

The effects of gibberellic acid and short-day treatments on growth and flowering of *Chrysanthemum morifolium* cv. Breeze cassus

Pesanai Zanamwe^{1*} and Rene Tong²

¹Department of Crop Science, Gwanda State University, P.O. Box 30, Filabusi, Zimbabwe,

²Department of Botany and Plant Biotechnology, University of Johannesburg, Johannesburg, 2006, South Africa

*Corresponding author: zanamwep@gmail.com

Abstract

Chrysanthemum morifolium cv. Breeze cassus that is widely grown as a commercial pot plant in South Africa grow short and branch poorly under the usual photoperiodic conditions. It was hypothesised that understanding the effects of gibberellic acid (GA₃), a plant hormone that promotes flowering and stem elongation in plants, may aid in improving its quality. To study this, the vegetative growth and flowering characteristics of *Chrysanthemum morifolium* cv. Breeze cassus plants treated with GA₃ once, i.e., three weeks after transplanting, GA₃ twice, i.e., three and five weeks after transplanting, gibberellin biosynthesis inhibitor, daminozide, and short-day photoperiod were evaluated. Data was collected on plant height, number of branches per plant, time to flowering, flower buds per plant and flower size and subjected to analysis of variance. Plants treated with a short day photoperiod flowered early compared to the control. However, the plant height and flower buds per plant were significantly ($p < 0.05$) reduced. The final plant height of short-day photoperiod and daminozide treated plants were not statistically ($p > 0.05$) different. Both GA₃ treatments significantly ($p < 0.05$) increased plant height compared to the control, daminozide and short-day photoperiod treated plants. Plants treated with GA₃ twice were significantly ($p < 0.05$) taller compared to plants treated with GA₃ once. Treatment with GA₃, either once or twice, had no effect on time to flowering, however, treating plants with GA₃ once, significantly increased flower buds and treating plants with GA₃ twice significantly increased flower diameter compared to the control. It was noted that treatment of *Chrysanthemum morifolium* cv. Breeze cassus with short-day photoperiod and GA₃ have different effects on plant growth and flowering. It was, therefore, concluded that, although GA₃ increases plant height of *Chrysanthemum morifolium* cv. Breeze cassus, it may not replace short day photoperiod treatment where early flowering is desired.

Keywords: *Gibberellic acid; Chrysanthemum; Short-day photoperiod; Flowering time; Plant size; Daminozide*

Introduction

Chrysanthemum morifolium cv. Breeze cassus is widely grown as a commercial pot plant in South Africa. However, the plant grows short and branch poorly under the usual photoperiodic conditions (promoting flowering by following long days with short days). Photoperiodic conditions are used in forced (off-season) production of flowering plants whose flowering is regulated by genes that are photoperiod responsive (Malleshaiah *et al.*, 2021). *Chrysanthemum* is one of the plants which are photoperiod sensitive where

short days induce flower bud initiation. In this plant, manipulation of the photoperiod to maintain plants in vegetative phase or to induce flowering was noted for revolutionising its cultivation (Palai *et al.*, 2018). However, in *Chrysanthemum morifolium* cv. Breeze cassus, forced flowering produces undesirably poor branched short plants (less than 20 cm tall). *Chrysanthemums* grown as pot plants should exhibit well-shaped growth form, branch readily, have relatively short stems (approximately 20 cm) and their flowers

should have desirable shape, size, and colour (Sharma *et al.*, 2016; Qureshi *et al.*, 2018).

Gibberellic acid (GA₃) and its synthesis inhibitors such as daminozide are generally known to regulate plant size and flowering in many plant species (Qureshi *et al.*, 2018). GA₃ promote stem elongation and reduce the number of days to flowering in some chrysanthemum cultivars (Padmapriya and Chezhiyan, 2002; Mohariya *et al.*, 2003; Muhammad Sajid *et al.*, 2016). Gibberellic acid stimulates shoot elongation by stimulating cell division and cell expansion (Bhattacharya *et al.*, 2010; Taiz *et al.*, 2015). In some chrysanthemum cultivars, GA₃ causes flower initiation and early flowering by decreasing the concentration of abscisic acid (ABA) in plant shoots as well as by increasing photosynthates (Lawrance and Copeland, 2000; Phengphachanh *et al.*, 2012). Gibberellic acid is also known to be a component of florigen, which is required for the formation of flowers in the plant system and for rapid mobilization and accumulation of metabolites which influence floral morphogenesis resulting in bigger flower sizes (Phengphachanh *et al.*, 2012). Gibberellins biosynthesis inhibitors are used where short and sturdy stems are desirable (Taiz and Zeiger, 2010). Bergstrand (2017) noted that relatively short (20 cm) plants that branch profusely are generally preferred by retailers and consumers. Chemical growth retardants with gibberellic acid inhibiting mode of action such as daminozide, chlormequat chloride (CCC) and paclobutrazol were developed and used to reduce plant height in floriculture and other branches of horticulture (Rademacher, 2016). Therefore, it was hypothesised that understanding the effects of gibberellins (GA) on growth and flowering may aid in improving the quality of *Chrysanthemum morifolium* cv. Breeze cassus. Moreover, studying the effect of short-day treatment on plant size and flowering may also help in understanding the problem of reduced growth and branching in *Chrysanthemum*

morifolium cv. Breeze cassus produced under the usual photoperiod.

