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EFFECTS OF DIFFERENT PHOTOPERIODS ON FLOWERING TIME OF QUALITATIVE LONG DAY ORNAMENTAL ANNUALS

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Abstract

Present study was carried out at the Agricultural Research Institute, Dera Ismail Khan, Pakistan, during the year 2005. Seeds of five qualitative LDPs (Pot Marigold cv. Resina, Annual Phlox cv. Astoria Magenta, Cornflower cv. Florence Blue, Oriental Poppy cv. Burning Heart, Flax cv. Scarlet Flax) were sown on 1st March 2005. The experiment was designed to study flowering response under four distinct controlled photoperiods (11, 13, 15 and 17 $h.d^{-1}$). A curvilinear qualitative response was observed in almost all cultivars studied. Pot Marigold, Annual Phlox, Cornflower, Oriental Poppy and Flax took minimum time to flower when grown under 17 $h.d^{-1}$ photoperiods however it was significantly ($p<0.05$) increased when photoperiod decreased to 11 $h.d^{-1}$.

Introduction

Flowering is the end result of physiological processes, biochemical sequences, and gene action, with the whole system responding to the influence of environmental stimuli (photoperiod, temperature) and the passage of time (Munir, 2003; Zheng *et al.,* 2006). Generally, after attaining a certain size (completing the 'juvenile' phase) plants enter into the 'reproductive' phase (initiation and development of flowering). Evans (1969) referred to flowering as the inductive processes occurring in the leaf, mediated by the photoreceptor, phytochrome that leads to the initiation of flowering at the meristem (evocation). Inductive processes occur in the leaf (O'Neil, 1992) and result in floral initiation in which the apical meristem changes towards floral development (McDaniel *et al.,* 1992). It is also believed that flowering is induced by a stimulus (florigen), which is produced within the leaf (Chailakhyan, 1936) but this hormone has not yet been identified. When the apical meristem of the plant is committed to flowering, its fate becomes irreversible (Bernier, 1988), although flower or inflorescence reversion to vegetative growth can also occur spontaneously in some species. This condition can be caused if plants are transferred to certain specific photoperiod or temperature regimes, which favour vegetative development (Battey & Lyndon, 1990).

Many flowering plants use a photoreceptor protein, such as phytochrome or cryptochrome, to sense seasonal changes in day length (photoperiod), which they take as signals to flower (Weller & Kendrick, 2008). The photoperiodic response of flowering is generally categorised into three main groups: short-day plants (SDPs) in which flowering is hastened by longer nights; long-day plants (LDPs) where shorter nights promote flowering; and day-neutral plants (DNPs) which flower irrespective to day length. SDPs and LDPs can be further classified as qualitative or obligate (species that require a specific minimum or maximum photoperiod for flowering) and quantitative or facultative (flowering process is hastened by a specific minimum or maximum photoperiod). It is actually the night length rather than day length that controls flowering, so flowering in a long day (LD) plant is triggered by a short night (which, of course, also means a long day). Conversely, short day (SD) plants will flower when nights get longer than a critical length. This can be observed by using night breaks. For example, a short day plant (long night) will not flower if a pulse (5 minutes) of artificial light is shone on the plant during the middle of the night. This generally does not occur from natural light such as moonlight, lightning, fire flies, etc, since the light from these sources is not sufficiently strong to trigger the response (Thomas & Vince-Prue, 1997). Keeping in view the importance of photoperiod on flower induction an experiment was designed to determine the flowering response of five qualitative LDPs to four photoperiods under the sub-tropical environmental conditions of Dera Ismail Khan.

