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Foliar Application of Growth Regulators Mitigates Harmful Effects of Drought Stress and Improves Seed Yield and Oil Quality of Rapeseed (*Brassica napus* L.)

Seyed Ahmad Kalantar Ahmadi[1] · **Hamed Eyni-Nargeseh**[2]

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Abstract

This study aimed to investigate the effect of plant growth regulators (PGRs) for improving the quantity and quality of rapeseed oil under different irrigation conditions. A 2-year experiment was conducted in a split-plot arrangement based on a randomized complete block design with three replications over 2 years (2015–16 and 2016–17) at Safiabad Agricultural and Natural Resources Research and Education Center. Main plots consisted of three irrigation regimes (optimal irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting) and subplots included 10 levels of foliar application of PGRs (100, 200, and 300 mg l^{-1} ascorbic acid; 100, 200, and 300 µM salicylic acid; 10, 20, and 30% [w/v] methanol; and foliar application of distilled water as control). The studied cultivar was Hyola401. Restricted irrigation from flowering and silique-setting stages led to decreases in rapeseed seed yield by 38 and 15% as compared with the full irrigation regime, respectively. The interaction of irrigation regime × foliar application showed that spraying ascorbic acid (300 mg l^{-1}) maximized the seed yield under optimal irrigation (4493.33 kg ha^{-1}) and restricted irrigation from the silique-setting stage (3884.72 kg ha^{-1}). Under restricted irrigation from the flowering stage, foliar spray of methanol (30%) produced the highest seed yield (2667.77 kg ha^{-1}). Restricted irrigation from flowering and silique stages resulted in a decrease in oil content by 9.28 and 5.83%, respectively. Overall, foliar application of ascorbic acid, salicylic acid, and methanol improved the seed yield and oil quality of rapeseed under optimal water supply and drought-stress regimes.

Keywords Ascorbic acid · Abiotic stress · Fatty acid profiling · Methanol · Salicylic acid

Plants are exposed to different environmental stresses and drought is one of the most important abiotic stresses influencing plant growth and development (Seleiman et al. 2021; Aliyari Rad et al., 2021; Jamshidi Zinab et al. 2022). Drought stress due to waste of water by evapotranspiration or depletion of available soil water adversely affects the growth of plants (Shafiq et al. 2014) and limits plant growth and yield in arid and semiarid regions (Kalantar Ahmadi et al. 2015a; Sun et al. 2013).

Rapeseed is a major oilseed crop, which is cultivated globally and ranks as the third largest oilseed crop worldwide, just behind palm and soybean (FAO 2020). Rapeseed seed yield is affected by the events prior to and during the flowering stage, but the reproductive period is more sensitive to drought stress (Khodabin et al. 2020). Plant growth depends on water uptake from the soil by roots and transfer to other plant parts (Mohammadi Alagoz et al. 2022). The reduction in growth characteristics when plants are subjected to drought stress could be due to the negative effects of drought stress on physiological and biochemical activities (Heshmat et al. 2021). Plants are susceptible to water-deficit stress during the flowering stage and exposure to drought stress during this stage causes fewer flowers and silique and flower abortion (El-Sabagh et al. 2017; Farahani et al. 2019; Zhu et al. 2021). A short period of drought stress can limit rapeseed seed yield during reproductive

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✉ Seyed Ahmad Kalantar Ahmadi
a.kalantarahmadi@areeo.ac.ir

Hamed Eyni-Nargeseh
heyni@tvu.ac.ir

[1] Department of Agronomy and Horticultural Science, Safiabad Agricultural and Natural Resources Research and Education Center, Agricultural Research, Education and Extension Organization (AREEO), PO Box 333, Dezful, Iran

[2] Department of Agricultural Science, Technical and Vocational University (TVU), Tehran, Iran

growth (Shirani Rad et al. 2021). Moreover, drought stress adversely influences silique formation, which ultimately results in a reduction in seed yield (Johnston et al. 2002). Environmental conditions play a key role in the produced oil content and quality (Zhu et al. 2021), and the effect of drought stress on reducing oil content and quality is well known (Shahsavari and Dadrasnia 2016; Eyni-Nargeseh et al. 2022). The composition of rapeseed fatty acids includes 66% monounsaturated, 27% polyunsaturated, and 7% saturated fatty acids (Safavifard et al. 2018) and determines oil quality. Fatty acid composition is highly affected by environmental conditions, management operations, and length of the grain-filling period (Pritchard et al. 2000; Enjalbert et al. 2013; Mokhtassi-Bidgoli et al. 2022).