Chrysanthemum is generally a short day plant whose flowering can be induced by natural shortening of day length and or could be induced by artificial shortening of the day length (Nxumalo and Wahome, 2010; Palai *et al.*, 2018). Nxumalo and Wahome (2010) noted that the critical photoperiod (light period) is 12 hours or less for reproductive growth and 14 hours or more for vegetative growth. The usual photoperiodic sequence for producing chrysanthemums is to provide long days in order to promote vegetative growth, followed by short-days which promote flowering. However, in *Chrysanthemum morifolium* cv. Breeze cassus, this photoperiodic sequence results in undesirably short and poor branching plants. This study was, therefore, carried out to understand the effect of increasing endogenous gibberellins with GA₃ treatment, inhibiting endogenous GA synthesis by daminozide and exposing plants to short day photoperiod on growth and flowering of *Chrysanthemum morifolium* cv. Breeze cassus.

Materials and Methods

Plant material

Forty six days old uniformly rooted cuttings of *Chrysanthemum morifolium* cv. Breeze cassus, procured from a commercial propagator (Bergbron Nursery; Johannesburg, South Africa) were transplanted to 1.5 litre plastic pots (each pot containing one plant) on the 13th of August in 2017 and 2018. The pots were filled with potting compost, also supplied by Bergbron Nursery (Johannesburg, South Africa). The plants were pinched after 14 days from transplanting and exposed to different treatments seven day (21 days from transplanting) after pinching.

Experimental design and treatments

The experiment was laid out in a complete randomized design (CRD) with five treatments including the control. Treatments

were replicated three times. The treatments comprised of spraying plants with 250 ppm GA₃ (Moond and Rakesh, 2006) once, i.e., three weeks (21 days) after transplanting, spraying plants with 250 ppm GA₃ twice, i.e., three and five weeks (21 and 35 days) after transplanting, spraying plants with 1000 ppm daminozide (Gautam *et al.*, 2006), three weeks (21 days) after transplanting, exposing plants to 8 hours light and 16 hours darkness short-day photoperiod from three weeks (21 days) after transplanting to flowering and the control (not treated). Gibberellic acid and daminozide were applied as foliar sprays using a hand sprayer of a volume of 30 ml.

The plants were raised in a wide span greenhouse set at $23 \pm 2^\circ\text{C}$ at the University of Johannesburg, South Africa (26.1832°S , 27.9990°E). The pots in which the plants were grown were spaced 10 cm apart on a 50 cm high greenhouse bench. Seven days after pinching, the short-day photoperiod treatment plants were transferred to a growth chamber set at $23 \pm 2^\circ\text{C}$ temperature and 8 hours light and 16 hours darkness photoperiod where they grew up to full bloom (flowering) stage. The control, GA₃ and daminozide treatment plants continued to grow in the greenhouse. All treatments plants were fertilized with 3 g compound fertilizer (15% N, 15% P₂O₅ and 15% K₂O) per pot at transplanting and 5 weeks from transplanting, according to Palai *et al.* (2018). The plants were irrigated once a week until water started dripping from the bottom holes of the pots.

Data collection

Plant height and number of branches developed after pinching, and reproductive characteristics; reaction time (time to flowering), number of flower buds per plant and flower size were measured. Plant height was determined by measuring the height of the plant from the rim of the pot to the top of the plant at 14 days interval from 45 days after transplanting to 77 days after transplanting using a ruler. The number of branches that developed after pinching

(branches per plant) were determined by counting the branches that developed during the first 21 days of growing after pinching. The reaction time was determined as the number of days from transplanting to first visual flower bud (Sajid *et al.* (2016). The number of flower buds were determined by counting all the flower buds after 63 and 77 days from transplanting (Sajid *et al.*, 2016). Flower size was determined by measuring flower diameter using Vernier calliper at full bloom growth stage (Delvadia *et al.*, 2009).

Data analysis

Analysis of variance (ANOVA) was performed to determine the effect of treatments on plant growth and development. The significant means were separated using Fisher method at 5 % level of significance using the General Linear Models (GLM) program in the Minitab statistical software. Graphs were drawn using Microsoft Office Excel, version 2007 (Microsoft Corporation, Redmond, WA 98052-7329, USA).

Results

Number of branches formed after pinching

There was no significant effect ($p = 0.596$) of treating *Chrysanthemum morifolium* cv. Breeze cassus plants with GA₃ once, GA₃ twice, daminozide, 8 hours light and 16 hours short-day photoperiod from three weeks after transplanting to flowering and the control on the number of branches formed after pinching (Table 1). In all treatments, *Chrysanthemum morifolium* cv. Breeze cassus formed an average of three branches after pinching (Table 1).

Table 1: The effects of various treatments on the number of branches formed by *Chrysanthemum morifolium* cv. Breeze cassus after pinching.

Treatments	Number of branches
^Control	3
GA ₃ , 3 WT	3
GA ₃ , 3 and 5 WT	3
Daminozide, 3 WT	3
Short day, 3 WT-F	3
p value	0.596

^Control = untreated plants, GA₃, 3WT = plants treated with GA₃ once, i.e., at three weeks after transplanting, GA₃, 3 and 5 WT = plants treated with GA₃ twice, i.e., at 3 and 5 weeks after transplanting, Daminozide, 3 WT = plants treated with daminozide at three weeks after transplanting, Short day, 3 WT-F = plants treated with 8 hours light and 16 hours short day photoperiod from three weeks after transplanting to flowering.

