Effects of Seasonal Variation and Gibberellic Acid Treatment on the Growth and Development of *Gypsophila paniculata*

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Abstract. We investigated the effects of different planting seasons and gibberellic acid treatments on the growth and development of *Gypsophila paniculata* to explore new approaches to controlling the flowering period. Four different cultivars were selected and continually planted in July, September, and November in the low-latitude and high-altitude region of Kunming, China (25° N, 102° E). Results showed that the vegetative growth and flowering time of *Gypsophila paniculata* were prolonged and postponed when the planting time was delayed. Specifically, ‘My Pink’ showed 20% and 80% rosette rates when grown in autumn and winter, respectively, thus indicating that *Gypsophila paniculata* is sensitive to planting time. Moreover, GA₃ treatment not only can significantly promote vegetative growth but also can stimulate early flowering and suppress the occurrence of rosettes during winter. This is more specific to ‘My Pink’, which showed 40% and 80% reductions in rosette rates with four and eight GA₃ treatment applications, respectively. Our study showed that seasonal variations in the growth and development of *Gypsophila paniculata* and GA₃ treatment can effectively stimulate early flowering and suppress rosettes during winter.

Gypsophila paniculata, a flowering plant of the Caryophyllaceae family, is a small perennial herbaceous shrub ≈1 m tall and ≈1 m wide (Toaima et al., 2016; Vettori et al., 2015). It is normally characterized by a rosette of grayish green basal leaves and linear lanceolate from which the flower stems propagate and end in mounds of branching stems covered by clouds of tiny white or pink flowers (Ataşlar et al., 2009; Cronquist, 1981). *Gypsophila paniculata* is a famous ornamental plant that is considered one of the 10 best-selling cut flower species worldwide (Zvi et al., 2008). Because of its ornamental value, *Gypsophila paniculata* has been grown not only for flower arrangements but also as a perennial garden plant (Wang et al., 2013).

The flowering behavior of *Gypsophila paniculata* is strongly influenced by growth conditions, mainly daylength and temperature (Thakur et al., 2013). It has been reported that

plants will stay in the vegetative growth stage and form a rosette under short daylighting and lower temperatures (Nishidate et al., 2012). The formation of the rosette significantly affects the flower production of *Gypsophila paniculata* because these rosette plants cannot bloom. Therefore, for year-round production of *Gypsophila paniculata*, specific techniques such as artificial lighting and heating are used to provide optimal light and temperature conditions. Although the light can be replaced by an LED source, artificial heating is energy-consuming during winter. To overcome this serious limitation and to optimize flower production during winter, the application of growth regulators may offer an inexpensive and relatively simple method of enhancing flower production by regulating the growth and development of plants (Rameau et al., 2015). Gibberellic acid is considered essential for the development of stamens and petals (Elfving et al., 2011; Ni et al., 2015) because it uses the response of the plant reproductive development to environmental stresses, especially unfavorable temperature conditions (Liu et al., 2017; Wu et al., 2016). More specifically, under lower temperatures, gibberellin signaling produces the effects of warm temperatures, which are essential for the response to the long-day induction effects (López and Gonzalez, 2006). Therefore, gibberellic acid could promote the development of flowers and help plants bloom under low temperatures.

Kunming is the main production area of *Gypsophila paniculata* in China, with 28 years of cultivation history (Yang, 2012). Due to its location (altitude, 1900 m; latitude, 25° N), Kunming has the mildest climate in China (Liang et al., 2017). The perpetual spring-like weather provides ideal growth conditions for plants, but it is still difficult for *Gypsophila paniculata* to bloom during winter because of the low temperatures at night. Usually, the seedlings of *Gypsophila paniculata* are planted from March until the end of August in Kunming; then, they are harvested twice during the forthcoming year. If the seedlings are planted after September, then the rosette plants will appear. These rosette plants are more significant during winter. Few studies have focused on applying gibberellins to induce floral stems and provoke plant blooming in *Gypsophila paniculata*. Therefore, we examined the seasonal variations in the growth and development of four different cultivars of *Gypsophila paniculata*. Our results showed that *Gypsophila paniculata* is season-sensitive, and that some cultivars cannot flower during autumn and winter. To overcome this, we applied growth regulator GA₃ to control the growth and flowering of different cultivars. The study showed that two different GA₃ treatments have positive effects on the vegetative growth and flowering regulation of *Gypsophila paniculata* during winter, thus providing a simple method of enhancing flower production.