Exogenous application of plant growth regulators (PGRs) such as ascorbic acid could substantially mitigate the negative effects of environmental stresses on plant growth and seed yield (Kalantar Ahmadi et al. 2015a; Shafiq et al. 2014). Ascorbic acid is a substantial metabolite involved in plenty of cellular processes including cell division and expansion (Pignocchi and Foyer 2003), enzymatic and nonenzymatic reactions (Kalantar ahmadi et al. 2015b; Smirnoff and Wheeler 2000), and photosynthesis and flowering (Barth et al. 2006). Some reports have indicated that ascorbic acid is an important factor involved in defense mechanisms and exogenous application of ascorbic acid is an effective agronomic method to enhance stress tolerance in different plants (Munir et al. 2013; Ahmad et al. 2012). A significant increase in proline, pigments, growth, and seed yield of rapeseed using foliar application of ascorbic acid ($200\,\mathrm{g}\ \mathrm{l}^{-1}$) was reported by Sakr and Arafa (2009). Improved seed yield caused by applying ascorbic acid can be attributed to increased leaf area, photosynthesis rate, and dry matter. Although the effect of ascorbic acid on fatty acids is not clearly understood, its application led to an increase in linolenic acid and a reduction in oleic acid (Bybordi 2012).

Salicylic acid is an important phenolic compound affecting various physiological aspects of crops (Hayat et al. 2010; Singh et al. 2017). Salicylic acid promotes uptake and translocation of nutrients (Khan et al. 2003), photosynthesis rate, and growth (Farhangi-Abriz and Ghassemi-Golezani 2016), as well as contributing to drought tolerance (Kalantar ahmadi et al. 2015b) through enhancement of leaf pigments, carboxylase activity of rubisco (Singh and Usha 2003), and scavenging of reactive oxygen species (ROS) due to an increase in the activity of antioxidant enzymes (Ghassemi-Golezani et al. 2019; Kalantar ahmadi et al. 2015b). Improvements in seed yield and oil quality of crops by foliar spray of salicylic acid under drought-stress conditions were reported by Ghassemi-Golezani et al. (2019) and Razmi et al. (2017). Moreover, oil content and fatty acids such as linoleic acid, linolenic acid, stearic acid, and total unsaturated fatty acids of rapeseed are influenced by interaction effects of salicylic acid and drought stress (Estaji and Niknam 2020).

In the early 1990s, it was reported that applying methanol is a way to improve yield and reduce water demands in crops (Nemecek-Marshall et al. 1995). Methanol plays a vital role in preventing increased photorespiration induction in stressed plants (Moran et al. 1994) and reducing crop water requirements (Madhaiyan et al. 2006). Among the solutions to diminish the adverse effect of drought stress on crops, foliar application of methanol is to reduce photorespiration and increase CO_2 fixation, which lead to improved crop yield under drought stress, and use of methanol as a carbon source has been reported to achieve this goal (Nonomura and Benson 1992; Abbasian et al. 2016). Some researchers have shown that application of methanol resulted in an increase in growth (Dewez et al. 2003), chlorophyll content, leaf weight and relative water content (Vojodi et al. 2017), acceleration of maturity (Ramirez et al. 2006), and change in the activity of enzymes (Kalantar Ahmadi et al. 2015a).

Rapeseed is one of the most important oilseed crops in many parts of the world and implementation of crop management practices has great importance to improve its seed yield and oil quality. Hyola401 is a spring commercial cultivar that is commonly cultivated by Iranian farmers and can potentially produce an acceptable oil content and yield under water-limited and head-stress conditions in Khuzestan province, Iran. Over recent years, the water shortage crisis has increasingly affected the agricultural sector in Iran, especially in Khuzestan province due to its hot and dry climate. It is always a great challenge for farmers to irrigate fall crops at the end of the growing period and spring crops at the beginning of the growing period. Withholding irrigation would be considered a management strategy to save water and allocate it to spring crops. On the other hand, the negative effects of withholding irrigation on quantitative and qualitative yields should be minimized. Since the PGRs involved directly or indirectly impact quantitative and qualitative characteristics and mitigate the adverse effects of environmental stresses on plants, the aim of this research was to investigate the beneficial effects of foliar application of ascorbic acid, salicylic acid, and methanol on seed yield, oil content, and fatty acid composition of rapeseed under optimal water supply and drought-stress conditions.

