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Effects of Temperature and Photoperiod on Flowering in Lentils (*Lens culinaris* Medic.)

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ABSTRACT

Factorial combinations of three photoperiods (10, 13 and 16 h), two day temperatures (18 and 28 °C) and two night temperatures (5 and 13 °C) were imposed on nodulated plants of six diverse genotypes (cultivars and land-races) of lentil (*Lens culinaris* Medic.) grown in pots in growth cabinets from vernalized (1·5±0·5 °C for 30 d) or non-vernalized seeds (i.e. 144 'treatment' combinations). The times from sowing to the appearance of first open flowers were recorded. Vernalization, long days and warm temperatures hastened flowering but genotypes differed in *relative* sensitivity to each of these factors and in time to flowering in the same most-inductive environment. Rates of progress towards flowering (i.e. $1/f$, the reciprocals of the times to first flower, f) in all genotypes, vernalized or not, were linear functions of both mean temperature, $\bar{t}$, and photoperiod, p, with no interaction between the two terms. So, over a wide range of conditions (covering the photo-thermal regimes experienced by lentil crops world-wide), time to flowering can be described by the equation: $1/f = a + b\bar{t} + cp$, where a, b and c are constants which differ between genotypes and the values of which provide a sound basis for screening germplasm for sensitivity to temperature and photoperiod. Although these two environmental factors affect the same phenological event (i.e. time to flowering) our data suggest the responses are under separate genetic control. Seed vernalization consistently increased the values of both a and b in all genotypes. The implications of these collective findings for the screening of lentil germplasm are discussed.

Key words: *Lens culinaris* Medic., lentil, flowering, photoperiod, temperature, germplasm screening.

INTRODUCTION

Little is known about genetic variation in, or environmental regulation of, the induction or development of lentil (*Lens culinaris* Medic.) flowers. That information which is available has been reviewed elsewhere (Saint-Clair, 1972; Summerfield, 1981).

The period of vegetative growth in lentil crops in countries such as Lebanon, Iran, Italy, Spain, Syria and Turkey coincides with progressively lengthening days and warmer temperature. In comparison, crops in India and Pakistan at this stage of development experience shortening days and cool, or even cold (0–2 °C), average air temperature (Sinha, 1977). Seasonal changes in photo-thermal conditions are likely to be important determinants of the rate of reproductive ontogeny (Summerfield, Muehlbauer and Roberts, 1985).

Some genotypes of lentil respond to cold vernalization; sensitive genotypes flower sooner when either seeds or seedlings have been chilled (Summerfield, 1981). A modest vernalization requirement may be advantageous in Mediterranean climates and for 'rabi' (winter) cropping in Asia: flowers would be less likely to appear before winter and until plants were well-established. But the consequence of vernalization for subsequent flowering depends on the genotypes tested, the severity and duration of the cold temperature imposed, and on post-vernalization photo-thermal conditions – and little is known about the relative importance of these factors (Summerfield *et al.*, 1985).

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The few genotypes examined so far show that lentils flower sooner in longer (16–24 h) than in shorter (6–12 h) photoperiods: some genotypes respond as qualitative, others as quantitative long-day plants and some have been described as 'day-neutral'. Then again, air temperatures outside the range used for studies on vernalization (i.e. from about 6 to 30 °C) also influence dates of flowering (warmer conditions favouring more rapid flowering), but which factor in natural environments is the more significant, photoperiod or temperature, remains purely speculative (Summerfield *et al.*, 1985). The duration of the crop from sowing to harvest is important, especially in the stressful environments in which lentils are often grown. Breeders therefore need to predict the responses of improved genotypes to various conditions in order to fit them to different seasons and farming systems (Solh and Erskine, 1981). And so, they need to screen large numbers of germplasm accessions in order to identify those potentially well-adapted (because of their phenological responses) to different climates (Muehlbauer, Cubero and Summerfield, 1985).

In previous papers we have described the quantitative responses of three species of grain legumes to photo-thermal effects on flowering – two short-day species, cowpea and soyabean (e.g. Hadley *et al.*, 1983, 1984) and one long-day species, chickpea (Roberts, Hadley and Summerfield, 1985). We describe here the responses of lentils, another long-day species, and suggest that it is now possible to draw general conclusions about photo-thermal effects on flowering in the grain legumes.

MATERIALS AND METHODS

Environmental conditions

Factorial combinations of three photoperiods (10, 13 and 16 h), two day temperatures (18 and 28 °C, both $\pm 0{\cdot}5$ °C) and two night temperatures (5 and 13 °C, both $\pm 0{\cdot}5$ °C) were imposed on nodulated plants of six lentil genotypes (Table 1) grown in pots in controlled environment growth cabinets. This wide range of conditions covers typical average values of photoperiod and temperature experienced by lentil crops during vegetative periods of two to five months duration in the principal production regions in Ethiopia, India, Turkey, Syria, the USA and Canada.