Materials and Methods

The experiment was conducted in Agricultural Research Institute, Dera Ismail Khan, Pakistan, during the year 2005. Seeds of qualitative LDPs such as Pot Marigold (*Calendula officinalis* L.) cv. Resina, Annual Phlox (*Phlox drummondii* L.) cv. Astoria Magenta, Cornflower (*Centaurea cyanus* L.) cv. Florence Blue, Oriental Poppy (*Papaver orientale* L.) cv. Burning Heart, Flax (*Linum usitatissimum* L.) cv. Scarlet Flax were sown on 1st of March 2005 into module trays containing locally prepared leaf mould compost. Seed trays were kept at room temperature at night and they were moved out during the day (08:00–16:00 h) under partially shaded area. After 70% seed germination, six replicates of each cultivar were shifted to the respective photoperiod chamber. Plants remained for 8h (from 08:00 to 16:00h) in the field (outside the photoperiod chambers) where they were exposed to natural daylight and temperature (Table 1). At 16:00h each day, all plants were moved into the photoperiod chambers where they remained until 08:00h the following morning. Photoperiod within each of the chambers was extended by two 60Watt tungsten light bulbs and one 18Watt warm white florescent long-life bulb (Philips, Holland) fixed above 1 m high from the trolleys providing a light intensity (Photosynthetic Photon Flux Density, PPFD) of 7μmol m^{-2} s^{-1}. In all photoperiod chambers, the lamps were switched on automatically at 1600 h for a duration dependents on the day length required (11, 13, 15, 17 $h.d^{-1}$). These chambers were continuously ventilated with the help of micro exhaust fan (Fan-0051, SUPERMICRO® USA) with an average air speed of 0.2 $m.s^{-1}$ over the plants when inside the chambers, to minimize any temperature increase due to heat from the lamps. Temperature and solar radiation were measured in the weather station situated one kilometer away from the research venue. Temperature was recorded with the help of Hygrothermograph (NovaLynx Corporation, USA) while solar radiation was estimated using solarimeters (Casella Measurement, UK). Plants were potted into 9cm pots containing leaf mould compost and river sand (3:1 v/v) after 6

leaves emerged. Plants were irrigated by hand and a nutrient solution [(Premium Liquid Plant Food and Fertilizer (NPK: 8-8-8); Nelson Products Inc. USA)] was applied twice a week. Plants in each treatment were observed daily until flower opening (corolla fully opened). Numbers of days to flowering from emergence were recorded at harvest and the data were analysed using GenStat-8 (Lawes Agricultural Trust, Rothamsted Experimental Station, U.K. and VSN International Ltd. U.K.). The rate of progress to flowering ($1/f$) is represented as the reciprocal of the time to flowering, therefore $1/f$ data of qualitative LDPs were analysed using the following linear model (Adams *et al.,* 1998; Munir, 2003):

$1/f = a + b\,P$ (where a and b are constants and P is photoperiod)

Results

Results of present work indicated that there was a significant ($p<0.05$) difference among four photoperiods regarding flowering time in qualitative LDPs such as Pot Marigold cv. Resina (Fig. 1A), Annual Phlox cv. Astoria Magenta (Fig. 1B), Cornflower cv. Florence Blue (Fig. 1C), Oriental Poppy cv. Burning Heart (Fig. 1D) and Flax cv. Scarlet Flax (Fig. 1E). Days taken to produce flower was increased significantly when these LDPs were grown under 11 $h.d^{-1}$ photoperiod, however it was linearly decreased when they were grown in 13, 15 and 17 $h.d^{-1}$ photoperiod chambers.

Flowering was delayed by 25 days when Pot Marigold cv. Resina (Fig. 1A) was grown under 11 $h.d^{-1}$ photoperiod i.e. 100 days in 11 $h.d^{-1}$ photoperiod and 75 days in 17 $h.d^{-1}$ photoperiod. Similarly, plants received 13 and 15 $h.d^{-1}$ photoperiod flowered after 88 and 77 days respectively. Similarly, Annual Phlox cv. Astoria Magenta (Fig. 1B) took 75 days to produce flower when grown under LD (17 $h.d^{-1}$ photoperiod) as compared to 11 $h.d^{-1}$ photoperiod (99 days to produce flower) i.e., 24 days early flowering in LD environment. Plants grown in 13 and 15 $h.d^{-1}$ photoperiod flowered in 87 and 75 days respectively. Similarly, time to flowering was increased up to 22 days when plants of Cornflower cv. Florence Blue (Fig. 1C) were grown under 11 $h.d^{-1}$ photoperiod (86 days) as compared to 17 $h.d^{-1}$ photoperiod (108 days). Plants grown in 13 and 15 $h.d^{-1}$ photoperiod took 96 and 88 days to flower respectively. Oriental Poppy cv. Burning Heart (Fig. 1D) flowered after 72 days in 17 $h.d^{-1}$ photoperiod followed by 73 days in 15 $h.d^{-1}$ photoperiod. However, in 11 $h.d^{-1}$ photoperiod plants bloomed after 96 days from emergence followed by 84 days in 13 $h.d^{-1}$ photoperiod. A difference of 25 days was recorded between the two extreme photoperiods. Similarly, 11 $h.d^{-1}$ photoperiod (110 days) delayed flowering time up to 24 days in Flax cv. Scarlet Flax (Fig. 1E) as compared to 17 $h.d^{-1}$ photoperiod (86 days) whereas plants grown in 13 and 15 $h.d^{-1}$ photoperiod took 98 and 88 days to flower respectively.