Materials and Methods

Experimental Site

The present field experiment was conducted in Safiabad Agricultural and Natural Resources Research and Educa-

Table 1 Characteristics of physical and chemical properties of soil used in the experiment

Year	Soil texture	OC (%)	P (ppm)	K (ppm)	pH	EC (ds/m)	Zn (ppm)	Mn (ppm)	B (ppm)
2015	Clay-loam	0.62	8.5	178	7.64	0.57	0.71	4.5	0.6
2016	Clay-loam	0.65	10.1	185	7.61	0.56	0.73	4.2	0.62

OC organic carbon, *EC* electrical conductivity

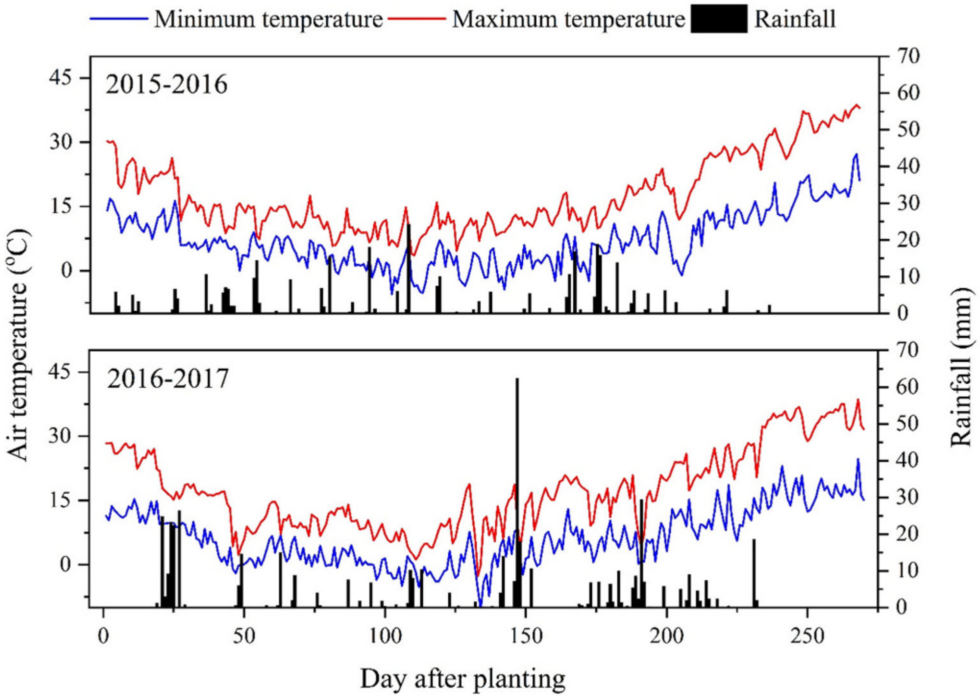

Fig. 1 Daily climatic parameters (air maximum and minimum temperatures and rainfall) over the growing periods (2015–2016 and 2016–2017) of rapeseed in Dezful

tion Center located in Khuzestan province in southwest Iran (82.9 masl, 48° 26′ E, 32° 16′ N) over 2 years (2015–2016 and 2016–2017). The soil characteristics are shown in Table 1. The climatic data including minimum and maximum temperatures and rainfall are provided in Fig. 1.

Experimental Design

A split-plot arrangement of treatments was used based on a randomized complete block design with three replications. Main plots were three levels of irrigation including full irrigation after 70 mm evaporation from a class A evaporation pan (control), restricted irrigation from the flowering stage (BBCH scale: 61) until the end of the growing season, and restricted irrigation from the appearance of siliques (BBCH scale: 75) until the end of the growing season. Subplots consisted of 10 levels of foliar application of PGRs (100, 200, and 300 mg l^{-1} ascorbic acid; 100, 200, and 300 µM salicylic acid; 10, 20, and 30% [w/v] methanol; and foliar application of distilled water as control). Hereafter, the foliar applications of PGRs are abbreviated to AsA100, AsA200, AsA300, Sa100, Sa200, Sa300, Me10, Me20, Me30, and control, respectively. It should be noted that the PGRs were applied at two stages of rosette (BBCH scale: 30) and opening of the first flower (BBCH scale: 61; Weber and Bleiholder 1990). PGRs used were manufactured by Merck, Darmstadt, Germany.