Relative humidity ($60 \pm 2{\cdot}5$ per cent) and CO_2 concentrations (340 ± 15 mg l^{-1}) were maintained constant in each cabinet, the former by appropriate adjustments of vapour pressure deficit in the different thermal regimes. Plants were illuminated by 'Daylight' fluorescent tubes giving a radiation flux density of 151 J m^{-2} s^{-1} at pot level. Photoperiod was controlled by switching the main light source rather than by extending a common short photoperiod by different durations of dim supplementary light. (The reasons for this approach have been discussed elsewhere; Hadley *et al.*, 1983). Thus, plants grown in the 13 and 16 h photoperiods received 30 and 60 per cent more photosynthetically active radiation, respectively, than those grown in the 10 h regime. Furthermore, since in natural conditions the diurnal change in air temperature lags asymmetrically behind that of radiation, so day temperatures in the cabinets were set to start 2 h after the lights came on and to end at 'lights off'. A useful consequence of this (inevitably confounded) factorially designed investigation is that it provides a wide range of mean temperatures (Fig. 1). Since the switch to the nominal day temperature was delayed for 2 h after the start of the photoperiod, these values are calculated as: [(duration of photoperiod -2) $\times$ nominal day temperature $+$ (duration of night period $+2$) $\times$ nominal night temperature]/24.

TABLE 1. *Origin and selected characteristics of the lentil genotypes grown in each photothermal regime*

Germplasm accession*	Name	Country of origin	Median seed weight (mg)	Comments†
ILL 9	78S-26004	Jordan	$51{\cdot}8 \pm 3{\cdot}1$	Selection from germplasm; medium maturity; wide adaptation
ILL 1744	EL79	Ethiopia	$26{\cdot}9 \pm 1{\cdot}7$	Early maturing land-race
ILL 2501	Pant-L-406	India	$20{\cdot}9 \pm 1{\cdot}7$	Released cultivar; early maturing
ILL 4349	Laird	Russia	$66{\cdot}4 \pm 5{\cdot}3$	Released cultivar (in Canada); very late to mature
ILL 4400	Syrian local large	Syria	$72{\cdot}9 \pm 2{\cdot}2$	Medium maturity land-race
ILL 4605	Precoz	Argentina	$48{\cdot}3 \pm 3{\cdot}5$	Released cultivar; early maturing

* Accession number of the world lentil germplasm collection maintained at the International Centre for Agricultural Research in the Dry Areas (ICARDA) at Aleppo in Syria.
† Based on experience and field observations at ICARDA (Tel Hadya Research Farm; 36° 10′ N).

Plant husbandry and culture

Techniques were based on those developed for tropical grain legumes in general (Summerfield, Huxley and Minchin, 1977) and subsequently modified for lentils as described in detail elsewhere (Summerfield and Muehlbauer, 1982). Seeds of all accessions were weighed individually and only those from the median range of each stock were used (Table 1). They were immersed in 50 per cent ethanol for 30 s, surface sterilised for 2 min in 0·2 per cent mercuric chloride in 0·01 M HCl and washed three times in sterile distilled water. Seeds of many *microsperma* (small-seeded), land-races and, depending on the time of harvest and conditions during storage, even those of improved cultivars may imbibe only poorly unless scarified (Summerfield, 1981). Because of this, the graded stocks were soaked in aerated distilled water at room temperature and only those seeds which were plump after 4 h (the typical duration to maximum imbibition, determined by preliminary trials) were selected. One half of each batch of seeds was blotted dry and inoculated with strain of *Rhizobium* SL 13 – a broadly effective strain from the ICARDA collection – using standard procedures (Summerfield *et al.*, 1977). The remaining batches of seeds were placed on moist filter paper in 9 cm diameter Petri dishes and vernalized for 30 d in the dark at $1{\cdot}5 \pm 0{\cdot}5$ °C. Based on experience elsewhere (reviewed by Summerfield, 1981) this chilling treatment was expected to be effective for those genotypes responsive to vernalization.

The non-vernalized seeds were sown immediately following inoculation, three to each 18 cm diameter plastic pot containing a mixture of vermiculite, sand, gravel and loamless peat compost without fertilizer (4:2:4:1, by volume) and soaked 24 h beforehand with deionized water. After sowing, the seeds were covered with a 2 cm layer of granite grit. All components of the rooting medium had been steam-sterilised at 96 °C and 3 kg cm^{-2} for between 10 and 20 min.

