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EFFECT OF PACLOBUTRAZOL AND ABSCISIC ACID APPLICATION ON GROWTH AND PRODUCTIVITY OF CANOLA (*Brassica napus* L.)

Raja Muhammad NAQQASH[1†], Ilkay YAVAS[2†], Kaleem UL DIN[3], Ghadeer M. ALBADRANI[4], Asif IQBAL[1], Saddam HUSSAIN[1,4]*

[1]Department of Agronomy, University of Agriculture, Faisalabad-PAKISTAN
[2] Department of Plant and Animal Production, Kocarli Vocational School, Aydin Adnan Menderes University, Aydın, TURKEY
[3]Department of Botany, University of Agriculture, Faisalabad-PAKISTAN
[4] Department of Biology, College of Science, Princess Nourah bint Abdulrahman University, Riyadh, SAUDI ARABIA
[†] These are the equal contributing first author
**Corresponding author: iyavas@adu.edu.tr*

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ABSTRACT

Canola (*Brassica napus* L.) is an important non-conventional oilseed crop that can be grown in diverse soil and climatic conditions. However, seed shattering after maturity is a major problem in canola production as it can cause up to 50% yield loss if harvesting is delayed due to adverse conditions. Different agronomic and physiological interventions can be opted to enhance yield stability and shattering resistance in canola. The present field study aimed at exploring the potential effects of paclobutrazol and abscisic acid on the growth and yield contributing attributes of canola. The treatments of the experiment included a control (no-spray), water spray, paclobutrazol (50 mg L^{-1}), abscisic acid (0.5 mM), and a combination of paclobutrazol+ abscisic acid (50 mg L^{-1}+0.5 mM). The current experiment examined two canola cultivars, Hybrid (45S42) and Inbred Sandal Canola. The experiment was laid out under Randomized Complete Block Design (RCBD) with a factorial arrangement having three replications. The results showed that exogenous application of paclobutrazol and abscisic acid alone or in combination significantly affected both canola cultivars. The plant height decreased significantly however, leaf area index, grain weight, number of seeds per silique, and number of branches per plant were increased compared with the control. Moreover, the combination of paclobutrazol and abscisic acid recorded a higher increase in leaf area index, dry matter accumulation, plant yield, and number of seeds per pod than their individual application. Between cultivars, the Hybrid cultivar (45S42) recorded better yield as compared to the Inbred Sandal Canola. These results thereby suggest that Hybrid (45S42) cultivar with combined application of paclobutrazol and abscisic acid can be a suitable option for gaining high yield and productivity.

Keywords: Growth retardants, Canola, Plant height, Seed Shattering, Production

INTRODUCTION

The increase in human population has changed the dietary habits of mankind along with increased demands for food and fuel. In the future, providing a balanced diet to the rising population will be a major challenge for global agriculturists. Fats and oils are an integral part of each daily diet (Karp and Richter, 2011). Pakistan has become the world's third-largest edible oil importer. Pakistan has been lacking in producing edible oil and spends a giant amount of budget on the import of edible oil and oilseeds to fulfil our necessities (Shahzad*,* 2003).

Rapeseed (*Brassica napus* L.) is an annual crop that ranks among the major five oil-seed crops cultivated worldwide (Khattab and Laila, 2002 and Liersch et al., 2013). The production potential of many crops in developing countries is underexplored mainly because of inadequate management and nutrition-related constraints (Gosh et al., 2006; Safdarı-monfared et al., 2020 and Sher et al., 2023). Among oilseed crops, rapeseed and mustard is the second main source in the globe which provides up to 13% of oil in the world. In Pakistan, canola was introduced in 1995 to replace low-quality conventional rapeseed and mustard crops. Seed of canola contains 40% oil and 30-35% protein. However, the average yield is very low compared to other developed countries of the world (Shahzad, 2003 and Saleem, 2018). Several factors such as, storage of seeds, agronomic practices, weed population, pod shattering, and lower harvest index contribute to a decrease in the productivity of canola

(Madani, 2016). Plant height has a great effect on the production of canola because greater height boosts the chances of lodging (Gan et al., 2008). In canola production, lodging is a major contributor of decreased yield under favorable climatic conditions (Kumar et al., 2012 and Wu and Ma, 2018).

