1987; Summerfield et al., 1989; Robertson et al., 1997; Abbo et al., 2002; Berger et al., 2005) hinders their frequent utilization in chickpea breeding programs, especially due to vernalization and extended photoperiod requirements for flowering (Abbo et al., 2002; Sharma and Upadhyaya, 2015). Vernalization and photoperiod sensitivity of wild *Cicer* species is also a serious matter of concern for their efficient regeneration and conservation in genebanks. In the previous study, we concluded

that wild *Cicer* species carry both vernalization- and photoperiod-responsive genes, with the preponderance of photoperiod-responsive genes in *C. reticulatum, C. echinospermum, C. judaicum, C. bijugum, C. pinnatifidum,* and *C. yamashitae,* whereas vernalization-responsive genes were predominant in *C. chorassanicum* and *C. cuneatum* (Sharma and Upadhyaya, 2015).

The present study is an attempt to study the response of wild and cultivated *Cicer* species to different extended photoperiod treatments in terms of reduction in the vegetative phase, and to study the effect of these treatments on yield-contributing traits. The study revealed that all four photoperiod treatments were effective in reducing the vegetative phase in wild *Cicer* species (Fig. 1). It is evident from the study that a linear trend of different photoperiod treatments was the most correct for explaining variation for days to first flowering and plant height; linear, cubic, and quadratic trends were the most correct for canopy width; cubic and linear trends were the most correct for number of pods and seeds per plant; linear and cubic trends were the most correct for seed weight per plant; and linear and quadratic trends were the most correct for 100-seed weight (Table 2).

Given the extent of reduction in number of days to first flowering, daylengths ≥15 h were similar and effective at reducing the vegetative phase by ~70%, thereby resulting in a strong response in *C. judaicum* and *C. yamashitae,* whereas daylengths ≥18 h were effective at reducing the vegetative phase by 70% in *C. reticulatum, C. pinnatifidum,* and *C. bijugum* (Fig. 1). The earlier study also showed the preponderance of photoperiod-responsive genes in *C. reticulatum, C. judaiucum, C. bijugum, C. pinnatifidum,* and *C. yamashitae* (Sharma and Upadhyaya, 2015), which was confirmed by the high response (~70% reduction in vegetative phase) of these species to extended photoperiod treatments compared with the control in the present study. The extended photoperiod treatments were less effective in reducing the vegetative phase in *C. chorassanicum,* wherein days to first flowering were reduced only by 10 to 27% under treatments ≥15 h. Similarly, in *C. cuneatum,* the vegetative phase was reduced only by ~38 to 39% under both 21- and 24-h treatments, 23% under 18-h treatments, and 16% under 15-h treatments, showing weak response to these treatments. These results are in accordance with the previous study, which indicated the preponderance of vernalization-responsive genes in *C. chorassanicum* and *C. cuneatum* (Sharma and Upadhyaya, 2015). Cultivated *C. arietinum* also exhibited a weak response to extended photoperiod treatments, as the vegetative phase was reduced only by 28 to 44% under treatments ≥15 h. These responses could be associated with the geographical origin and distribution of these species in different environments. Two species, *C. chorassanicum* and *C. cuneatum,* are found in areas >2400-m elevation with low temperatures (<4°C) during

pod set and are therefore exposed to vernalization under natural field conditions (Berger et al., 2003). The remaining species, which are found at lower elevations, such as *C. reticulatum* (966-m mean elevation), *C. judaicum* (322-m mean elevation), *C. bijugum* (957-m mean elevation), and *C. pinnatifidum* (935-m mean elevation) were found to be more photoperiod responsive (Sharma and Upadhyaya, 2015), which is evident from the high response of these species to extended photoperiod treatments compared with *C. chorassanicum* and *C. cuneatum* in the present study. Further, the present study suggests that within photoperiod responsive species, treatments ≥18 h were more effective at reducing the vegetative phase in species such as *C. reticulatum, C. bijugum,* and *C. pinnatifidum,* which are found at comparatively higher elevations, whereas treatments ≥15 h were effective at reducing the vegetative phase in *C. judaicum,* which is found at much lower elevation. The weak response to extended photoperiod treatments in cultivated *C. arietinum* could be associated with the origin and domestication of cultivated chickpea from the wild progenitor, *C. reticulatum.* Major genes and alleles controlling photoperiod sensitivity might have been lost during the evolution and domestication of cultivated chickpea.