Plant height

There was significant treatments effects ($p < 0.05$) on plant height at the end of 35, 49, 63 and 77 days from transplanting (Fig. 1). The plants that were treated with GA₃ once (three weeks after transplanting) and GA₃ twice (three and five weeks after transplanting) were significantly taller than the plants that were treated with daminozide, plants that were treated with 8 hours light and 16 hours darkness short-day photoperiod and the control after 35, 49, 63 and 77 days after transplanting, respectively. The plants that were treated with GA₃ twice were significantly taller than the plants that were treated with GA₃ once from 49 to 77 days after transplanting. The plants that were

treated with daminozide were significantly shorter than the control plants after 49, 63 and 77 days from transplanting. The plants that were treated with 8 hours light and 16 hours darkness short-day photoperiod were significantly shorter than the control plants after 63 and 77 days from transplanting, however, the heights of these (8 hours light and 16 hours darkness short-day photoperiod treated and the control) treatments plants were not significantly different from transplanting to 49 days after transplanting. The plants that were treated with 8 hours light and 16 hours darkness short-day photoperiod were significantly taller than daminozide treated plants after 49 and 63 days from transplanting, but, not significantly different after 77 days from transplanting.

Reaction time

There was significant treatments effects ($p < 0.001$) on the number of days to flowering (Table 2). *Chrysanthemum morifolium* cv. Breeze cassus started flowering (visible flower bud stage) after 49 days from transplanting (the control plants). The number of days to flowering that were taken by plants that were treated with GA₃ once, GA₃ twice and daminozide were not significantly different from the control plants (49 days from transplanting, respectively). The plants that were treated with 8 hours light and 16 hours darkness short-day photoperiod from three weeks after transplanting to flowering flowered significantly early (35 days from transplanting) compared to plants that were treated with GA₃ once, GA₃ twice, daminozide and the control.

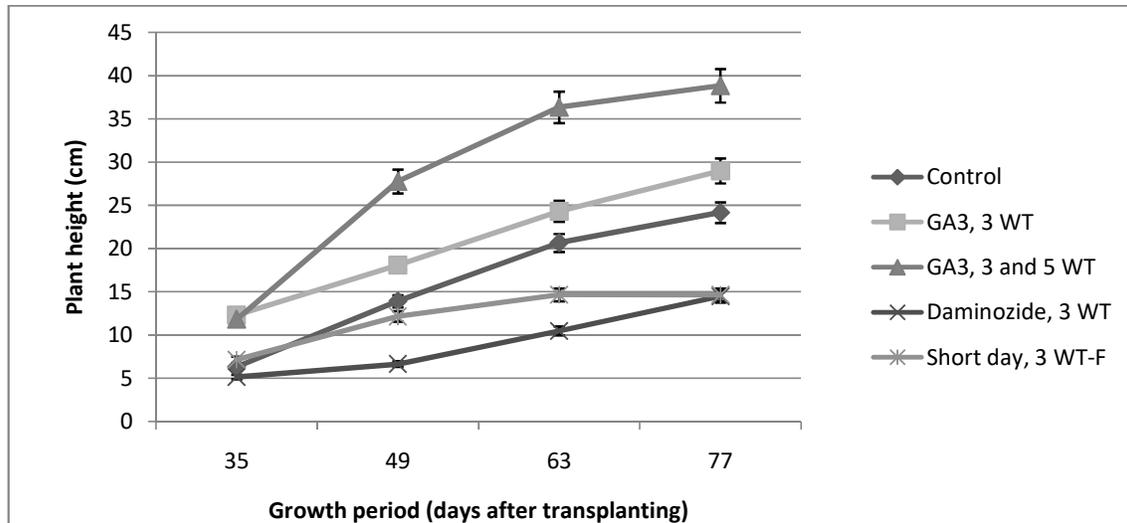


Figure 1: The effects of various treatments on plant height of *Chrysanthemum morifolium* cv. Breeze cassus after 35, 49, 63 and 77 days from transplanting.

The marks on line graphs represents mean height. Error bars = $p < 0.05$, Control = untreated plants, GA₃, 3WT = plants treated with GA₃ once, i.e., at three weeks after transplanting, GA₃, 3 and 5 WT = plants treated with GA₃ twice, i.e., at 3 and 5 weeks after transplanting, Daminozide, 3 WT = plants treated with daminozide at three weeks after transplanting, Short day, 3 WT-F = plants treated with 8 hours light and 16 hours short day photoperiod from three weeks after transplanting to flowering.

Table 2: The effects of various treatments on the number of days to visible flower bud formation on *Chrysanthemum morifolium* cv. Breeze cassus plants.

Treatments	Days to first bud
Control	49 ^a
GA ₃ , 3 WT	49 ^a
GA ₃ , 3 and 5 WT	49 ^a
Daminozide, 3 WT	49 ^a
Short day, 3 WT-F	35 ^b
p value	0.000

^aMeans carrying the same superscripts on the same column are not significantly different at $\alpha = 0.05$.

Control = untreated plants, GA₃, 3WT = plants treated with GA₃ once, i.e., at three weeks after transplanting, GA₃, 3 and 5 WT = plants treated with GA₃ twice, i.e., at 3 and 5 weeks after transplanting, Daminozide, 3 WT = plants treated with daminozide at three weeks after transplanting, Short day, 3 WT-F = plants treated with 8 hours light and 16

hours short day photoperiod from three weeks after transplanting to flowering

Number of flower buds

There was significant treatment effects ($p < 0.05$) on the number of flower buds produced after 63 and 77 days from transplanting (Fig. 2). After 63 and 77 days from transplanting, the plants that were treated with GA₃ once, had a higher number of flower buds compared to the plants that were treated with GA₃ twice, plants treated with daminozide, plants treated with 8 hours light and 16 hours darkness short-day photoperiod from three weeks after transplanting to flowering and the control, respectively. The number of flower buds produced by the plants that were treated with GA₃ twice, daminozide and the control were not significantly different, but, were significantly high compared to plants that were treated with 8 hours light and 16 hours darkness short-day photoperiod from three weeks after transplanting to flowering.