The best fitted model describing the effects of mean photoperiod (P) on the rate of progress to flowering ($1/f$) can be written as:

Pot Marigold cv. Resina (Fig. 2A) and (Fig. 3A):
$1/f$ = -120.86 (±2.87) + 2.87 (±0.22) P (r^2=0.97, d.f. 23) Eq. 1
Annual Phlox cv. Astoria Magenta (Fig 2B) and (Fig 3B):
$1/f$ = -118.72 (±3.18) + 2.79 (±0.25) P(r^2=0.98, d.f. 23) Eq. 2
Cornflower cv. Florence Blue (Fig. 2C) and (Fig. 3C):
$1/f$ = -124.72 (±2.93) + 2.43 (±0.23) P(r^2=0.99, d.f. 23) Eq. 3
Oriental Poppy cv. Burning Heart (Fig. 2D) and (Fig. 3D):
$1/f$ = -116.36 (±3.03) + 2.82 (±0.24) P(r^2=0.98, d.f. 23) Eq. 4
Flax cv. Scarlet Flax (Fig. 2E) and (Fig. 3E):
$1/f$ = -129.94 (±2.55) + 2.76 (±0.20) P(r^2=0.99, d.f. 23) Eq. 5

Above equations 1-5 are based on individual arithmetic means of respective factors, although all data were originally tested. The values in parenthesis show the standard errors of the regression coefficients. The outcome of this model indicated that photoperiod had significant effects on the rate of progress to flowering in all qualitative LDPs studied. For validation of the model actual data of rate of progress to flowering were plotted against the predicted ones. To develop a fitted relationship and almost all values were successfully plotted near the line of identity which also showed that the photoperiod had a significant effect on the rate of progress to flowering.

Discussion

Results of present experiment showed a qualitative LD photoperiodic response of Pot Marigold cv. Resina, Annual Phlox cv. Astoria Magenta, Cornflower cv. Florence Blue, Oriental Poppy cv. Burning Heart and Flax cv. Scarlet Flax i.e. photoperiod is essential for flowering. These results are in line with the findings of Erwin & Warner (2002) who reported that plants LD photoperiod hastened flowering in many LDPs studied. Present results indicated that flowering time was hastened up to 25 (Pot Marigold cv. Resina and Oriental Poppy cv. Burning Heart), 24 (Annual Phlox cv. Astoria Magenta and Flax cv. Scarlet Flax) and 22 days (Cornflower cv. Florence Blue) under LD environment (17 $h.d^{-1}$). The response of LDPs observed in present study supporting the fact that these plants are from Mediterranean or temperate origin where the day length/photoperiod is much longer than in the tropics and plants originating from this region prefer an open environment with ample sunshine (Summerfield *et al.,* 1997). Studies have been carried out previously to support this evidence in Annual Phlox (Runkle *et al.,* 1998), Oriental Poppy (Gentner *et al.,* 1975; Acock *et al.,* 1996; Wang *et al.,* 1998, 1999) and Flax (Kurt & Bozkurt, 2006).

Table 1. Environmental detail of the experiment.