Besides days to first flowering, other yield-contributing traits such as plant height, number of pods and seeds per plant, seed weight per plant, and 100-seed weight were also affected by different photoperiod treatments in wild and cultivated *Cicer* species. However, differential response was observed in each species (Fig. 3). These results have major implications for the utilization of wild *Cicer* species in hybridization programs for chickpea improvement by synchronizing flowering with cultivated chickpea, and for efficient regeneration and conservation in the genebanks.

In *C. reticulatum,* daylengths ≥18 h were similar and led to a strong response. As *C. reticulatum* yielded the highest number of pods and seeds per plant, seed weight per plant, and 100-seed weight at 18 h compared with other treatments (Fig. 3), 18 h may be regarded as the critical daylength required for synchronizing flowering and for efficient regeneration of this species.

In *C. judaicum* and *C. yamashitae,* daylengths ≥15 h resulted in strong responses, and plants yielded the highest number of pods, seeds per plant, and seed weight per plant under 15 h (Fig. 3). Thus, 15 h could be regarded as the critical daylength required for both synchronizing flowering with cultivated chickpea and for efficient regeneration of these two species.

In *C. pinnatifidum* and *C. bijugum,* although daylengths ≥18 h were equally effective in reducing the vegetative phases, plants yielded significantly highest number of pods and seeds per plant, seed weight per plant, and 100-seed weight under 15 h (Fig. 3). Thus, it can be concluded that 15 h is the most effective treatment for regeneration of this accession, whereas for a crossing program, 18-h treatment

should be used, as this treatment will help to synchronize the flowering of *C. pinnatifidum* with cultivated chickpea genotypes, which usually flower within 35 to 50 d.

In *C. chorassanicum*, and *C. cuneatum*, which are mainly vernalization responsive (Sharma and Upadhyaya, 2015), and in cultivated *C. arietinum* with weak response to extended photoperiod treatments, the present study revealed that daylengths ≥15 h in *C. chorassanicum* and *C. arietinum* and daylengths ≥18 h in *C. cuneatum* were effective at reducing the vegetative phase to some extent and yielding more pods and seeds per plant compared with the control (Fig. 3).

For each trait, the high percentage of variation explained by cubic polynomials indicated that the response could be modeled using the cubic polynomials in the exposure time, but further improvement became possible by exploring other nonlinear forms such as exponential.

The present study holds great potential for improving the understanding of mechanisms controlling the physiological behavior of wild and cultivated *Cicer* species. In this study, photoperiod sensitivity and critical daylength in wild *Cicer* species clearly exhibited latitudinal clines, wherein the lower the latitude, the shorter the critical daylength. The results indicated the requirement of long photoperiods (varying from 15 to 18 h) in wild *Cicer* species for their transition from vegetative into reproductive phase. It is also evident from the study that optimum photoperiods improve agronomic traits such as pod and seed yield, which could be due to the better assimilation and translocation of photosynthates from source to sink. Overall, it can be concluded that critical photoperiods for transition from the vegetative to reproductive phase and for net assimilation rate varies across wild and cultivated *Cicer* species. The optimum daylength requirement for different *Cicer* species identified in this study will be helpful in utilizing the wild *Cicer* species in chickpea improvement programs, as well as in their efficient regeneration for conservation in genebanks.

Conflict of Interest

The authors declare that there is no conflict of interest.

Acknowledgments

References

Abbo, S., S. Lev-Yadun, and N. Galwey. 2002. Vernalization response of wild chickpea. New Phytol. 154:695–701. doi:10.1046/j.1469-8137.2002.00405.x

Berger, J., S. Abbo, and N.C. Turner. 2003. Ecogeography of annual wild *Cicer* species: The poor state of the world collection. Crop Sci. 43:1076–1090. doi:10.2135/cropsci2003.1076

Berger, J.D., R. Buck, J.M. Henzell, and N.C. Turner. 2005. Evolution in the genus *Cicer*-vernalization response and low temperature pod set in chickpea (*C. arietinum* L.) and its annual wild relatives. Aust. J. Agric. Res. 56:1191–1200. doi:10.1071/AR05089

Daba, K., B. Tar'an, and T.D. Warkentin. 2016. Flowering response of diverse chickpea (*Cicer arietinum* L.) accessions to photoperiod. Genet. Resour. Crop Evol. 63:1161–1172. doi:10.1007/s10722-015-0308-5

FAOSTAT. 2018. FAOSTAT: Crops. FAO, Rome. http://www.fao.org/faostat/en/#data/QC (accessed 5 Oct. 2018).