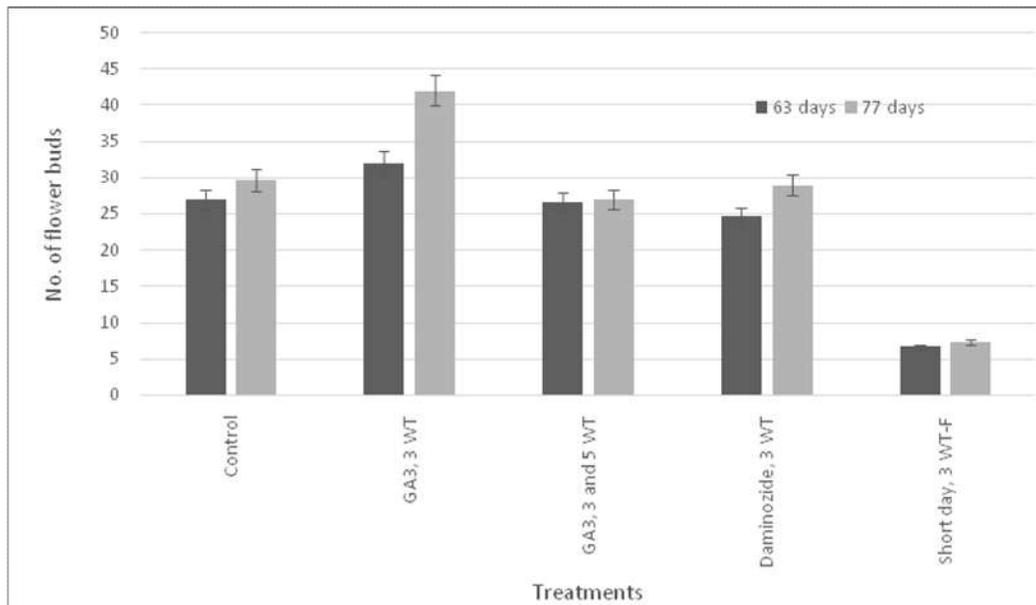


Figure 2: The effects of various treatments on the number of flower buds of *Chrysanthemum morifolium* cv. Breeze cassus after 63 and 77 days from transplanting.

The column bars show the mean flower buds.

Error bars = $p > 0.05$, Control = untreated plants, GA₃, 3WT = plants treated with GA₃ once, i.e., at three weeks after transplanting, GA₃, 3 and 5 WT = plants treated with GA₃ twice, i.e., at 3 and 5 weeks after transplanting, Daminozide, 3 WT = plants treated with daminozide at three weeks after transplanting, Short day, 3 WT-F = plants treated with 8 hours light and 16 hours short day photoperiod from three weeks after transplanting to flowering

Flower diameter

There was significant treatment effects ($p < 0.001$) on diameter of *Chrysanthemum morifolium* cv. Breeze cassus flowers at full bloom (Table 2). The flowers of plants that were treated with GA₃ twice were significantly larger (8.2 cm) compared to plants that were treated with GA₃ once (6.7 cm), daminozide (6.8 cm), 8 hours light and 16 hours darkness short-day photoperiod from three weeks after transplanting to flowering (6.9 cm) and the control (6.9 cm). However, flowers of the plants that were treated with GA₃ once, daminozide, 8 hours light and 16 hours darkness short day photoperiod from three weeks after transplanting to flowering and the control were not significantly different. GA₃ treatment appeared to promote secondary branching, while short-day photoperiod to reduce it (Fig. 3).

Table 3: The effects of various treatments on diameter of *Chrysanthemum morifolium* cv. Breeze cassus flowers at full bloom.

Treatments	Flower diameter (cm)
^Control	6.9 ^b
GA ₃ , 3 WT	6.7 ^b
GA ₃ , 3 and 5 WT	8.2 ^a
Daminozide, 3 WT	6.8 ^b
Short day, 3 WT-F	6.9 ^b
<i>P value</i>	0.000

^aMeans carrying the same superscripts in the same column are not significantly different at $\alpha = 0.05$.

^Control = untreated plants, GA₃, 3WT = plants treated with GA₃ once, i.e., at three weeks after transplanting, GA₃, 3 and 5 WT = plants treated with GA₃ twice, i.e., at 3 and 5 weeks after transplanting, Daminozide, 3 WT = plants treated with daminozide at three weeks after transplanting, Short day, 3 WT-F = plants treated with 8 hours light and 16 hours short day photoperiod from three weeks after transplanting to flowering