Materials and Methods

Plant material and growth conditions. Four commercial cultivars of *Gypsophila paniculata* (‘My Pink’, ‘Million Stars’, ‘Cloudstar 8’, and ‘Cloudstar 5’) (Fig. 1) were selected for this study. ‘My Pink’ and ‘Million Stars’ are the commercial cultivars that were introduced by Danziger, a company in Israel (Moshav Mishmar Hashiva). However, ‘Cloudstar 8’ and ‘Cloudstar 5’ are specifically bred based on the environmental conditions of Kunming. The plant seedlings were obtained from Yuxi Yunxing Biological Technology Co., Ltd. (Yunnan province, China), and they were separately planted in pots (diameter, 25 cm; height, 30 cm) filled with commercial potting medium (peat and perlite mixture). Then, all plants were grown in the greenhouse (without heating) during the natural photoperiod (Table 1) at the experimental farm of Yunnan Academy of Agricultural Sciences.

Planting season and GA₃ treatment. Seedlings were continually planted on 1 July, 1 Sept., and 1 Nov. to explore the seasonal variations in growth and development. Ten seedlings of each cultivar were grown during each month. Two different GA₃ treatments (four applications and eight applications) were applied to plants that were planted on 1 Nov.; four applications and eight applications of 125 mg/L GA₃ were sprayed during the entire growing season. The GA₃ treatments were started on 1 Dec. at a frequency

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of once per week, and each treatment was used for 10 individual plants.

Data collection and statistical analysis. During the flowering period, the growth parameters, such as plant height, crown width, flowering time, and plant rosette rate, of each treatment were measured using a ruler and vernier caliper. Then, the entire individual plant was collected to measure the fresh weight and dry weight using an electronic scale. These data were used to evaluate the effects of the planting season and GA₃ treatment on the growth and development of each cultivar. Data analysis and statistics were performed using Microsoft Excel 2016 (Microsoft Corporation) and Data Processing System (Hangzhou Ruifeng Information Technology Co., Ltd., China) (Tang and Zhang, 2013). A one-way analysis of variance with Tukey's honestly significant difference post hoc test was used for the mean comparison of

multiple samples at a significance level of 5% ($\alpha = 0.05$).

Results

Seasonal variations in growth and development for *Gypsophila paniculata*. We continually planted seedlings for 3 months to explore and simulate the effects of different planting seasons on the growth and development of *Gypsophila paniculata* during the natural photoperiod of a low-latitude and high-altitude region. In general, different planting seasons had a significant impact on the plant growth of four cultivars (Fig. 2). With the delayed planting time, the plant height significantly increased, but the crown width decreased. This reflects the fact that plants will continually undergo bolting and vegetative growth under low-temperature conditions; consequently, the flowering time

was delayed. Consistent with this, the fresh weight and dry weight exhibited the same increases, but the flowering time was postponed. From these results, we can conclude that *Gypsophila paniculata* is sensitive to the planting season, and that vegetative growth will be prolonged when planting is performed during autumn and winter. However, seasonal sensitivity varies in different cultivars. 'My Pink' was the most sensitive to the planting season, exhibiting rosette rates of 20% and 80% when planted in September and November, respectively. 'Million Stars' was less sensitive than 'My Pink', with a rosette rate of only 10% when it was planted in November. Moreover, two cultivars, 'Cloudstar 8' and 'Cloudstar 5', were not sensitive to the planting season, indicating that they have adapted to the environmental conditions of low-latitude and high-altitude regions; therefore, they could be planted during winter. These results suggest that different cultivars require different planting seasons, and that these planting seasons are determined by the genetic background of the cultivar.

During the natural photoperiod in Kunming, different planting seasons had significant influences on plant growth, such as extended vegetative growth and delayed flowering time. In the worst case, it caused plant rosettes for 'My Pink' and 'Million Stars', which was disastrous for flower producers. However, 'Cloudstar 8' and 'Cloudstar 5' were able to bloom and flower during the winter even though the flowering time was delayed. This could be acceptable to flower producers because the flower yield was also increased. Therefore, 'Cloudstar 8' and 'Cloudstar 5' could lead to more income for flower producers during winter, when the supply of other flowers is lacking.

Effects of GA₃ on the growth and development of *Gypsophila paniculata*. To overcome plant rosettes during autumn and winter, we applied the growth regulator GA₃ to the plants that were planted on 1 Nov. Compared with the control, the plants showed increased plant height, crown width, fresh weight, and dry weight with the increased spray applications (Fig. 3). This indicated that GA₃ could stimulate and promote vegetative growth under low-temperature conditions. More specifically, eight applications of GA₃ treatment not only had a greater impact on the fresh weight and dry weight of all cultivars but also could promote early flowering compared with the control. Four applications of GA₃ treatment suppressed biomass accumulation in 'Million Stars' and 'Cloudstar 5', possibly because fewer GA₃ treatments could promote early flowering and, consequently, reduce vegetative growth. Furthermore, GA₃ also suppressed the occurrence of rosettes in 'My Pink' and 'Million

Fig. 1. Four commercial cultivars of *Gypsophila paniculata* were used in this study: (A) *Gypsophila paniculata* 'My Pink'; (B) *Gypsophila paniculata* 'Million Stars'; (C) *Gypsophila paniculata* 'Cloudstar 8'; and (D) *Gypsophila paniculata* 'Cloudstar 5'. Each cultivar refers to a different phenotype. Bar = 1 cm.