After the lapse of 30 d, the vernalized seeds were inoculated with *Rhizobium* and sown in an identical manner to those planted earlier (i.e. on 7 September and 9 August 1983, respectively).

Pots were not irrigated until 10 d after sowing, when seedlings in all photo-thermal regimes were well-established. Thereafter, sufficient nutrient solution, complete except for containing only 20 mg l^{-1} inorganic nitrogen (Summerfield *et al.*, 1977), was delivered automatically four times each day so that drainage through the pots occurred at every application. At this concentration of applied nitrogen we expect that lentils will derive

a large proportion of their nitrogen from symbiotic dinitrogen fixation, but this has yet to be confirmed.

Four replicate pots of both vernalized and non-vernalized seeds of each accession were sown in each environment. Seedlings were thinned after 10–20 d to leave single uniform plants in each pot. At thinning, all pots were given a surface dusting of Captan, as an effective precautionary measure against seedling pathogens which may lead to 'damping-off'.

Records were made on individual plants of the time (d) from sowing until the appearance of the first open flower (corolla colour visible).

RESULTS

General comments

When diverse genotypes are grown in a wide range of photo-thermal conditions, there is a considerable difference in time to flowering between treatments. Consequently, a decision to terminate the experiment is a compromise between the need to collect as complete a set of data as possible and the economics of maintaining artificial environments for a small number of plants of particularly sensitive genotypes in the least inductive regime(s). On this occasion, the experiment was terminated after 140 d from sowing the non-vernalized seeds (i.e. 110 d from sowing the vernalized ones), by which time all replicates of 91 per cent of the 144 'treatment' combinations (i.e. six genotypes, vernalized or not, in each of 12 environments) had come into flower.

TABLE 2. *Time (d) from sowing to first flowering in diverse lentil genotypes in the most- and least-inductive treatment combinations*

	Days from sowing to first flowering in different treatments*			
	Most inductive†		Least inductive†	
Genotype	−V	+V	−V	+V
ILL 4605	34	27 (20·6)‡	83·7	69·8 (16·6)
ILL 9	51	32·5 (36·1)	> 140	> 110 (?)
ILL 1744	50·5	34·3 (32·1)	120·3	> 110 (< 8·6)
ILL 2501	51·5	35·5 (31·1)	130·3	> 110 (< 15·6)
ILL 4349	66	44·8 (34·8)	111	101 (9)
ILL 4400	72	45·3 (37·1)	> 140	> 110 (?)

* Mean values of four replicates.

† For all genotypes +V and −V denote vernalized and non-vernalized seeds, respectively; and the most- and least-inductive growing conditions were 16 h/28–13 °C ($\bar{t}$ = 21·75 °C) and 10 h/18–5 °C ($\bar{t}$ = 9·33 °C), respectively.

‡ Figures in parentheses are the relative hastening in days to flowering (per cent) due to vernalization.

Without exception, seed vernalization and subsequent photo-thermal regime all significantly affected the number of days to flowering. Although genotypes differed in *relative* sensitivity to each of the factors investigated, they all flowered soonest when grown from vernalized seeds in the regime in which the longest day was combined with the warmest temperature; conversely, flowering was most delayed when plants grown from non-vernalized seeds experienced the combination of the shortest day with the coolest temperature (Table 2). Irrespective of genotype, photoperiod or vernalization,