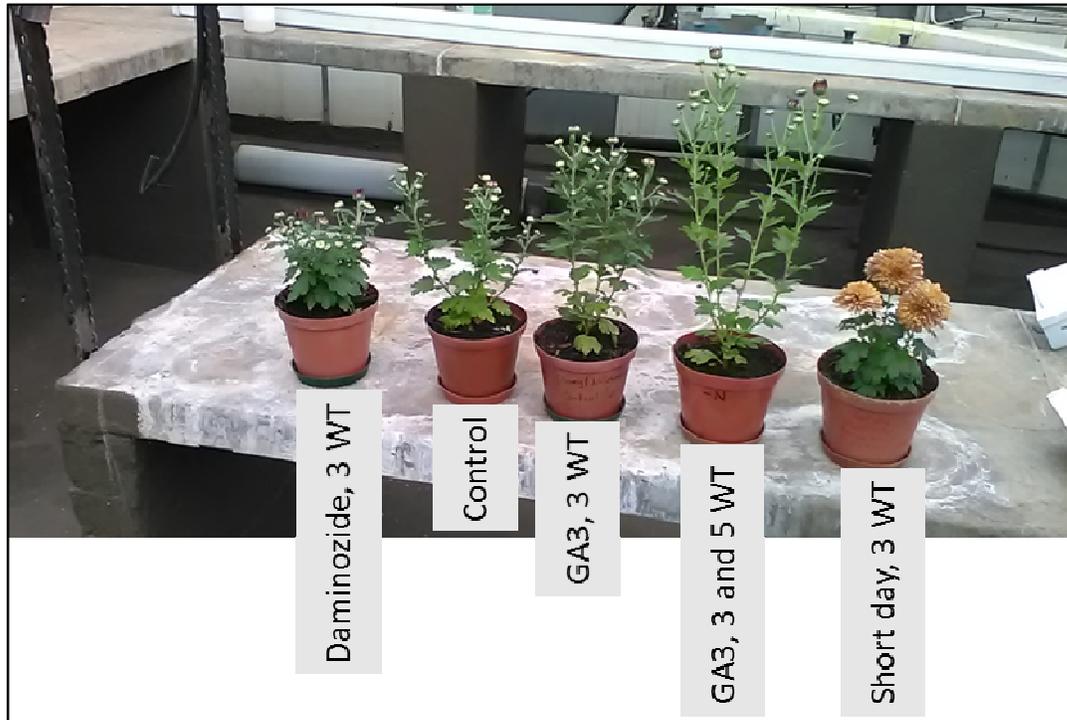


Figure 3: The effects of various treatments on plant height, number of flower buds or flowers and stage of flowering after 63 days from transplanting of *Chrysanthemum morifolium* cv. Breeze cassus. Control = untreated plants, GA₃, 3WT = plants treated with GA₃ once, i.e., at three weeks after transplanting, GA₃, 3 and 5 WT = plants treated with GA₃ twice, i.e., at 3 and 5 weeks after transplanting, Daminozide, 3 WT = plants treated with daminozide at three weeks after transplanting, Short day, 3 WT-F = plants treated with 8 hours light and 16 hours short day photoperiod from three weeks after transplanting to flowering.

Discussion

Treatment of *Chrysanthemum morifolium* cv. Breeze cassus with GA₃ resulted in an increase in plant height. The plants that were sprayed with GA₃ twice (at three weeks and five weeks) were taller than those that were sprayed with GA₃ once (at three weeks after transplanting). Moreover, spraying plants with GA₃ twice, but not once, increased flower size (diameter). On the contrast, spraying plants with GA₃ once, but not twice, increased the number of flower buds per plant. Short-day photoperiod reduced the time to flowering, plant height and the number of flower buds per plant. All treatments did not affect the number of branches produced by the plant after pinching.

Gibberellic acid is thought to promote auxin synthesis which stimulates apical dominance (Patel *et al.*, 2010; Tannirwar *et al.*, 2011, Sajidet *et al.*, 2016; Sharma *et al.*, 2016). Apical dominance is a condition where growth of the terminal bud suppresses the development of lateral buds hence branching. However, the results of this study showed that both GA₃ and short-day photoperiod treatments had no effect on branching of *Chrysanthemum morifolium* cv. Breeze cassus (Table 1). Spraying *Chrysanthemum morifolium* cv. Breeze cassus plants with GA₃ once i.e., three weeks after transplanting or twice (three and five weeks after transplanting) had no effect on the number of branches formed after pinching. Sharma *et al.* (2016) also reported no significant effect of GA₃ on both primary (branches formed after pinching) and

secondary branches (branches formed towards and during the flowering stage) in Garland chrysanthemum (*Dendranthema grandiflora* Tzvelev). However, the present study did not determine the number of secondary branches. Besides, GA₃ treatment seems to promote secondary branching and short-day photoperiod to reduce the secondary branching (Fig. 3). The results also showed that inhibiting gibberellins synthesis by spraying plants with daminozide three weeks after transplanting had no effect on branching (primary). Qureshi *et al.* (2018) also found no significant daminozide effect on the number of branches when daminozide was sprayed 2 and 6 weeks after transplanting. Notwithstanding the results of afore said studies, GA₃ was reported to increase branching in other plant species as well as other chrysanthemum cultivars. In Gaillardia (*Gaillardia pulchella* Foug cv. Lorenziana), GA₃ increased branching and this was attributed to increase in nodal count per main axis which increased the number of dormant buds from which branches were formed (Delvadia *et al.*, 2009). Although the number of nodes per stem were not counted in this study, the number of primary branches were not affected by both GA₃ treatment and gibberellins synthesis inhibitor, daminozide. These results suggest that gibberellins do not affect primary branching in *Chrysanthemum morifolium* cv. Breeze cassus. In *Chrysanthemum* cv. Paintball, treatment with 300 ppm GA₃ increased the number of branches per plant (Alhajhoj *et al.*, 2017), however, the study did not indicate the type of branches increased, i.e., primary or secondary. It is important to report branching as primary, secondary or a combination of primary and secondary branches in chrysanthemum plants. It is, therefore, not clear if the effect of GA₃ on branching of *Chrysanthemum* plants varies with cultivars. Besides, the results of the present study showed that GA₃ has no effect on primary branching. However, Figure 3 suggest that the secondary branches formed by plants treated with GA₃ once, GA₃ twice, daminozide and

the control were higher compared to those of the plants that were exposed to 8 hours light and 16 hours darkness short-day photoperiod. However, the number of branches formed after pinching (primary branches) were also not affected by treating plants with a short-day photoperiod of 8 hours light and 16 hours darkness. Light was reported to be indispensable for bud burst through its effect in mobilisation of carbohydrates (Girault *et al.*, 2010). It is, therefore, noted that both GA₃ and short-day photoperiod treatments did not affect primary branching of *Chrysanthemum morifolium* cv. Breeze cassus plants.

