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Temperature Affects Growth and Flowering of the Balloon Flower [*Platycodon grandiflorus* (Jacq.) A. DC. cv. Astra Blue]

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Abstract. **The effects of temperature from 14 to 29 °C on the growth and flowering of *Platycodon grandiflorus* (Jacq.) A. DC. cv. Astra Blue are described. Under greenhouse conditions, seedlings flowered earliest (after 45 days) when grown at 29 °C, vs. after 108 days when grown at 14 °C. Height at flowering was greatest at 19 °C and branch number was greatest (13 per plant) at 22 °C. Fresh and dry mass and leaf area at flowering increased with decreasing temperature. Imbibing seeds at 4 °C prior to germination had no significant affect on the time to flowering or the number of nodes below the flower. In a growth chamber experiment, apical dissections showed that flowers were initiated earlier at 20 °C than at 10 °C. These results show that the balloon flower is highly sensitive to temperature, but cool temperatures delay the time to flower initiation or flowering.**

Platycodon is receiving considerable attention from growers for production and sale as a herbaceous perennial, cut flower (Armitage, 1993), or pot plant. However, little is known regarding its responses to environmental variables. Many growers believe that the long time from seedling to flowering may reduce the commercial potential of this crop. Therefore, techniques to advance flowering, or an understanding of factors affecting it, would have considerable value.

To date, most experimentation on *Platycodon* has focused on its use as a cut flower. Iversen and Weiler (1994) conducted experiments with overwintered crowns and showed that a period of cool temperatures was necessary to break crown dormancy, and that plants appeared to be day-neutral thereafter. However, the effect of chilling temperatures on flower induction was not clear. Iles and Agnew (1995) investigated flowering in *P. grandiflorus* 'Sentimental Blue' after transferring plants overwintered under a thermal blanket to a 21 °C glasshouse at regular intervals. Temperatures under the thermal blanket were as low as –25 °C. Time to flowering following transfer to a warm glasshouse decreased with each subsequent transfer date, suggesting that flowering may have been hastened by prolonged exposure to cold temperatures. This suggests a role of cool temperatures in the flowering of this plant, although the evidence is not conclusive.

Here we report how temperature affects flowering in *P. grandiflorus* 'Astra Blue'. Emphasis was placed on determining 1) whether cool temperatures promote flowering; 2) whether cool treatments can be applied to imbibed seed (as shown on many other species that flower earlier in response to cool temperatures, see Purvis, 1961); and 3) the optimal temperatures for the production of the crop as a pot plant.

Materials and Methods

Experiment 1. Effects of temperature on time to flowering and plant growth. Seed of *P. grandiflorus* 'Astra Blue' were sown in a standard P84 (volume per cell = 40 mL; Plantpak, Maldon, U.K.) plug tray containing a peat-based compost (SHL; William Sinclair Horticulture, Lincoln, U.K.) on 18 Mar. 1996 and germinated in a greenhouse compartment maintained at a constant day/night set point temperature of 22 ± 1 °C. After 29 d, the seedlings were potted in 9-cm (0.37-L) square pots containing a 2 vermiculite : 1 sand : 2 gravel mix (VSG). Ten plants (with one pair of true leaves) were then placed in each of six temperature-controlled greenhouses, which comprised the inner six of eight compartments (each having dimensions of 3.7 × 7 m). The two coolest compartments were equipped with air-conditioning units to maintain temperatures throughout the experimental period. Temperatures within the compartments were recorded with a data logger (Datataker DT500; Data Electronics, Letchworth, U.K.; scanned every 15 s, recording hourly averages) using aspirated PT100 temperature sensors. The actual mean temperatures for the five highest temperature treatments were 13.7, 18.9, 21.9, 24.7, and 28.9 °C; no data were gathered for the coolest regime, since plants died prior to flowering. The outside light integral were measured with a pyranometer, and the average over the first 60 d of the experiment was 25.6 $mol \cdot m^{-2} \cdot d^{-1}$. The photoperiod during the experimental period ranged from 13.8 to 15.6 $h \cdot day^{-1}$. Plants were watered via a drip irrigation system with a nutrient solution containing Sangral 1:1:1 (William Sinclair Horticulture) liquid feed diluted to a conductivity of 1500 µS (N, P, K of 182, 78, and 150 $mg \cdot L^{-1}$, respectively, plus micronutrients) and maintained at a pH of 5.8.