Growth period	Diurnal temperature (°C)			Daily light integral 08:00-16:00
	Maximum	Minimum	Average	
March 2005	26.19	13.29	19.74	8.43 $MJ.m^{-2}.d^{-1}$
April 2005	32.87	15.73	24.30	9.45 $MJ.m^{-2}.d^{-1}$
May 2005	36.39	20.35	28.37	9.40 $MJ.m^{-2}.d^{-1}$
June 2005	42.27	30.70	36.48	9.99 $MJ.m^{-2}.d^{-1}$
July 2005	36.77	25.68	31.23	9.42 $MJ.m^{-2}.d^{-1}$

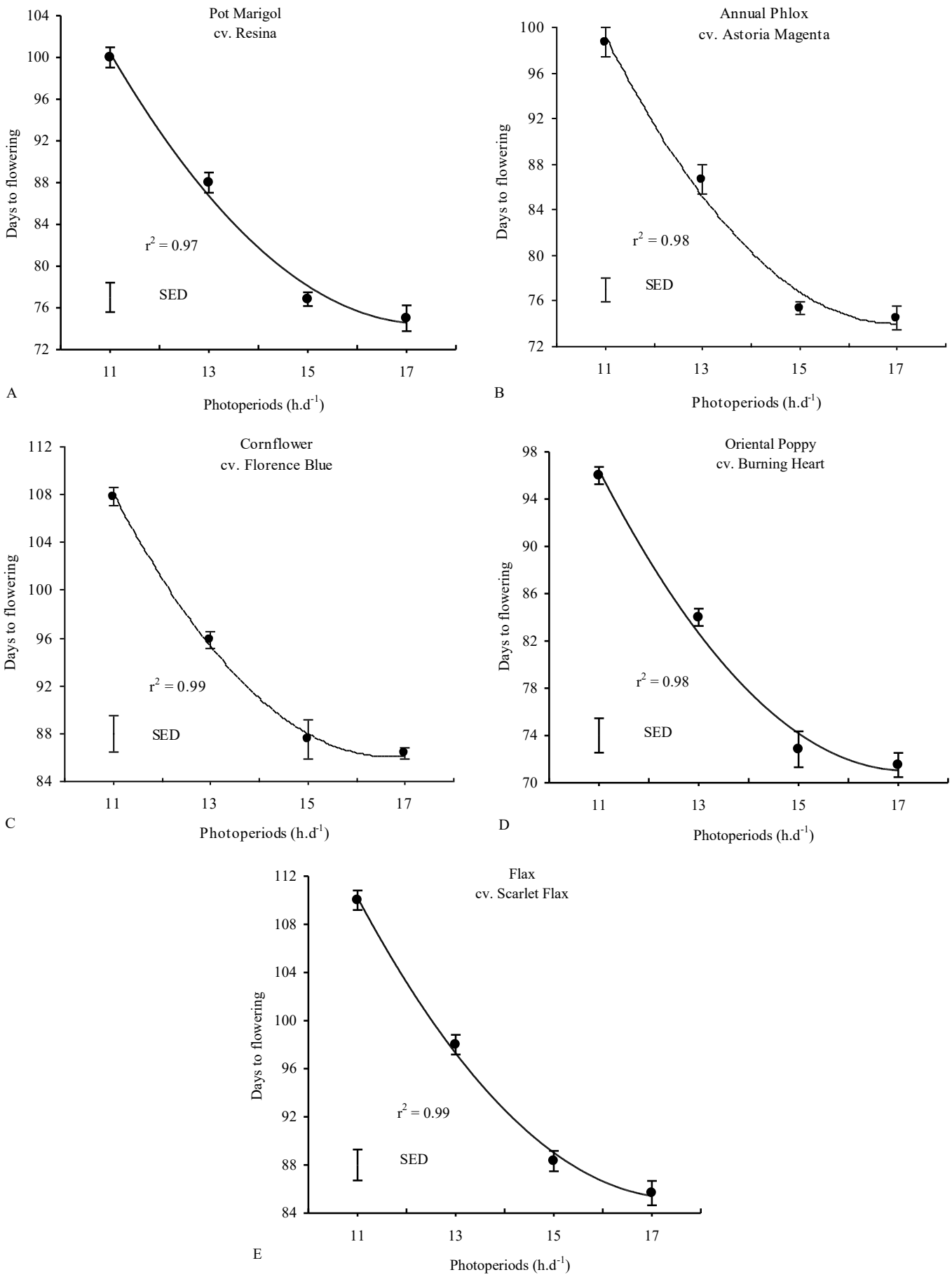

Fig. 1. Effect of different photoperiods on flowering time of (A) Pot Marigold cv. Resina, (B) Annual Phlox cv. Astoria Magenta, (C) Cornflower cv. Florence Blue, (D) Oriental Poppy cv. Burning Heart and (E) Flax cv. Scarlet Flax. Each point represents the mean of 6 replicates. Vertical bars on data points (where larger than the points) represent the standard error within replicates whereas SED vertical bar showing standard error of difference among means.