Different chemical substances can be applied to control various aspects of development and growth in plants. Paclobutrazol [(2RS, 3RS)-1-(4-chlorophenyl)-4,4-dimethyl-2-(1H-1,2,4-triazol-1-yl)-pentan-3-ol] is a member of the triazole family, and can alter the levels of key plant hormones, including gibberellins, abscisic acid, and cytokinins (Kum and Gebeloglu, 2024). Paclobutrazol impacts the isoprenoid pathway and modifies plant hormone levels by inhibiting gibberellin synthesis, increasing cytokinin levels, and consequently reducing stem elongation. Application of paclobutrazol also reduced the stem height in *Solanum tuberosum* L., (Mabvongwe et al., 2016), Mango (*Mangifera indica* L.), (Kumar et al., 2020), *Cymbidium hybridum* L. (Li et al., 2020), and ornamental tomatoes (de Moraes et al., 2005). Paclobutrazol application has also been reported to improve canola yield (Zhou and Xi, 1993).

However, several studies have reported the role of paclobutrazol and abscisic acid in reducing shattering losses and crop lodging (Kumar et al., 2012). However, the combined exogenous application of paclobutrazol and abscisic acid has not been studied and needs to be further explored. Furthermore, the application of these growth retardants has not been studied yet in canola cultivars. The objective of the current study was to evaluate the vital role of the above-mentioned growth retardants on the morphological and yield attributes of canola cultivars. It has been hypothesized that exogenous application of paclobutrazol and abscisic acid may suppress the plant height and improve the productivity of two canola cultivars Hybrid Canola (45S42) and Inbred Sandal Canola.

MATERIALS AND METHODS

Experimental Location, Setup, Design, and Treatments

The field experiment was conducted during the winter season of 2019-2020. The investigational area is at 31° North latitude, 73° East longitude, at an elevation of 186 meters from sea level (Agronomy Farm, University of Agriculture, Faisalabad). The experimental design was randomized complete block design (RCBD) under factorial arrangement and each treatment was replicated thrice. The treatments of the experiment included a) control, b) water spray (WS), c) paclobutrazol (50 mgL^{-1}), d) abscisic acid (0.5 mM), and e) paclobutrazol (Pbz=50 mg L^{-1}) + abscisic acid (ABA=0.5mM). The exogenous application of these growth retardants was done separately and combined on the canola cultivars Hybrid 45S42 and Inbred Sandal Canola at the bud initiation stage only once. The seedbed was prepared with a cultivator followed by planking. The net plot size of the trial plot was 4 m × 1.8 m. Seeds of both cultivars were sown in mid of October by using a 4.5 kg ha^{-1} seed rate with the help of drill at the row spacing of 45 cm. The nitrogen and phosphorus fertilizer doses at 90:60 kg ha^{-1} were applied to fulfil the nutrient requirement. Three irrigations were applied throughout the experiment. Standard plant protection measures were adopted to protect crops from insects and diseases. For weed control, hand weeding was carried out after 35 days.

Soil Analysis

Before the sowing of the crop, soil samples were collected from the experimental site by using an auger to different depths from 0 to 30 cm. The samples were air-dried, ground, and passed through a 2 mm strainer and were analyzed (Table 1).

Table 1. Physicochemical properties of experimental soil

Sand (%)	60
Silt (%)	14
Clay (%)	26
Textural Class	Sandy clay Loam
EC (dSm^{-1})	1.59
pH	7.9
Organic Matter	0.72
Total Nitrogen	0.051
Available P (ppm)	5.08
Available K (ppm)	172

Morphological Attributes

The morphological attributes were determined according to standard procedures. The leaf area index was calculated by using a leaf area meter having model number (LI-3100). The stem diameter was measured with the help of a Vernier caliper. Plant height was recorded by using measuring tape from the soil level to the growing tip of the plant. Five plants were used from every plot randomly for the number of branches per plant counted physically. For seed yield, one line of 1 m was harvested at maturity from each block, then threshed and weighted out then converted into kg ha^{-1} by using the unitary method.

Yield Contributing Attributes

Five plants were used from every plot randomly for the number of siliques per plant. Ten siliques opened in the tray which were taken randomly from each plot and No. of seeds per silique were physically counted. The electrical weight balance was used for thousand-grain weight. Two lines from each plot are harvested and weighed out and then converted into kg ha^{-1}. The seed yield is determined by the harvested two lines from all experimental blocks then threshed and weighted out then converted into kg ha^{-1} by using the unitary method.

The Harvest index was calculated with the help of the flowing formula,

$$Harvest\ Index\ \% = economic\ yield / Biomass \times 100$$

Statistical Analysis

A two-way ANOVA analysis was carried out to find the influence of treatments and cultivars on the growth, and yield-related traits of canola. Tukey's HSD test was used to test the treatments' difference at 5% probability level (Steel et al., 1997). The clustered heat map and correlation matrix were done by the use of R-Studio and Origin software respectively.