Treating *Chrysanthemum morifolium* cv. Breeze cassus plants with GA₃ increased plant height (Fig. 1). Plants that were treated with GA₃ (both once and twice) were taller than the control plants at all observed time points (i.e., at 35, 49, 63 and 77 days after transplanting, respectively). Moreover, treating *Chrysanthemum morifolium* cv. Breeze cassus with GA₃ twice (3 and 5 weeks after transplanting) increased the height of the plants more than that of plants that were treated once (three weeks after transplanting) after 35, 49 and 77 days from transplanting, respectively. Again, plants that were treated with a gibberellins synthesis inhibitor, daminozide, were shorter than the control plants after 63 and 77 days from transplanting, respectively. Earlier studies by Gupta and Datta, (2001); Gautam *et al.* (2006); Delvadia *et al.* (2008); Pahade (2015); and Sajid *et al.* (2016) also reported that active forms of gibberellins (e.g., GA₃) increased plant height and GA synthesis inhibitors such as daminozide (Bergstrand, 2017) and chlormequat chloride (Alhajhoj *et al.*, 2017) reduced plant height in other chrysanthemum cultivars. Generally, gibberellins regulates the size of plants by increasing internodal length due to increased cell division and elongation (Delvadia *et al.*, 2008; Sajid *et al.*, 2016). However, it was reported that GA₃ failed to increase stem height in *Chrysanthemum* cv. Faroe and Yoko ono (Vieira *et al.*, 2011). This suggests

that GA₃ treatment have different effects on the height of different chrysanthemum cultivars. It should, therefore, be noted that, the results of this study suggests that GA₃ increases and daminozide decreases plant height in *Chrysanthemum morifolium* cv. Breeze cassus. The plant height increasing effect of GA₃ treatment was enhanced by treating plants twice (i.e., three and five weeks after transplanting). Short-day photoperiod also affected plant height, however, the effect of short-day photoperiod was opposite that of GA₃ treatment.

Treating *Chrysanthemum morifolium* cv. Breeze cassus plants with a short-day photoperiod of 8 hours light and 16 hours darkness from three weeks after transplanting to flowering reduced plant height (Fig. 1). The height of plants that were treated with 8 hours light and 16 hours darkness photoperiod were shorter than the control plant after 49, 63 and 77 days from transplanting, respectively. This could be probably due to reduced photosynthesis. Light is required for photosynthesis, a process through which carbohydrates that are important for plant growth are synthesised (Taiz *et al.*, 2015). Warrington and Norton (1991) also reported reduced plant height in chrysanthemum plants that were treated with a short-day photoperiod and they attributed this to reduced levels of assimilates. Nxumalo and Wahome (2010) noted that big chrysanthemum plants can be obtained if the time of long days provided to promote vegetative growth before floral inductive short-day photoperiod, is increased. In the present study, the plants were exposed to short-day photoperiod three weeks after transplanting. The potential of increasing (more than three weeks from transplanting) the time of exposure to long day photoperiod before treating plants with a short-day photoperiod should be examined. Short-day photoperiod and GA₃ treatments also showed significant different effects on time of flowering (reaction time).

Chrysanthemum morifolium cv. Breeze cassus started flowering 49 days after transplanting (Table 2; the control plants). Gibberellic acid treatment had no effect on time of flowering. The number of days to flowering for plants that were treated with GA₃ (either once or twice) were not significantly different from the control. Moreover, inhibiting gibberellins synthesis by treating plants with daminozide had no effect on the number of days to flowering. Gilbertz (1992) also found no differences on time to flowering between daminozide treated *Dendranthema × grandiflorum* (Ramat.) plants and the control. However, GA₃ was reported to reduce the number of days to flowering in *Chrysanthemum morifolium* Ramat cv. Jayanti (Gupta and Datta, 2001) and *Chrysanthemum* cv. ZIPRI (50 ppm but not 200 ppm; Tannirwar *et al.*, 2018). Sharma *et al.* (2016) reported a reduced time to flowering due to GA₃ treatment, but, no effect regardless of GA₃ treatment where pinching was done. In this study, the plants were pinched 14 days after transplanting and this could be the reason why GA₃ had no effect on flowering time. Future studies should, therefore, seek to understand the effect of pinching on time of flowering of GA₃ treated chrysanthemum plants. However, gibberellins were reported to play a subsidiary role in flowering of chrysanthemums (Dong *et al.*, 2017). Treatment with short-day photoperiod of 8 hours light and 16 hours darkness three weeks after transplanting to flowering, reduced the number of days to flowering by 14 days (Table 2). Generally, chrysanthemum is a short-day plant; hence, long nights (11 to 12 hours of darkness or longer, depending on cultivar) are required for rapid flower initiation and development (Runkle and Vaid, 2013). It can, therefore, be noted that, early flowering can be induced by a short-day photoperiod of 8 hours light and 16 hours darkness in *Chrysanthemum morifolium* cv. Breeze cassus. However, the number of flowers per plant is reduced by this short day photoperiod.