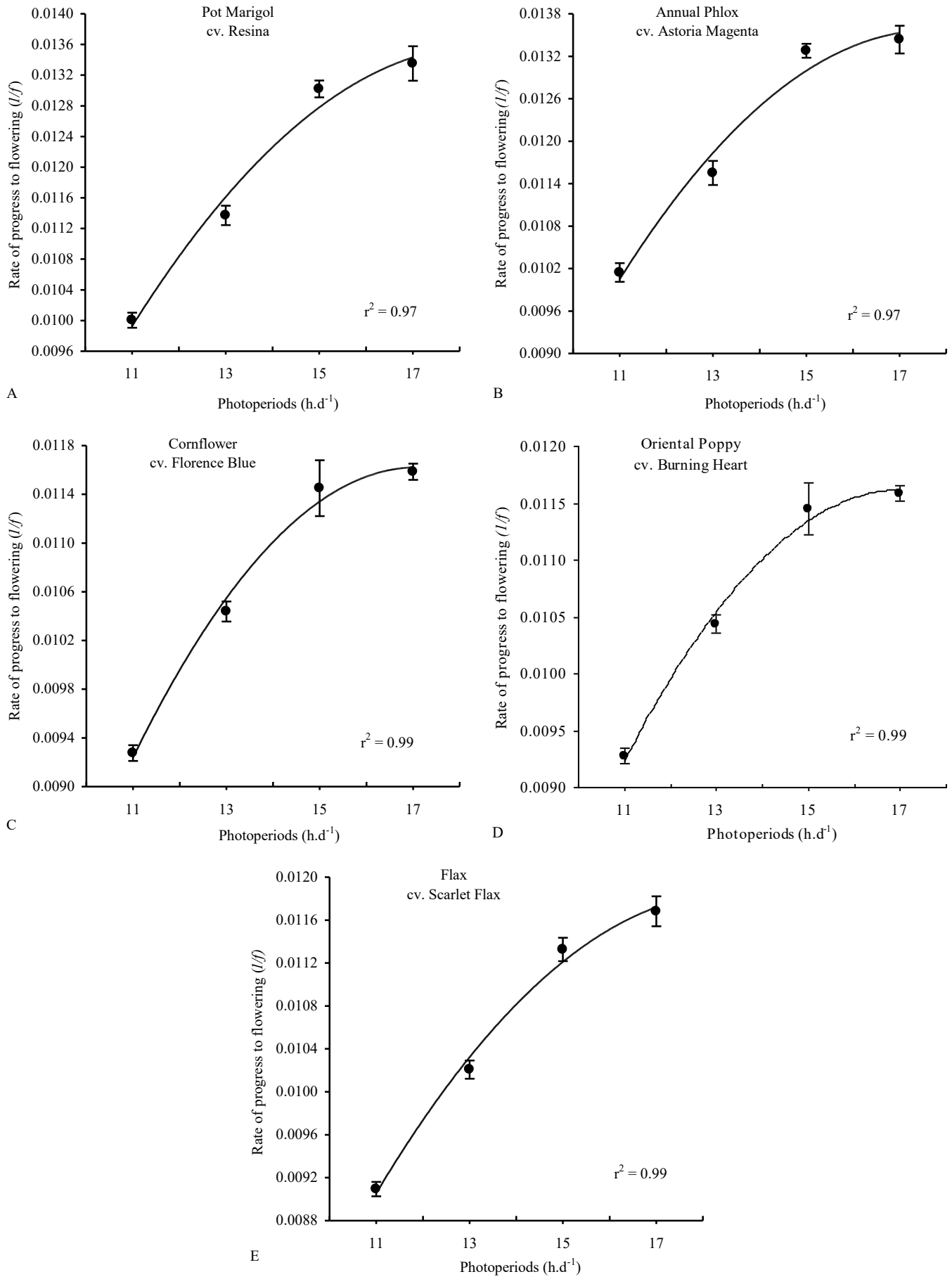

Fig. 2. Effect of different photoperiods on rate of progress to flowering (1/*f*) of (A) Pot Marigold cv. Resina, (B) Annual Phlox cv. Astoria Magenta, (C) Cornflower cv. Florence Blue, (D) Oriental Poppy cv. Burning Heart and (E) Flax cv. Scarlet Flax. Each point represents the mean of 6 replicates. Vertical bars on data points (where larger than the points) represent the standard error within replicates.