RESULTS

Morphological attributes

Morphological attributes were significantly affected by the exogenous application of growth retardants in both cultivars of canola (Tables 2 and 3). The interaction between the cultivars × exogenous application was significant for total dry matter, stem diameter and plant height while non-significant for other morphological attributes (Table 5). Average across cultivars, exogenous application of paclobutrazol and abscisic acid increased the maximum leaf area index, stem diameter, number of branches, and total dry matter while causing a reduction in the plant height as compared to control conditions. When we used them separately their effect was less significant in most of the cases. However, the combined application of paclobutrazol and abscisic acid showed a significant improvement in these morphological attributes such as maximum leaf area index and total dry matter increased by 14% and 10% respectively, while suppressing the plant height by 17% as compared to control conditions. However, among the cultivars the Hybrid 45S42 showed better performance in the above-mentioned morphological attributes as compared to the Inbred Sandal Canola.

Table 2. Effect of exogenous application of abscisic acid and paclobutrazol on the morphological attributes of canola cultivars.

Canola Cultivars	Treatments	LAI	Stem Diameter (cm)	Plant Height (cm)	TDM (g m^{-2})
Hybrid (45S42)	Control	3.03 e	6.43 c	212.11 a	500.19 c
	WS	3.26 bc	5.37 ef	213.14 a	526.35 b
	Pbz	3.15 d	7.26 b	190.33 b	530.63 b
	ABA	3.35 ab	7.31 b	188.54 b	526.64 b
	Pbz+ABA	3.43 a	9.39 a	166.31 d	536.90 a
Inbred Sandal Canola	Control	2.85 f	4.73 f	190.74 b	421.31 h
	WS	2.95 ef	5.66 de	192.10 b	431.17 g
	Pbz	2.96 e	6.13 cd	181.35 c	464.75 e
	ABA	3.21 cd	5.90 cde	180.47 c	457.23 f
	Pbz+ABA	3.27 bc	7.71 b	164.74 d	472.01 d
HSD Value		0.18	1.68	8.77	19.05

WS (Water spray) Pbz (Paclobutrazol), ABA (Abscisic acid), and Pbz + ABA, Difference among values (means of three replicates) show a significant difference across the mean at $p<0.05$ according to the Tukey's HSD test.

Table 3. Effect of exogenous application of abscisic acid and paclobutrazol on the morphological and yield contributing attributes of canola cultivars.

Canola Cultivars	Treatments	Number of Branches/ Plant	Number of Seeds/Silique	Number of Siliques/Plant
Hybrid (45S42)	Control	7.33 de	24 cde	246.67 f
	WS	8.33 d	25 bcd	250.67 ef
	Pbz	9.33 b	26 abc	266.67 bc
	ABA	10.00 c	28 a	270.33 b
	Pbz+ABA	13.33 a	28 a	294.33 a
Inbred Sandal Canola	Control	4.66 f	22 e	231.67 g
	WS	6.66 e	24 de	227.67 g
	Pbz	7.67 d	23 de	249.00 ef
	ABA	8.33 de	25 bcd	256.00 de
	Pbz+ABA	10.67 c	27 ab	261.00 cd
HSD Value		3.20	3.85	12.11

WS (Water spray) Pbz (Paclobutrazol), ABA (Abscisic acid), and Pbz + ABA, Difference among values (means of three replicates) show a significant difference across the mean at $p<0.05$ according to the Tukey's HSD test.

Yield contributing attributes

Yield-contributing attributes were significantly affected by the exogenous application of paclobutrazol and abscisic acid in both cultivars of canola (Tables 3 and 4). The interaction among the cultivars × exogenous application was significant for number of siliques per plant and thousand-grain weight while non-significant for other morphological attributes (Table 5). Average across cultivars, exogenous application of paclobutrazol and abscisic acid increased the number of siliques per plant, number of seeds per silique, thousand grain weight, seed yield, biological yield, and harvest index as compared to control conditions. When we used them separately their effect was less significant in most of the cases. However, the combined application of paclobutrazol + abscisic acid showed a significant increase in these yield-contributing attributes such as the number of siliques per plant and biological yield increased by 16% and 4% respectively as compared to control conditions. However, among the cultivars the Hybrid 45S42 showed better performance in the above-mentioned yield-contributing attributes as compared to the Inbred Sandal Canola.

Table 4. Effect of exogenous application of abscisic acid and paclobutrazol on the yield contributing attributes of canola cultivars.