Notwithstanding the reduced reaction time effect of short-day photoperiod treatment, a short-day photoperiod of 8 hours light and 16 hours darkness reduced the number of flower buds per plant compared to the control, GA₃ (both once and twice) and daminozide treatments, respectively (Figures 2 and 3). This could be attributed to few secondary branches. Although, the number of secondary branches were not counted in this study, they were observed to be fewer compared to the control and all other treatments (Fig. 3). It can be noted that a short-day photoperiod of 8 hours light and 16 hours darkness reduce flower production in *Chrysanthemum morifolium* cv. Breeze cassus by reducing secondary branching. Treating *Chrysanthemum morifolium* cv. Breeze cassus plants with GA₃ once (three weeks after transplanting) increased the number of flower buds, however, spraying the plants with GA₃ twice had no effect on the number of flower buds per plant. This could be attributed to promotion of apical dominance. Gibberellic acid was reported to promote apical dominance by stimulating auxin synthesis (Patel *et al.*, 2010; Tannirwar *et al.*, 2011, Sajidet *et al.*, 2016; Sharma *et al.*, 2016). Auxin is known to promote apical dominance and inhibit branching by keeping lateral buds dormant (Taizet *et al.*, 2015). The effect of early treatment (three weeks after transplanting) on apical dominance appears to terminate earlier allowing increased secondary branching (Delvadiaet *et al.*, 2009). Although the number of flower buds were not reduced below the control, apical dominance appeared to allow branching to occur to the same level as the control where plants were treated with GA₃ two times (three and five weeks after transplanting). Daminozide had no effect on the number of flower buds per plant. It may be noted that treatment of *Chrysanthemum morifolium* cv. Breeze cassus with GA₃ three weeks after transplanting improves flower production in addition to increasing plant height. However, treatment with GA₃ twice (three and five weeks after transplanting), although it increased plant height more than treatment

with the same hormone only once, do not improve flower production.

In addition to the opposite effect of GA₃ and short-day photoperiod on the time of flowering and number of flower buds per plant, GA₃ and short-day photoperiod had opposite effects on flower size. Short-day photoperiod had no effect on flower size and GA₃ sprayed two times (three and five weeks after transplanting) increased flower size (Table 3). Gupta and Datta (2001) reported increased flower size when GA₃ was sprayed 4 times on *Chrysanthemum morifolium*, Ramat cv. Jayanti. In China aster (*Callistephus chinensis* cv. Nees), spraying plants with 50 ppm GA₃, three times increased flower size (Mishra *et al.*, 2018). In the present study, GA₃ did not affect flower size where it was applied once (three weeks after transplanting), but increased flower size significantly when applied twice. Again treating plants with daminozide had no effect on flower diameter. Qureshi *et al.* (2018) also reported that daminozide did not affect flower diameter of *Chrysanthemum morifolium* cv. Flirt. These findings may suggest that, GA₃ increases the size of flowers when applied more than once. However, Alhajhoj *et al.* (2017) reported significant increase in flower size when *Chrysanthemum* cv. Paintball was treated once with 300 ppm GA₃. Further studies including many cultivars of *Chrysanthemum* is, therefore, required to understand the effect of treating chrysanthemum with GA₃ on flower size.

Conclusion

Treatment of *Chrysanthemum morifolium* cv. Breeze cassus with GA₃ and short-day photoperiod has opposite effects on growth and flowering. Eight hours light and 16 hours darkness short-day photoperiod treatment caused early flowering, fewer flower buds and reduced plant height. Gibberellic acid did not affect the time of flowering, however, it increased plant height, number of flower buds when applied once and flower size when applied twice. It could

be concluded that short-day photoperiod and GA₃ treatments may be used in achieving different objectives in *Chrysanthemum morifolium* cv. Breeze cassus production. An 8 hours light and 16 hours darkness short-day photoperiod treatment can be used where the objective is to speed up flowering without paying much attention to plant height and flower yield and gibberellic acid and its synthesis inhibitors could be used where the objective is to manipulate plant height but not time of flowering.

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References

- Alhajhoj MR. (2017). Effects of Foliar Application of Plant Growth Regulators on Growth and Flowering Characteristics of *Chrysanthemum* CV. Paintball. *Pakistan Journal of Life & Social Sciences*, **15**(2), 114-119
- Bergstrand KI. (2017). Methods for growth regulation of greenhouse produced ornamental pot- and bedding plants – a current review. *Folia Horticulture*, **29**(1), 63-74.
- Bhattacharya A, Kourmpetli S, Davey MR. (2010). Practical applications of manipulating plant architecture by regulating gibberellin metabolism. *Journal of plant growth regulation*, **29**(2), 249-256. <https://doi.org/10.1007/s00344-009-9126-3>
- Delvadia DV, Ahlawat TR, Meena BJ. (2016). Effect of Different GA₃ Concentration and Frequency on Growth, Flowering and Yield in *Gaillardia* (*Gaillardia pulchella* Foug.) Cv. Lorenziana. *Journal of Horticultural Science*, **4**(1), 81-84.
- Dong B, Deng Y, Wang H, Gao R, Stephen GUK, Chen S, Jiang J, Chen F. (2017). Gibberellic acid signaling is required to induce flowering of chrysanthemums grown under both short and long days. *International Journal of Molecular Sciences*, **18**(6), 1259. doi.org/10.3390/ijms18061259
- Gilbertz DA. (1992). Chrysanthemum response to timing of paclobutrazol and uniconazole sprays. *HortScience*, **27**(4), 322-323. <https://doi.org/10.21273/HORTSCI.27.4.322>
- Girault T, Abidi F, Sigogne M, Pelleschi-Travier S, Boumaza R, Sakr S, Leduc N. (2010). Sugars are under light control during bud burst in *Rosa* sp. *Plant, Cell & Environment*, **33**(8), 1339-1350. doi.org/10.1111/j.1365-3040.2010.02152.x
- Gupta VN, Datta SK, Banerji BK. (2001). Influence of Gibberellic Acid (GA₃) on Growth and Flowering in *Chrysanthemum* (*Chrysanthemum morifolium*, Ramat) cv. Jayanti. *Indian Journal of Plant Physiology*, **6**(4), 420-422
- Mishra PP, Pandey G, Moharana, DP. (2018). Influence of various concentrations of gibberellic acid (GA₃) and spraying frequencies on growth, yield and post-harvest parameters of China aster [*Callistephus chinensis* (L.) Nees.]. *International Journal of Chemical Studies*, **6**(3), 89-92.
- Moond SK, Rakesh RS. (2006). Effect of GA₃, CCC and MH on vegetative growth and yield of chrysanthemum. *Horticultural Science*, **35**, 258-59.
- Mohariya AD, Patil BN, Wankheda SG, Band PE, Kartikeyan R. (2003). Effect of GA₃ and Tiba on Growth, Flowering and yield of Different varieties of *Chrysanthemum*. *Advances in Plant Sciences*, **16**(1), 143-146.
- Nxumalo SS, Wahome PK. (2010). Effects of application of short-days at different periods of the day on growth and