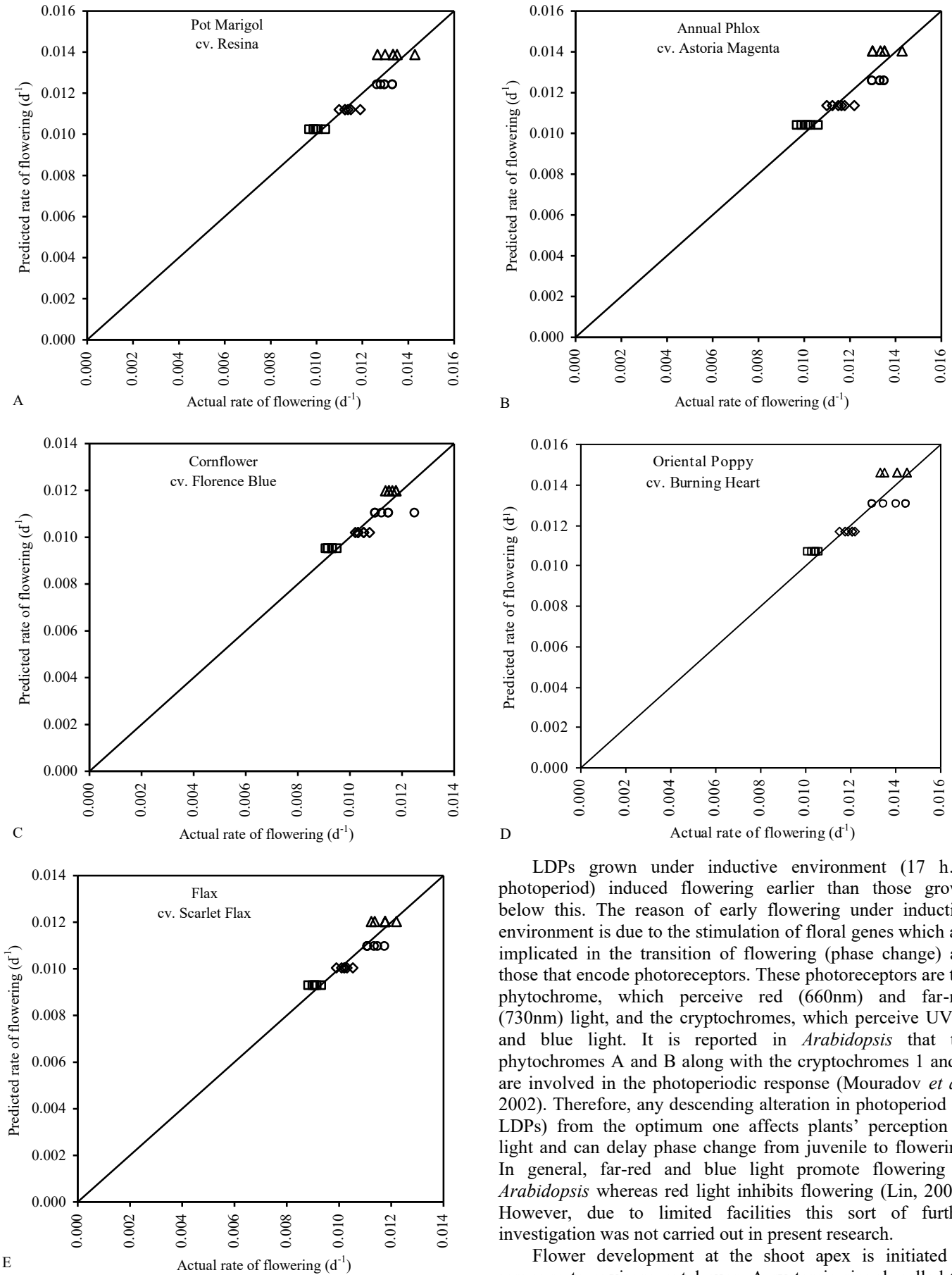

Fig. 3. The relationship between the actual rate of progress to flowering against those fitted by the flowering model ($1/f = a + bP$) for (A) Pot Marigold cv. Resina, (B) Annual Phlox cv. Astoria Magenta, (C) Cornflower cv. Florence Blue, (D) Oriental Poppy cv. Burning Heart and (E) Flax cv. Scarlet Flax grown under 8 (□), 11 (◊), 14 (○) and 17 (Δ) h.d^{-1} photoperiod. The sold line is the line of identity.