Canola Cultivars	Treatments	1000 grain weight (g)	Seed yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Harvest index (%)
Hybrid (45S42)	Control	3.93 d	1965 bc	7694.00 abcd	24.92 a
	WS	4.02 d	1973 bc	7744.70 abc	24.97 a
	Pbz	6.65 b	2013 b	7845.00 ab	25.62 a
	ABA	6.77 b	2060 ab	7995.70 a	26.36 a
	Pbz+ABA	9.03 a	2160 a	8071.30 a	26.88 a
Inbred Sandal Canola	Control	1.89 f	1760 e	7141.00 e	24.19 a
	WS	2.70 e	1792 de	7204.00 de	24.40 a
	Pbz	4.99 c	1840 de	7354.30 cde	24.76 a
	ABA	6.44 b	1847 de	7385.00 bcde	24.74 a
	Pbz+ABA	7.11 b	1870 cd	7432.30 bcde	25.14 a
HSD Value		0.79	109.99	490.17	4.73

WS (Water spray) Pbz (Paclobutrazol), ABA (Abscisic acid), and Pbz + ABA, Difference among values (means of three replicates) show a significant difference across the mean at *$p<0.05$* according to the Tukey's HSD test.

Correlation matrix and heat map

The morphological and yield-contributing attributes show strong positive and negative correlations under the influence of the exogenous application of growth retardants and control conditions according to Pearson's correlation analysis as shown in (Fig 1). The maximum leaf area, stem diameter, total dry matter, and the number of branches showed a strong positive correlation with the yield-contributing attributes in both cultivars of canola. However, these aforementioned indices showed a strong negative correlation with the plant height of both cultivars of canola.

Heat map analysis was done among the morphological and yield contributing attributes in both cultivars of canola. The interaction strength between the recorded aforementioned attributes and the treatments was shown by variation in the color of boxes of the heat map. The colors from dark blue (positive strongly) to dark red (negative strongly) in the scale of the heat map closely resemble the boxes colors. The highest enhancement in the morphological and yield contributing while reduction in the plant height was observed in the treatment V1T4 followed by the treatment V2T4. The treatment V2T0 showed a significant reduction in these attributes (Fig. 2).

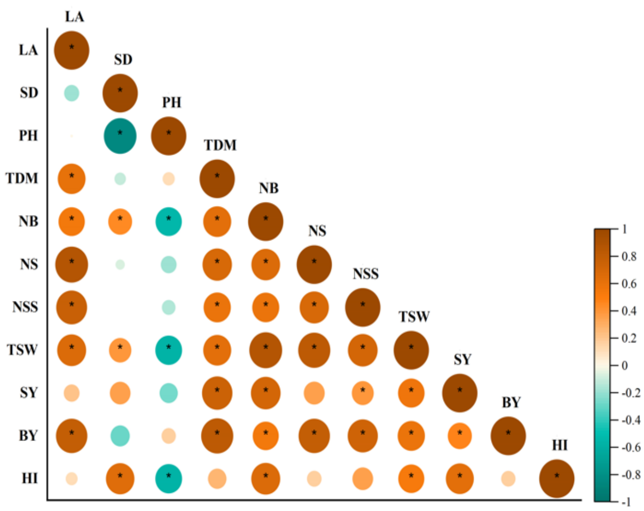

Figure 1. Pearson correlation analysis among the morphological and yield contributing indices of both cultivars of canola. The abbreviations of the attributes are leaf area index (LAI), stem diameter (SD), number of branches per plant (NB), number of silique per plant (NS), number of seeds per silique (NSS), thousand grain weight (TGW), seed yield (SY), biological yield (BY), and harvest index (HI).

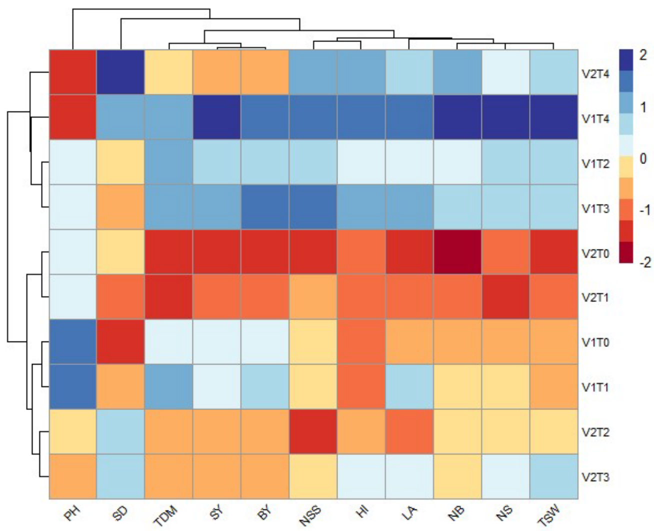

Figure 2. Heat map analysis showed the morphological and yield contributing indices of both cultivars of canola. Please see the caption of Fig. 2 for abbreviations of the attributes while in the group of treatment T0 (Control), T1 (Water spray), T2 (Paclobutrazol), T3 (Abscisic acid), and T4 (Paclobutrazol + Abscisic acid). The canola cultivars are represented by V1 (Hybrid 45S42) and V2 (Inbred Sandal Canola).