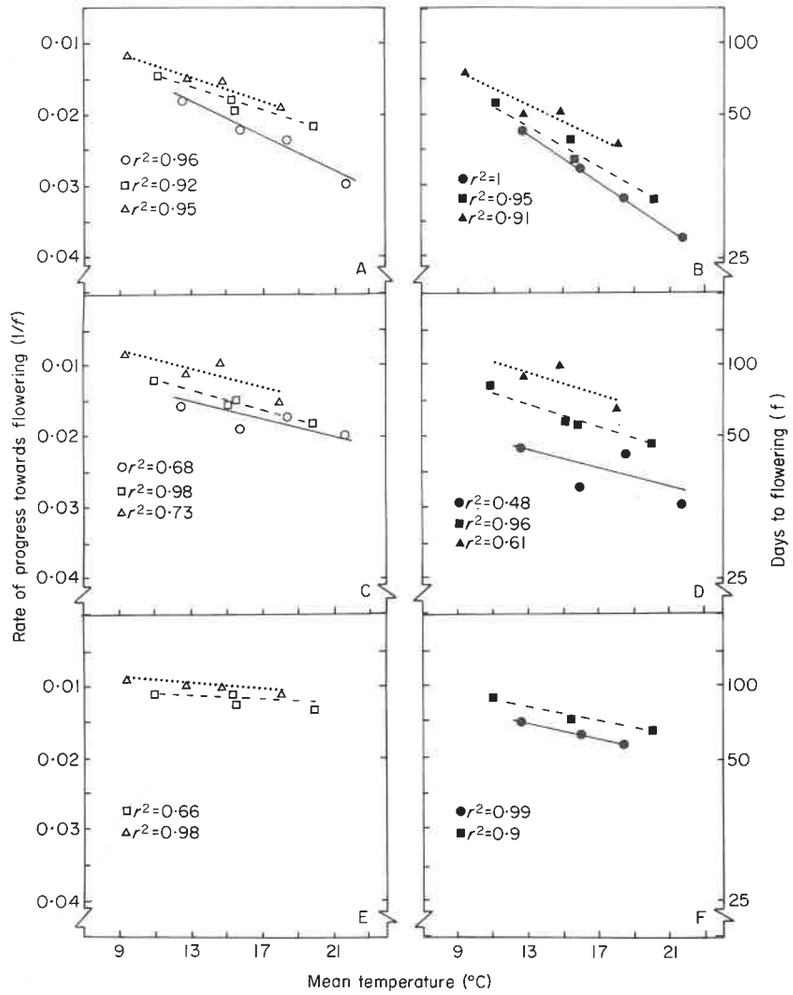

FIG. 1. Effects of mean temperature ($\bar{t}$ °C) and photoperiod (h) on rates of progress towards flowering ($1/f$) and days to first flower (f) in selected lentil genotypes grown from nonvernalized (−V) or vernalized (+V) seeds. (A, B) the early-maturing Argentinian cultivar 'Precoz', designated ILL 4605; (C, D) an early maturing land-race from Ethiopia, ILL 1744; and (E, F) the late-maturing Canadian cultivar 'Laird', ILL 4349. The symbols ○, □ and △ (−V) and ●, ■ and ▲ (+V) denote the experimental mean values in different thermal regimes in photoperiods of 16, 13 and 10 h, respectively, with fitted regression lines (——, - - -, and ·····, respectively) as described by Eqn (1).

time to flowering was obviously modulated by mean diurnal temperatures rather than by any specific effect of day or night temperature or by any diurnal difference in temperature.

Genotypes differed inherently in time to flowering in the most inductive conditions (i.e. in their relative earliness or 'flowering tendency'; Wallace, 1985), from the Argentinian cultivar Precoz (34 d) to the Syrian land-race (72 d); these two genotypes were also the least and most sensitive, respectively, to vernalization in this most inductive

TABLE 3. *Values of the constants in Eqns (4) and (5) derived from regressions of the inverse durations (days from sowing) to first open flower (1/f) against mean temperature ($\bar{t}$ °C) and photoperiod (h) for each of six lentil genotypes grown from either non-vernalized (−V) or vernalized (+V) seeds*

Genotype	Environments fitted*	Vernalization status	Fitted parameter values				
			a	*b*	*c*	*d*	r^2
Precoz (ILL 4605)	12	−V	−0·0052226	+0·00093643	+0·00075104	—	0·96
	12	−V	+0·0101590	−0·00008401	−0·00044067	+0·000077692	0·97
	12	+V	−0·0075941	+0·00146190	+0·00075104	—	0·96
	12	+V	+0·0077875	+0·00044144	−0·00044067	+0·000077692	0·97
Pant-L-406 (ILL 2501)	12	−V	−0·0086030	+0·00019484	+0·0014637	—	0·91
	12	−V	−0·0039023	−0·00010325	+0·0010940	+0·000023015	0·92
	10	+V	−0·0240300	+0·00058480	+0·0023845	—	0·91
	10	+V	−0·0054737	−0·00051361	+0·0010940	+0·000076200	0·92
EL79 (ILL 1744)	12	−V	−0·0047545	+0·00068262	+0·0006950	NS	0·87
	11	+V	−0·0185025	+0·00068262	+0·0019835	NS	0·87
78S-26004 (ILL 9)	11	−V	−0·0057408	+0·00020113	+0·0012292	NS	0·89
	10	+V	−0·0196948	+0·00078441	+0·0019110	NS	0·89
Laird (ILL 4349)	10	−V	+0·0014689	+0·00030622	+0·00044640	NS	0·90
	7	+V	+0·0015094	+0·00030622	+0·00085502	NS	0·90
Syrian local large (ILL 4400)	8	−V	−0·002918	NS	+0·0010093	NS	0·56
	8	+V	−0·020910	+0·00045813	+0·0020210	NS	0·77

* Poor germination and/or emergence of Laird and Syrian local large ± failure to flower (as in other genotypes) within duration of experiment (i.e. $f = > 110$ or > 140 d from sowing in +V (vernalized) and −V (non-vernalized) treatments, respectively).