LDPs grown under inductive environment (17 h.d^{-1} photoperiod) induced flowering earlier than those grown below this. The reason of early flowering under inductive environment is due to the stimulation of floral genes which are implicated in the transition of flowering (phase change) are those that encode photoreceptors. These photoreceptors are the phytochrome, which perceive red (660nm) and far-red (730nm) light, and the cryptochromes, which perceive UV-A and blue light. It is reported in *Arabidopsis* that the phytochromes A and B along with the cryptochromes 1 and 2 are involved in the photoperiodic response (Mouradov *et al.,* 2002). Therefore, any descending alteration in photoperiod (in LDPs) from the optimum one affects plants' perception of light and can delay phase change from juvenile to flowering. In general, far-red and blue light promote flowering in *Arabidopsis* whereas red light inhibits flowering (Lin, 2000). However, due to limited facilities this sort of further investigation was not carried out in present research.

Flower development at the shoot apex is initiated in response to environmental cues. A systemic signal, called the floral stimulus, is transmitted from the leaves through the phloem and induces floral development at the shoot apex. An *et al.* (2004) identified a pathway of genes required for the initiation of flowering in response to photoperiod in *Arabidopsis*. The nuclear zinc-finger protein CONSTANS

(CO) plays a central role in this pathway and in response to LD activates the transcription of FT (FLOWERING LOCUS T) gene, which encodes a RAF-kinase-inhibitor-like protein. After the activation of FT, CO regulates the synthesis or transport of a systemic flowering signal, thereby positioning this signal within the established hierarchy of regulatory proteins that controls flowering. It can be related to present study in a way that qualitative LDPs committed to flower earlier when they received sufficient duration of LD.

The transduction of the light signals involves a complex web of interactions between photoreceptors and their corresponding interacting proteins. In term of floral induction, perception of photoperiod appears to be one of the most important transducers of the plant's environment. An important mechanism used by the plants phytochromes and cryptochromes to communicate photoperiod activity involves the entrainment of the circadian rhythms, a self-reinforcing endogenous clock that allows light/dark coordinated gene expression. Mizoguchi *et al*., (2005) reported that GIGANTEA (GI) gene regulates circadian rhythms and acts earlier in the hierarchy than CO and FT and suggested that GI acts between the circadian oscillator and CO to promote flowering by increasing CO and FT mRNA abundance.

These studies established an understanding that different genes control flowering process and these genes are evoked when a leaf is fated to respond to the inductive photoperiod, the leaf exports floral stimulus towards apex. In most cases, when the photoperiod becomes non-inductive (11 $h.d^{-1}$, in present study), the leaf stops exporting signal. The important developmental event in leaf formation, as far as photoperiodic induction is concerned, appears to be the commitment of a leaf to develop the capacity to respond to the inductive photoperiod (McDaniel, 1996). In present study, it is revealed that after completing the juvenile phase (attaining a specific leaf numbers), the competent leaf (newly developed one) respond to the inductive photoperiod (day length) and induced floral signal toward apex to produce flower that is why an early flowering response was observed under inductive photoperiod environment in LDPs.

Conclusion

It can be concluded from the findings of present research that days taken to produce flower (flowering time) in Pot Marigold, Annual Phlox, Cornflower, Oriental Poppy and Flax can be prolonged under 11 $h.d^{-1}$ non-inductive environment in order to continuous supply of these plants in the market and to enhance their flower display period. However, these LDPs can be subjected to LD inductive environment if an early flowering is required. These plants can be grown under non-inductive environment (11 $h.d^{-1}$) during juvenile phase to improve their quality for marketing viewpoint. The outcome of present study also indicated a possibility of year-round production of these plants, which will eventually increase the income of ornamental growers.

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