Plants reaching anthesis were recorded each day. At anthesis, 10 plants were harvested to assess fresh and dry mass (excluding roots), leaf area, branch numbers, and leaf number on the main stem below the flower. Data were analyzed using linear or multiple linear regression analysis; only linear or quadratic terms

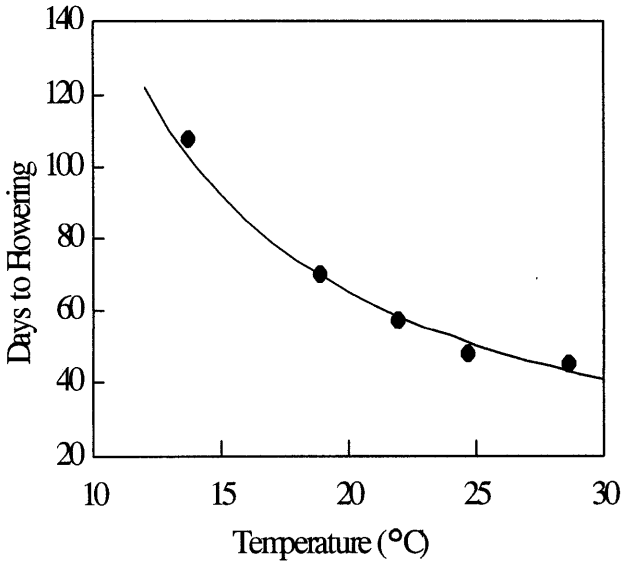

Fig. 1. The effects of temperature on time to flowering of *Platycodon grandiflorus* 'Astra Blue' from the commencement of the experimental treatments. Each point is the mean value for 10 replicate plants. The curve was fitted by linear regression where days to flowering = $1/(-0.00262 + 0.0009T)$, T is mean temperature and $r^2 = 0.97$, 3 df.

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with a level of significance >0.05 were considered. Regressions were performed on data from individual replicates, not mean values for the treatments.

Experiment 2. Effects of seed vernalization on time to flowering. This experiment was conducted to assess whether seed could be vernalized to advance time to final flowering. The vernalization treatments were conducted with imbibed seed (on moist filter paper inside petri dishes) in an incubator set at 4 °C for 0, 4, 8, 12, 20, 24, or 28 d. The dates when chilling commenced were staggered so that all treatments ended on the same date (30 Apr. 1996). Seed were then germinated in a P84 plug tray containing a peat-based compost (SHL) inside a greenhouse maintained at 22 °C. After 36 d, 10 seedlings (with one to two pairs of true leaves) from each treatment were randomly selected and transferred to 9-cm pots containing VSG and grown on until flowering inside a greenhouse compartment maintained at 18 °C, according to the protocol described above.

Experiment 3. Effects of temperature on time to flower initiation. Experiment 1 examined the effects of temperature on time to flowering from seed sowing and did not discriminate between effects of temperature on time to flower initiation vs. subsequent flower development. To examine whether cool temperatures advanced time to flower initiation, a third experiment was conducted using repeated apical dissections.

Seed of *P. grandiflorus* 'Astra Blue' were sown on 18 Mar. 1996 in P84 plug trays containing the same compost as in Expt. 1 and germinated at a temperature of 22 °C. After 29 d, plants (one pair of true leaves) were transferred into 9-cm pots containing VSG. Sixty plants were placed in each of two identical growth rooms, one set at a constant 10 ± 0.5 °C and the second at 22 ± 0.5 °C. A temperature of 10 °C is considered to be sufficiently cool to induce "vernalization" responses in a range of species (Bernier et al., 1981; Purvis, 1961). To examine whether flower initiation could be promoted by an initial period of cold, followed by warm temperatures in long days, i.e., following a typical classical vernalization response (Vince-Prue, 1975), an additional 30 plants were transferred from the 10 °C room to the 22 °C room after 21 d. The light level inside the room was maintained at 90 mol·m^{-2}·s^{-1} for a 16-h day from warm white fluorescent lamps (93.7% intensity) plus tungsten bulbs (6.3% intensity, as calculated on the basis of nominal wattage). Plants were irrigated with the same nutrient solution used in Expt. 1.