Table 1. Average natural daylength and temperature of each month in Kunming.

Month	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
Daylength (h)	10.78	11.31	12.02	12.75	13.36	13.66	13.51	12.98	12.29	11.56	10.94	10.63
Temperature (°C)	10.70	13.18	16.02	19.01	21.42	22.66	22.05	21.78	20.69	17.79	14.62	10.87

Data regarding the average natural daylength and temperature were observed from the detectors at the experimental farm at Yunnan Academy of Agricultural Sciences.

Fig. 2. Effects of different planting seasons on the growth and development of *Gypsophila paniculata*. Statistical significance was analyzed based on a one-way analysis of variance. *P* was calculated with Tukey's honestly significant difference post hoc test ($\alpha = 0.05$).

Stars'; it especially reduced the 40% and 80% rosette rates of 'My Pink' that occurred four applications and eight applications of GA₃ treatment, respectively. This demonstrated that GA₃ treatments during low temperatures could relieve the rosette rate, which is important for the flower production of *Gypsophila paniculata*.

Discussion

In this study, four cultivars of *Gypsophila paniculata* were planted during different months (July, September, and November) of the natural photoperiod. The results showed that the general growth and development were affected by planting seasons, with a trend of extended vegetative growth occur-

ring with delayed planting time. Specifically, some cultivars showed significant sensitivity to planting seasons and generated rosettes when seedlings were planted in autumn and winter, such as 'My Pink' and 'Million Stars'. This was mainly caused by the low temperatures in winter, and GA₃ treatment was able to provide effective improvements. Moreover, low temperatures are not the only limitation to the growth and development of *Gypsophila paniculata*. The photoperiod is an important factor for growth because *Gypsophila paniculata* is a long-day species. It has been reported that a photoperiod of at least 14 h is needed for the flowering of *Gypsophila paniculata* (Nishidate et al., 2012). However, this is not the case for 'Cloudstar 8' and 'Cloudstar

5', which has no sensitivity to the photoperiod. A natural photoperiod of only ≈ 11 h is provided from December to February, but 'Cloudstar 8' and 'Cloudstar 5' are able to flower, with little delay in flowering time. This difference may be caused by the genetic background of different cultivars; 'Cloudstar 8' and 'Cloudstar 5' are specifically bred based on the environmental conditions of Kunming.

Using different quantities of GA₃ applications enhanced the plant height, width, and weight of *Gypsophila paniculata* compared with control, thus indicating that GA₃ treatment could effectively promote plant vegetative growth during winter. More importantly, GA₃ treatment was able to prevent the occurrence of rosettes in 'My Pink' and 'Million

Fig. 3. Effects of different GA₃ spray times on the growth of *Gypsophila paniculata*. Statistical significance was analyzed based on a one-way analysis of variance. *P* was calculated with Tukey's honestly significant difference post hoc test ($\alpha = 0.05$).

Stars'. This demonstrated that GA₃ treatment could stimulate and promote plant growth and development. Gibberellins are a type of high-performance and broad-spectrum plant growth regulator and critical plant hormone (Hedden and Thomas, 2012; Patrick et al., 2009), functioning either from the seedling stage to the maturity stage (Sun, 2010) or from nutritional development to reproductive development (Eriksson et al., 2006). According to the current study of the model plant *Arabidopsis*, flowering was controlled by the interaction between three different pathways (long-day pathway, autonomous pathway, and gibberellin pathway), but the gibberellin pathway was the main regulator of floral induction (Boss

et al., 2004; Putterill and Laurie, 2004; Simpson, 2004). The molecular mechanisms of gibberellins regulation could be partially explained by the regulation of the flower meristem identity gene *LEAFY* and flowering time gene *SUPPRESSOR OF CONSTANS 1* (Blázquez et al., 1998; Blázquez et al., 1997; Mouhu et al., 2013; Tyagi et al., 2018). However, the molecular mechanisms controlling the regulation of flowering in *Gypsophila paniculata* are not well-known and require further study. Our research provides a simple and efficient way to prevent rosettes when planting *Gypsophila paniculata* during winter. This method could be applied during the flower production of *Gypsophila paniculata*.

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