Table 5. Analysis of variance regarding the effect of exogenous application of abscisic acid and paclobutrazolon the yield contributing attributes of canola cultivars.

Attributes	Cultivar	Treatment	Cultivar × Treatment
LAI	0.284***	0.166***	0.006^{ns}
No. of Branches	32.03***	29.78***	0.45^{ns}
No. of seeds/silique	34.13***	14.63***	2.13 ^{ns}
No. of siliques/plant	3203.33***	1633.63***	92.67**
Plant Height	1117.28***	1374.23***	113.53***
Stem Diameter	9.50***	9.04***	1.02***
TDM	41716.30***	1810.60***	245.50**
1000 seed weight	16.13***	28.93***	0.72***
Seed Yield	338553***	21585***	3470^{ns}
Biological Yield	2409467***	119148**	5178^{ns}
Harvest Index	5.04^{ns}	12.09**	0.59^{ns}

ns=non-significant, significant at p≤0.05*, p≤0.01**, p≤0.001***

DISCUSSION

The morphological attributes such as maximum leaf area index, stem diameter, number of branches, and total dry matter were significantly increased while plant height was reduced with the exogenous application of growth retardants (paclobutrazol, abscisic acid, and their combinations) in both cultivars of canola as compared to control conditions (Table 2 and 3) as reported in the previous studies (Hazar and Bora, 2018 and Desta and Amare, 2021). These findings are in line with Li et al. (2020), who reported that the use of plant growth retardants can decrease plant height. The decline in plant height is primarily due to a decrease in the elongation of the internodes. It is also observed that when growth retardant was applied, the upper internodes were shortened (Syaputra et al., 2013 and Tesfahun and Menzir, 2018). By preventing stem expansion, growth retardants in rapeseed promote lateral growth and fruit/flower development, producing bushier plants with more flowers (Ijaz et al., 2015). Our research also indicated that the hybrid canola cultivar (45S42) showed maximum values of these traits than the inbred canola cultivar (Sandal). These results are similar to Fernandez et al. (2023) who reported that hybrids have the potential advantage to perform better as compared to inbred cultivars.

In both cultivars of canola yield attributes such as the number of seeds per silique, 1000 grain weight, seed yield, biological yield, and harvest index significantly increased with the exogenous application of growth retardants (paclobutrazol, abscisic acid, and their combination) as compared with control conditions (Table 3 and 4) as found in the previous investigations (Qian-Mei et al., 2023). Ali et al. (2024) also reported that the use of foliar application of paclobutrazol (a compound present in triazole fungicide) increased the yield of canola (*Brassica napus* L.). The number of branches and pods per plant increased because of the growth retardant treatment. Furthermore, there was a significant increase in seed yield because when a plant does not use all of its energy to elongate its stems, it can use that energy to produce more flowers, fruits or seeds, and the parts of the plant that humans typically harvest for yield (Ahmad et al., 2023 and Sima et al., 2024). It is also reported that the application of growth regulators can be used to increase stem diameter, number of branches per plant, number of siliques per plant, number of seeds per silique, and grain weight. These findings were similar to the results of Mccaskill et al. (2019) who reported that the use of plant growth retardants can reduce plant height and increase crop yield by reducing lodging and shattering losses.

CONCLUSION

The present study revealed that the use of plant growth retardants such as paclobutrazol and abscisic acid had beneficial effects on plant growth and yield contributing attributes leading to improved yield of canola Furthermore, overly large canopies were efficiently handled via the use of plant growth retardants, which reduced plant height and promoted the formation of seeds and productivity. Better results were obtained when paclobutrazol and abscisic acid were applied in combination, as compared to their alone application. Overall, foliar Pbz and ABA treatment shows potential for increasing canola yields in a sustainable manner. Future studies should examine their processes, the best times to apply them, and the approaches that work best for various cultivars and environments. These tools have complicated and situation-specific impacts, but they may be useful parts of climate-smart canola production systems.

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