At the start of the treatments and every 3 d thereafter, apices of three plants from each treatment were fixed for examination under a scanning electron microscope (SEM). Dissected apices were fixed in glutaraldehyde 4% (v/v) in 0.1 M phosphate buffer (pH 7) for 2 h in sealed bottles at room temperature. The fixative was decanted off and the material washed with three changes of glass distilled water. The material was then fixed in 1% (m/v) aqueous osmium tetroxide (OsO_4) for 2 h and again washed with three changes of glass distilled water. The material was then dehydrated through a graded acetone series (30%, 50%, 70%, 90%, 95%, 100% followed by absolute 100%), the samples were in each acetone dilution for 30 min. The material for SEM was dried using the critical point drying technique, mounted on stubs using silver paint and coated with gold using a sputterer coater. Observations were conducted with a Jeol T20 (Tokyo, Japan) SEM at ≈20KV.

Results

Effects of temperature on time to flowering. Time to flowering from sowing was earliest ($P < 0.001$) at high temperatures (Fig. 1). As temperature decreased, time to flowering increased; plants required 45 d for flowering at 29 °C vs. 108 d at 14 °C. Regression analysis indicated a significant linear relationship ($r^2 = 0.97$, 3 df) between the reciprocal of time to flowering (1/f) and mean temperature (T), such that:

$$1/f = -0.00262 + 0.0009T$$

There was no significant effect of temperature on the number of nodes below the flower, although number of nodes in the 19 °C treatment was slightly greater (15 nodes) than in the other treatments, which averaged ≈13 to 14 nodes (Table 1). Morphology of *Platycodon* was very sensitive to temperature. Plant fresh and dry mass, as well as leaf area, were greatest at the coolest temperatures, such that fresh mass and leaf area were ≈5- to 2-fold as great at 13.7 than at 29 °C, respectively. Both parameters decreased curvilinearly with increasing temperature ($P < 0.001$). Height at flowering was greatest at 19 to 22 °C, and decreased with cooler or warmer temperatures. However, the responses were small, such that height at 19 °C was only 4.4 cm greater than at 29 °C. Flower bud number was also very sensitive to temperature with bud number at 14 °C being about twice that at 29 °C ($P < 0.001$). Branch number was greatest at 19 to 22 °C. Although sensitivity over the whole temperature range was small, plants at 29 °C had only two fewer branches than those at 22 °C. Flower diameter and mean branch length were not consistently affected by temperature (Table 1).

Effects of seed vernalization on time to flowering. Prechilling of the seed had no effect on subsequent time to flowering or on other growth parameters (e.g., fresh and dry mass, leaf number, etc., data not shown).

Effects of temperature on flower initiation. The leaves were initiated in sub-opposite pairs around an ovate meristem (Fig. 2A). As the meristem enlarged and began to initiate flowers, four floret primordia appeared in the corners of a rectangular apex. The meristem was symmetrical about two axes, orientated 90° from each other (Fig. 2B). These four primordia then developed to form the outer petals of the flower (Fig. 2C).

Apical dissections showed that plants at 22 °C initiated flowers 35 days from the start of the treatments, whereas plants at 10 °C, and those transferred from 10 to 22 °C, were still vegetative at the time of the last sampling (60 d after potting).

Table 1. The effects of temperature on plant height, fresh and dry mass, leaf area, branch number and length, flower diameter and number of flower buds of *Platycodon grandiflorus* cv. Astra Blue[z].