NS, not significant; r^2 is the coefficient of variation for each fitted regression.

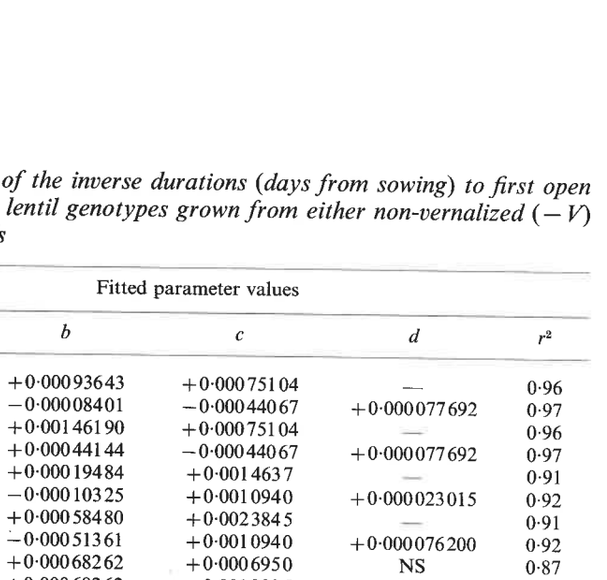

FIG. 2. Effects of mean diurnal temperature ($\bar{t}$ °C) and photoperiod (h) on rates of progess towards flowering ($1/f$) and days to flowering (f) in lentil ILL 4605 grown from non-vernalized (——) or vernalized (------) seeds. Symbols are the experimental mean values in each of 12 environments (as in Fig. 1). Each response surface has been calculated using appropriate constants (Table 3) in Eqn (4).

growth environment, although the relative differences between all genotypes other than Precoz were small (Table 2). Where data are complete (i.e. plants flowered in all regimes before the experiment was terminated), they show that the relative effect of vernalization on the number of days to flower (f) is less in less-inductive photo-thermal environments (Table 2).

Responses to temperature and photoperiod

When grown in the most, intermediate or least-inductive photoperiod (16, 13 and 10 h days, respectively) all genotypes, both cultivars and land-races, responded in a similar manner to temperature, i.e. their rate of progress towards flowering ($1/f$), the reciprocal of the time taken to flower, was a linear function of mean temperature ($\bar{t}$). Thus, as shown in Fig. 1, the flowering response to temperature *in any given photoperiod* can be described by the equation:

$$1/f = a + b\bar{t} \qquad (1)$$

where f denotes the number of days from sowing to the onset of flowering, $\bar{t}$ is the mean temperature (°C) and a and b are constants.

The thermal sum relation described by Eqn (1) infers that at any fixed photoperiod flowering will occur when a constant number of thermal units, T_f (day degrees above a base temperature of t_o), have accumulated after sowing. The value of the theoretical base temperature can be calculated from the constants in Eqn (1) using the expression:

$$t_o = -a/b \qquad (2)$$

Furthermore, the thermal sum required for flowering is given by:

$$T_f = 1/b \tag{3}$$

Seed vernalization consistently increased both the value of the theoretical base temperature for flowering within a given photoperiod (e.g. from the range −1·71 to −5·86 °C to the range −0·58 to −3·88 °C in ILL 4605; by extrapolations of Fig. 1A, B) and the value of the slope constant b in Eqn (1) – see Fig. 1A–F. However, as we consider later (see Discussion), the values of the theoretical base temperatures for flowering in species responsive to vernalization, such as lentils, may not have the same biological significance as base temperatures for other plant processes (e.g. germination or leaf initiation; Monteith, 1977), or as base temperatures for flowering in species or genotypes unresponsive to vernalization or to photoperiod (e.g. Hadley *et al.*, 1983, 1984). Then again, an increase in the value of the slope constant b does not necessarily mean that time (d) to flowering *per se* is more sensitive to post-vernalization temperatures in plants grown from vernalized seeds, as we also discuss later.

There is no evidence from Fig. 1 (or for the three remaining genotypes tested here; Table 1) of a critical photoperiod over the range 10–16 h (i.e. throughout the entire range of daylengths experienced by lentil crops) or of any *strong* interactions between photoperiod and post-vernalization mean temperature on rates of progress towards flowering. Nevertheless, since the possibility of significant interactions (in statistical as well as agronomic terms) could not be excluded with complete confidence (e.g. see Fig. 1A, B), the relative reliability of two algebraic relations for describing the overall effects of mean temperature and photoperiod on flowering in lentils was tested. These two models are described by the equations:

$$1/f = a + b\bar{t} + cp \tag{4}$$

and

$$1/f = a + b\bar{t} + cp + d(\bar{t} \times p) \tag{5}$$

where $\bar{t}$ is mean temperature, p is the photoperiod (h), and a, b, c and d are constants. The outcome of fitting these equations to the flowering data for individual genotypes is shown in detail in Table 3. And, as a typical example, Eqn (4) is fitted to the data for ILL 4605 in Fig. 2.