- flowering in chrysanthemum (*Dendranthema grandiflorum*). *Journal of Agriculture and Social Sciences*, **6**(2), 39-42.
- Padmapriya S, Chezhiyan N. (2002). Influence of gibberellic acid and certain other chemicals on flowering characters of chrysanthemum (*Dendranthema grandiflora* Tzeled) cultivars-I. *South Indian Horticulture*, **50**(4/6), 437-443.
- Pahade V. (2015). Effect of Plant Growth Regulators on Growth and Flowering of Chrysanthemum (*Dendranthema grandiflora* Tzvelev). M.Sc, Thesis. RajmataVijayarajeScindiaKrishiVishwa Vidyalaya, Gwalior, MP.
- Palai SK, Madhuri G, Nath MR, Bhuyan S. (2018). Effect of planting dates and photoperiod on growth and flowering of chrysanthemum (*Chrysanthemum morifolium* Ramat) cv. yellow Reagan. *The Pharma Innovation*, **7**(5), 106-108.
- Patel SR, Parekh NS, Parmar AB, Patel HC. (2010). Effect of growth regulators on growth, flowering and yield of chrysanthemum (*Chrysanthemum morifolium* Ramat.) cv." IIHR-6" under middle Gujarat conditions. *International Journal of Agricultural Sciences*, **6**(1), 243-245.
- Phengphachanh B, Naphrom D, Bundithya W, Potapohn N. (2012). Effects of day-length and gibberellic acid (GA3) on flowering and endogenous hormone levels in *Rhynchostylis gigantea* (Lindl.) Ridl. *Journal of Agricultural Science*, **4**(4), 217-222. doi.org/10.5539/jas.v4n4p217
- Qureshi IA, Gulzar S, Dar AR, Rehman RU, Tahir I. (2018). Effect of growth retardants on the growth and flowering of *Chrysanthemum morifolium* cv. Flirt. *Indian Journal of Agricultural Research*, **52**(3), 319-322.
- Rademacher W. (2016). Chemical regulators of gibberellin status and their application in plant production. *Annual Plant Reviews*, **49**, 359-403. doi.org/10.1002/9781119210436.ch12
- Reddy YTN, Sulladmath UV. (1983). Effect of growth regulators on growth and flowering of China aster (*Callistephus chinensis* Nees). *South Indian Horticulture*, **31**, 95-98
- Runkle E, Vaid T. (2013). Temperature on Chrysanthemum. Technically speaking, 62.
- Sajid M, Amin N, Ahmad H, Khan K. (2016). Effect of gibberellic acid on enhancing flowering time in *Chrysanthemum morifolium*. *Pakistan Journal of Botany*, **48**(2), 477-483.
- Scott TK, Case DB, Jacobs WP. (1967). Auxin gibberellin interaction in apical dominance. *Plant Physiology*, **42**, 1329-1333
- SharathKumar M, Heuvelink E, Marcelis LF, Van Ieperen W. (2021). Floral induction in the short-day plant chrysanthemum under blue and red extended long-days. *Frontiers in Plant Science*, **11**: 1-13. doi: 10.3389/fpls.2020.610041.
- Sharma G, Patanwar M, Mishra P, Shukla N. (2016). Effect of plant growth regulators and pinching on garland chrysanthemum (*Dendranthema grandiflora* Tzvelev). *International Journal of Bio-resource and Stress Management*, **7**(4), 766-769.
- Taiz L, Zeiger E. (2010). *Plant Physiology*. 5th Edition. Sinauer Associates Inc., Sunderland, Massachusetts, U.S.A.
- Taiz L, Zeiger E, Møller IM, Murphy A. (2015). *Plant physiology and development*. 6th Edition. Sinauer Associates Inc., Sunderland, Massachusetts, U.S.A.
- Tannirwar AV, Dange NR, Brahmankar SB. (2011). Effect of growth regulators and nutrients on growth and flowering of chrysanthemum cv. ZIPRI. *Asian Journal of Horticulture*, **6**(1), 269-270.
- Vieira M, de Souza AV, Santos, CMG, Alves L, Cerqueira RC, Paes R, de Souza AD, Fernandes L. (2011). Stem diameter and height of chrysanthemum cv. Yoko ono as affected by gibberellic acid.

African Journal of Biotechnology,
10(56), 11943-11947.

Warrington I.J, Norton RA. (1991). An evaluation of plant growth and development under various daily quantum integrals. *Journal of the American Society for Horticultural Science*, **116**(3), 544-551. doi.org/10.21273/JASHS.116.3.544

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