Temp (°C)	Plant ht (cm)	Fresh mass (g)	Dry mass (g)	Leaf area (cm^2)	Node[y] no.	Flower diam (cm)	Flower bud no.	Branch no. >1 cm	Avg branch length (cm)
13.7	12.8	27.0	5.1	1466	13	7.4	16	9	11.7
18.8	18.6	19.4	3.3	1430	15	6.8	11	12	16.5
21.9	18.1	11.8	2.1	1221	14	6.8	12	13	15.8
24.7	13.7	7.5	1.3	905	13	8.0	9	11	12.1
28.9	14.2	5.5	1.1	766	14	6.5	8	11	13.1
Constant[x]	–12.3	73.3	13.7	1795	NF	NF	21.7	–11.2	0
SE	(6.1)	(13.1)	(2.38)	(74)			(2.93)	(5.02)	(0)
Linear	2.89	–4.25	–0.78	NF	NF	NF	–0.49	2.16	1.51
SE	(0.66)	(1.27)	(0.23)				(0.13)	(0.48)	(0.10)
Quadratic	–0.07	0.061	0.012	–1.30	NF	NF	NF	–0.05	–0.037
SE	(0.016)	(0.03)	(0.005)	(0.14)				(0.011)	(0.004)
r^2	0.35	0.77	0.75	0.69	---	---	0.25	0.35	0.21
P	***	***	***	***	NS	NS	***	***	*

[z]Means are from 10 replicate plants.
[y]Below the flower.
[x]Data were analyzed via regression ($Y = a + bT + cT^2$). Only terms with a level of significance greater than the 0.05 level are stated. Regressions were performed using individual replicate values. NS represents not significant and NF not fitted, i.e. the term was not significant when used in a regression.
NS, *, ***Nonsignificant or significant at $P < 0.05$ or 0.001, respectively.