Jaiswal, H.K., B.D. Singh, A.K. Singh, and R.M. Singh. 1986. Introgression of genes for yield and yield traits from *C. reticulatum* into *C. arietinum*. Int. Chickpea News 14:5–8.

Knights, E.J., R.J. Southwell, M.W. Schwinghamer, and S. Harden. 2008. Resistance to *Phytophthora medicaginis* Hansen and Maxwell in wild *Cicer* species and its use in breeding root rot resistant chickpea (*Cicer arietinum* L.). *Aus*. J. Agric. Res. 59:383–387. doi:10.1071/AR07175

Malhotra, R.S., K.B. Singh, M. Vito, N. Greco, and M.C. Saxena. 2002. Registration of ILC 10765 and ILC 10766 chickpea germplasm lines resistant to cyst nematode. Crop Sci. 42:1756. doi:10.2135/cropsci2002.1756

Robertson, L.D., B. Ocamppo, and K.B. Singh. 1997. Morphological variation in wild annual *Cicer* species in comparison to the cultigen. Euphytica 95:309–319. doi:10.1023/A:1003004516921

Savary, S., A. Nelson, A.H. Sparks, L. Willocquet, E. Duveiller, G. Mahuku, et al. 2011. International agricultural research Tackling the effects of global and climate changes on plant diseases in the developing World. Plant Dis. 95:1204–1216. doi:10.1094/PDIS-04-11-0316

Sethi, S.C., D.E. Byth, C.L.L. Gowda, and J.M. Green. 1981. Photoperiodic response and accelerated generation turnover in chickpea. Field Crops Res. 4:215–225. doi:10.1016/0378-4290(81)90073-3

Sharma, S., and H.D. Upadhyaya. 2015. Vernalization and photoperiod response in annual wild *Cicer* species and cultivated chickpea. Crop Sci. 55:2393–2400. doi:10.2135/cropsci2014.09.0598

Singh, K.B. 1987. Chickpea breeding. In: M.C. Saxena and K.B. Singh, editors, The chickpea. CAB Int., Wallingford, UK. p. 127–162.

Singh, K.B., and B. Ocampo. 1997. Exploitation of wild *Cicer* species for yield improvement in chickpea. Theor. Appl. Genet. 95:418–423. doi:10.1007/s001220050578

Singh, M., G.K. Kanji, and K.S. El Bizri. 1992. A note on inverse estimation in non-linear models. J. Appl. Stat. 19:473–477. doi:10.1080/02664769200000042

Singh, S., R.K. Gumber, N. Joshi, and K. Singh. 2005. Introgression from wild *Cicer reticulatum* to cultivated chickpea for productivity and disease resistance. Plant Breed. 124:477–480. doi:10.1111/j.1439-0523.2005.01146.x

Summerfield, R.J., R.H. Ellis, and E.H. Roberts. 1989. Vernalization in chickpea (*Cicer arietinum*): Fact or artefact? Ann. Bot. (Lond.) 64:599–603. doi:10.1093/oxfordjournals.aob.a087882

Summerfield, R.J., and E.H. Roberts. 1987. Effect of illuminance on flowering in long- and short-day grain legumes: A reappraisal and unifying model. In: J.G. Atherton, editor, Manipulation of flowering. Butterworths, London. p. 203–223. doi:10.1016/B978-0-407-00570-9.50020-X

Talip, M., A. Adak, A. Kahraman, J. Berger, D. Sari, H. Sari, et al. 2018. Agro-morphological traits of *Cicer reticulatum* Ladizinsky in comparison to *C. echinospermum* PH Davis in terms of potential to improve cultivated chickpea (*C. arietinum* L.). Genet. Resour. Crop Evol. 65:951–962. doi:10.1007/s10722-017-0587-0

Upadhyaya, H.D. 2008. Crop germplasm and wild relatives: A source of novel variation for crop improvement. Korean J. Crop Sci. 53:12–17.

VSN International. 2015. The guide to the Genstat command language (Release 18), Part 2. Statistics. VSN Int., Hemel Hempstead, UK.