It is clear from Table 3 that the inclusion of an interaction term where statistically significant, as in Eqn (5), contributes little to improve the precision of the fitted regressions. In the four cases (out of a total of 24) in which the inclusion of an interaction term was justified, it contributed a maximum of only an additional 1 per cent to explaining the variation already accounted for if Eqn (4) were used instead. Since Eqn (4) is simpler and can be applied to all six genotypes, it is argued that it provides the best description for lentils – as it has proved to be for chickpea, the other long-day grain legume in which the flowering response has been thoroughly investigated with respect to temperature at different photoperiods (Roberts *et al.*, 1985).

The hastening of flowering by the single vernalization treatment imposed here could not be accommodated in any simple manner into Eqn (4) and so separate sets of regression constants were calculated for plants grown from vernalized seeds (Table 3). It remains to be seen if the vernalization effect can be included in a subsequent revision of Eqn (4) – and data from an investigation designed to test this possibility are being collected.

Genotypes differed considerably in their relative sensitivity to both photoperiod and temperature (Tables 2 and 3) but the differences in relative 'earliness' and sensitivity to temperature were not paralleled by similar differences in relative responsiveness to photoperiod (Table 4). This fact, together with the absence of any strong interaction between these environmental parameters, suggests that while the responses to temperature and photoperiod affect the same phenological event (i.e. time of flowering), they are under separate genetic control.

TABLE 4. *Time (d) to first flower at mean temperatures of 9 and 22 °C combined with a photoperiod of either 10 or 16 h (calculated from Eqn 4) and the resultant ranking according to earliness and sensitivity to temperature and photoperiod in six lentil genotypes*

Genotype and vernalization status	Time to flowering (d) (Ranking according to earliness in parentheses)*				Ranking of relative sensitivity to temperature and photoperiod			
					Temperature		Photoperiod	
	9 °C/10 h	22 °C/10 h	9 °C/16 h	22 °C/16 h	10 h	16 h	9 °C	22 °C
ILL 4605 −V	92·9 } (1)	43·7 } (1)	65·7 (3)	36·5 } (1)	6	5	2	1 }
ILL 4605 +V	76·5 }	31·2 }	56·9 (5)	27·3 }	2	6	1	1 }
ILL 2501 −V	128·5 } (5)	97·0 } (5)	60·4 } (1)	52·4 (3)	3	2 }	6	5 }
ILL 2501 +V	196·9 }	78·0 }	51·5 }	37·1 (4)	5	2 }	5	5 }
ILL 1744 −V	119·9 (4)	58·1 (2)	79·9 (5)	46·8 } (2)	5	6	3 }	2
ILL 1744 +V	133·8 (3)	61·2 (4)	51·7 (2)	35·4 }	3	3	3 }	4
ILL 9 −V	119·7 (3)	91·1 (4)	63·5 (2)	54·5 (4)	2	3	4 }	4
ILL 9 +V	154·4 (4)	60·0 (3)	55·5 (3)	35·5 (3)	4	5	4 }	3
ILL 4349 −V	115·3 } (2)	79·0 (3)	88·2 (6)	65·3 } (5)	4	4	1	3
ILL 4349 +V	78·0 }	59·5 (2)	55·8 (4)	45·6 }	1	1	2	2
ILL 4400 −V	139·3 } (6)	139·3 } (6)	75·6 (4)	75·6 } (6)	1	1	5	6 }
ILL 4400 +V	292·1 }	106·5 }	64·3 (6)	46·5 }	6	4	6	6 }

* Earliness and relative sensitivity to temperature or photoperiod are ranked from 1, earliest/least sensitive to 6, latest/most sensitive; relative sensitivity depends on the number of days alteration in time to first flowering due to the environmental factor considered. Plants grown from non-vernalized (−V) or vernalized (+V) seeds (and −V and +V treatments are ranked separately). Where time to flowering exceed either 110 (+V) or 140 d (−V) then values are extrapolations.