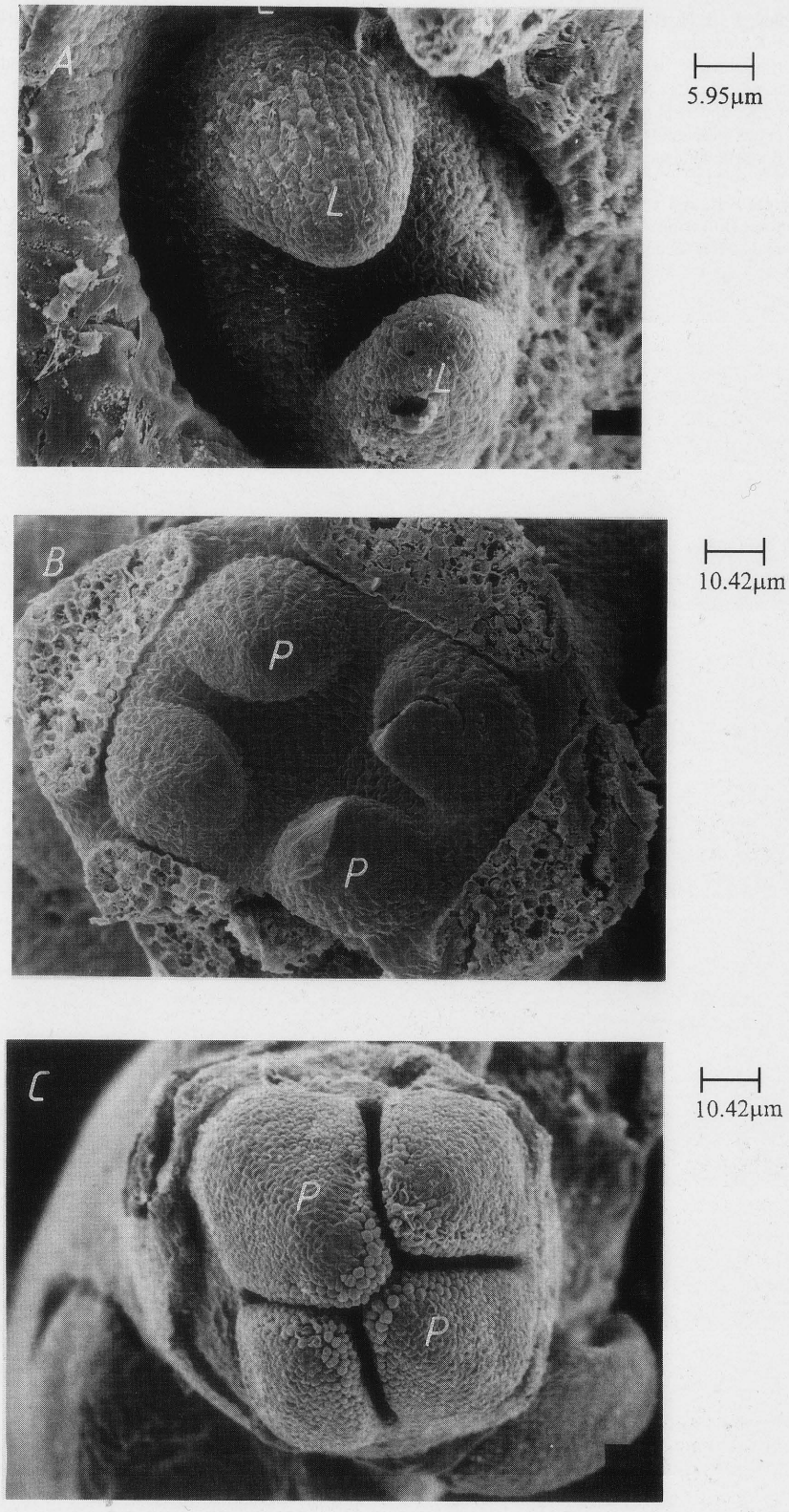

Fig. 2. Scanning electron micrographs of *Platycodon* apical meristems during the transition from vegetative development (**A**: mag. ×840), to flower initiation (**B**: mag. ×480), and development (**C**: mag. ×480). P and L designate floral and leaf primordia, respectively.

Discussion

In these experiments low temperatures delayed, rather than hastened, the time to flowering. The third experiment in particular suggested no evidence for a cold requirement, at least in this cultivar, since plants at 10 °C, and those subjected to 10 °C followed by 22 °C, did not initiate flowers during the course of the experiment. Arguably 10 °C is probably not as effective as 4 to 7 °C in terms of inducing a vernalization response, but the reviews of Purvis (1961) and Bernier et al. (1981) suggest that this temperature is usually sufficient to evoke a response to cold, if one exists. Given the lack of any cool temperature requirement for flowering, it was not surprising seed chilling had no effect on final time to flowering.

The lack of a response to cool temperatures noted here contrasts with Iles and Agnew's (1995) observations on *Platycodon*. These differences may be due to varietal differences or to experimental techniques used to examine vernalization responses. For instance, Iles and Agnew (1995) forced flowering by transferring plants from outdoors into a glasshouse compartment at different time intervals. This experimental approach has two potential deficiencies: the degree of chilling (or even devernalization) supplied is unknown, and, as plants are transferred to a glasshouse at different times, each successive batch will be of different chronological age and will have received a systematically varying exposure to light.

The delayed time to flowering at the cooler temperatures suggests little evidence of a cold requirement for flower induction. We have shown that *P. grandiflorus* 'Astra Blue' is highly sensitive to temperature in terms of time from seed sowing to flowering, and a relationship has been developed that growers may be able to use to forecast time to flowering at any given temperature. The linear relationship between *reciprocal* of time to flowering and temperature implies that there is a hyperbolic relationship between temperature and days to flowering. Hadley et al. (1983) showed that the reciprocal of the slope of the relationship between temperature and the reciprocal of days to flowering represents the thermal time required for flowering (1/0.0009), equal to 1110 degree-days. The base temperature can be determined by dividing the negative value of the intercept of the relationship by the slope (0.00262/0.0009), which here equalled 2.9 °C. However, prior to applying this "model," further work would be required to examine potential effects of variation in light integral.

Platycodon's sensitivity to temperature suggests that small changes in temperature may greatly influence production time. However, increasing temperature would be at the expense of final quality. Similar high temperature reductions in quality are known for a range of other crops (Armitage, 1994; Larson, 1992). For example, Kaczperski et al. (1991) showed in *Petunia ×hybrida* that branch number was greatly reduced as temperatures increased above 13 °C. Similarly, plant height tended to increase with average daily temperature.

In conclusion, we have shown that *P. grandiflorus* 'Astra Blue' is highly sensitive to temperature with no evidence of a vernalization response for flowering. Experimentation is now required to determine whether other environmental manipulations (e.g., CO_2 and supplementary lighting) can be used to advance and manipulate flowering and plant quality.

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