DISCUSSION

The results analysed here show that a relatively simple equation provides a satisfactory model for describing the effects of wide ranges of photo-thermal conditions on rates of progress towards flowering in diverse genotypes of lentil. The model is identical to that found to apply to another long-day grain legume, chickpea (*Cicer arietinum* L.) (Roberts *et al.*, 1985). The responses of short-day grain legumes can also be described by simple equations (Hadley *et al.*, 1983, 1984): though more complicated, they show some similarities with the long-day response described here. In all species investigated, including lentils, there has been considerable variation in the relative responsiveness of genotypes to photoperiod *and* temperature (from almost neutral to acutely sensitive) and also in their relative earliness in the most inductive regimes. It is now clear that previous attempts to understand the modulation of flowering by environmental and genetic factors in lentils (see the reviews by Summerfield, 1981 and Summerfield *et al.*, 1985) have been inadequate.

As discussed previously for chickpea (Roberts *et al.*, 1985), the values of the constants b and c in Eqn (4) provide a direct measure of the relative responsiveness of rates of

TABLE 5. *A comparison of predicted durations from sowing to first flowering (d) based on Eqn 4 with those observed for six lentil genotypes grown in the field in Northern Italy**

Genotype	Time from sowing to first flowering (d)				
	Recorded in the field	Calculated using Eqn (4) and various time sequences of field temperature data†			
		1	2	3	4
ILL 9	41	41·4	42·6	59·2	59·8
ILL 1744	41	40·7	41·7	54·1	56·0
ILL 2501	37	42·5	43·5	57·0	57·6
ILL 4349	47	49·7	50·2	72·6	73·8
ILL 4400	44	54·2	55·3	54·2	54·2
ILL 4605	45	33·2	34·5	43·3	44·7

* Sown on 27 June 1984 at Leonessa, Italy (925 m OD; 42 ° 34′ N).

† 1, 2 here the constants calculated for +V plants in Eqn (4) have been used (Table 3), with $\bar{t}$ °C [(Σ(day maximum+night minimum)/2)/days to first flower] calculated from emergence or sowing, respectively; 3, 4 as for 1 and 2, respectively, except that constants for −V plants (Table 3) have been used in Eqn (4). A common average photoperiod (p) of 15·4 h (Francis, 1972) was used throughout. Numbers underlined within body of table are the most accurate predictions – except for the Syrian land-race ILL 4400 for which reliable predictions were not expected (see text).

progress towards flowering in different genotypes to temperature and photoperiod, respectively. But, since these constants relate to reciprocals of the times taken to flower, an early flowering genotype with a relatively large value of b or c may be less responsive to mean temperature or photoperiod in terms of changes in the number of days to flowering per unit change in either environmental factor than a later flowering genotype with a relatively small b or c value. This is because a unit change in $1/f$ gives a much greater change in f, the larger the initial f value. A simple expedient to overcome this potential difficulty is to evaluate Eqn (4) for individual genotypes over specific values of $\bar{t}$ and p, as has been done in Table 4 using the extremes of temperature and photoperiod involved in this investigation.

Relatively few data were available for the land-race ILL 4400 (Table 3) which is likely to be a genetically heterogeneous seed stock, and so the regression constants calculated for this genotype are unreliable. However, it is clear from Table 4 that other genotypes do not necessarily have identical rankings with respect to either their earliness or relative sensitivity to temperature and/or photoperiod over a wide range of photo-thermal conditions, or in their responsiveness to vernalization in more or less inductive post-vernalization regimes. In other species, vernalization effects are said to depend in a complex manner on both the temperature and duration of the treatment imposed (e.g. Hillman, 1969); responsiveness to vernalization can be governed by the same or different genes to those which govern daylength sensitivity (Napp-Zinn, 1984); and long-day species, when vernalized, can come into flower earlier or later than their nonvernalized counterparts when grown in less-inductive shorter days (Ruiter and Taylor, 1979).

In those short-day grain legumes unresponsive to vernalization which we have investigated hitherto (e.g. Hadley *et al.*, 1983, 1984), the values of the base temperature for different genotypes, calculated using Eqn (2), have been clearly related to the origins of the genotypes tested. For example, soyabean genotypes of tropical origin have considerably warmer base temperatures for flowering than those from temperate regions (about 10 °C and 4–6 °C, respectively; Summerfield and Roberts, 1985). However, when

all genotypes show a response to both temperature and photoperiod of the type described by Eqn (4) it is doubtful whether the concept of a base temperature for flowering has any physiological significance because, as discussed previously for chickpea (Roberts *et al.*, 1985), the value of the base temperature varies with photoperiod according to the equation:

$$t_o = (-a\text{-}cp)/b, \tag{6}$$

where the values of a, b and c are those calculated using Eqn (4). Furthermore, it is clear from the results presented here that vernalization also apparently alters t_o through its effect on the values of a, b and c (Table 3). Finally, under some conditions, the calculated base temperature is sometimes extreme (e.g. for ILL 2501 unvernalized in a 10 h photoperiod the calculated value of t_o is −50·9 °C), and outside the range which could be considered physiologically meaningful.

Ongoing research, in both controlled environments and in the field, has the objective of advancing further our understanding of the consequences and predictability of vernalization effects on flowering in lentils (and also the genetic control of these effects and post-vernalization responses in general). However, application of Eqn (4) and the constants derived from this investigation in growth cabinets to the field (within the wide ranges of each of photoperiod and mean temperature investigated) should prove reliable. This assertion was tested by a comparison of predicted and actual times to flowering of these six genotypes in the field in Northern Italy. As Table 5 shows, Eqn (4) and the constants calculated in cabinets (Table 3) gave precise predictions of times to flowering – at least under the relatively inductive photo-thermal conditions prevailing during June–August at that field location.

It is also clear from Table 5 that proper account must be taken of the relative responsiveness of different genotypes to vernalization if field predictions based on Eqn (4) are to prove reliable. For Precoz (ILL 4605), the least responsive genotype of those tested here to vernalization when grown in strongly inductive photo-thermal conditions (Table 2), the constants in Eqn (4) calculated for non-vernalized (−V) plants (Table 3) proved outstandingly better for predictive purposes than those calculated for vernalized ones. In comparison, for the remaining genotypes – which were all more sensitive to vernalization than Precoz when grown in strongly inductive photo-thermal conditions (Table 2) – the constants calculated from the responses of vernalized plants (+V) in artificial climates (Table 3) were the ones most reliable for predicting field behaviour accurately (Table 5). Pre-flowering, night-time minimum *air* temperatures during the field trial varied between about 5 and 15 °C, with an average close to 10 °C. We are currently investigating in controlled environments the effects of lentils of 'vernalization' treatments which cover most of this range (−5 to +10 °C for 15–45 d) on subsequent flowering of plants grown in long or short days.

One objective of our research programme has been to develop techniques for screening large numbers of genotypes which may be more or less sensitive to photoperiod and temperature and which, therefore, are likely to be well-adapted to either a relatively few or wide range of environments, respectively (e.g. Hadley *et al.*, 1983, 1984; Roberts *et al.*, 1985). With lentils, since the post-vernalization photoperiod-temperature response surface is a single plane (Fig. 2) defined by Eqn (4), then the minimum number of environments required to establish it is three – providing these include two photoperiods *and* two mean temperatures. Results obtained from this trio of environmental conditions would establish the magnitude of both the photoperiodic and temperature responses as well as the relative earliness of the genotypes tested. On the basis of current data, and the traditional and potential areas of lentil crop cultivation world-wide (Muehlbauer *et al.*, 1985), it would seem appropriate for this purpose to use an 11 h and a 16 h photoperiod and a combination of day and night temperatures which will give a mean

temperature of 10 °C or 22 °C. But, until we can better quantify the effects of vernalization (in terms of temperature and duration) on subsequent development, any recommendations on screening germplasm for responsiveness to this factor must be provisional. In the interim, the treatment used here (exposing imbibed seeds to $1{\cdot}5 \pm 0{\cdot}5$ °C for 30 d) could be used so that both vernalized and non-vernalized seeds are sown in all three environments. If geographical or political constraints restrict accessibility to suitable field sites in those regions where lentils are an important food legume (Muehlbauer *et al.*, 1985), then the screen proposed here could be carried out successfully in glasshouses – providing that photo-thermal controls in such facilities are precise and reliable (e.g. Hadley *et al.*, 1983, 1984; Roberts *et al.*, 1985).

The photo-thermal responsiveness of lentil germplasm originating from the Mediterranean region limits the dissemination of these materials into Asia, and especially into the Indian subcontinent – where most of the world's lentils are grown. In general, the Mediterranean types flower late in India and produce only a few fruits as they undergo 'forced maturation' in conditions of rapidly increasing heat and aridity (Erskine and Hawtin, 1983). Furthermore, it is difficult to synchronize the flowering periods of the imported genotypes with those of local types for hybridization purposes (Tyagi and Sharma, 1981). We have suggested recently a strategy of growing diverse genotypes along an illuminance gradient in the field to overcome the latter problem (Summerfield, Muehlbauer and Roberts, 1984). We now suggest that the relations described here could lead to simple field screening techniques which should facilitate the search for exotic germplasm potentially well-adapted to the major Indian production zone, where yields are traditionally lamentably small (Summerfield, 1981), either for direct introduction or for parents in a hybridization programme. When supplemented with suitable parameters which quantify the consequences of vernalization more fully (i.e. the durations and severity of cold), the suggestions made here could be further developed and should prove even more useful in the search for lentil genotypes well adapted to the environments for which they are intended.

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