Experiment Details

Each experimental plot consisted of eight 6-m lines with an interrow distance of 30 cm and a density of 80 plants m^{-2}. According to the results of soil analysis and fertilizer recommendations, 200 kg ha^{-1} potassium sulfate, 150 kg ha^{-1} triple superphosphate, and 390 kg ha^{-1} urea were applied. All potassium sulfate, triple superphosphate, and one third of the urea were used before planting, and the remaining urea was applied equally at two stages of stem elongation (BBCH scale: 30) and flowering (BBCH scale: 61). Hyola401 genotype was sown on 05 November in two study years. Weeds were controlled by applying 2.5 l ha^{-1} Botizan star at the cotyledon stage (BBCH scale: 09) and by hand weeding as needed during the growth period. Irrigation intervals were adjusted on the basis of 70 mm water evaporation from the class A evaporation pan under standard conditions and the quantity of the water used was 80% of the evaporated water. The water volume entering the field was measured by water meter. Accordingly, the full irri-

Table 2 Irrigation dates and amounts of experimental fields used in the model calibration and validation steps

Year	Irrigation regime	Amount of irrigation (m^3)	Irrigation date							
First year (2015–2016)	IR1	4480	6 Nov 2015	24 Nov 2015	6 Jan 2016	14 Feb 2016	11 Mar 2016	30 Mar 2016	13 Apr 2016	24 Apr
	IR2	2800	6 Nov 2015	24 Nov 2015	6 Jan 2016	14 Feb 2016	29 Feb 2016	–	–	–
	IR3	3360	6 Nov 2015	24 Nov 2015	6 Jan 2016	14 Feb 2016	11 Mar 2016	21 Mar 2016	–	–
Second year (2016–2017)	IR1	3920	6 Nov 2016	18 Nov 2016	20 Dec 2016	23 Jan 2017	19 Feb 2017	14 Mar 2017	18 Apr 2017	–
	IR2	2240	6 Nov 2016	18 Nov 2016	20 Dec 2016	23 Jan 2017	–	–	–	–
	IR3	2800	6 Nov 2016	18 Nov 2016	20 Dec 2016	23 Jan 2017	19 Feb 2017	–	–	–

IR1, IR2, and *IR3* are full irrigation, restricted irrigation from flowering stage, and restricted irrigation from silique setting stage, respectively

gation, restricted irrigation from flowering, and restricted irrigation from silique setting applications received 4480, 2800, and 3360 $m^3\ ha^{-1}$ in the first year and 3920, 2240, and 2800 $m^3\ ha^{-1}$ in the second year, respectively. The irrigation dates of experimental treatments are provided in Table 2.

Agronomic Traits

To calculate seed yield, four center rows in each plot were separately cut off at the physiological maturity stage (BBCH scale: 89) in an area of $6\ m^2$. Ten plants were randomly selected from each plot, and the number of siliques per plant, seed number per silique, and thousand-seed weight were measured in three replications.

Oil Content and Fatty Acid Composition

To measure oil content, 5-g seed samples were selected from each treatment (three replications) and nuclear magnetic resonance (NMR) was used to estimate the oil content (minispec mq20; Bruker, Bremen, Germany; International Standard [ISO] 5511 1992) at the laboratory of the Seed and Plant Improvement Institute, Karaj, Iran. Fatty acid methyl esters obtained from the oil samples were analyzed using gas chromatography (GC; Agilent Technologies, Santa Clara, CA, US) according to the method proposed by Azadmard-Damrichi et al. (2005): 2 ml of 0.01 M NaOH in methanol was added to the oil sample dissolved in 0.5 ml hexane. Thereafter, it was kept in a water bath at 60 °C for 10 min. In the next stage, B trifluoride in methanol (20% of BF3 in methanol) was mixed with it and left in the water bath at 60 °C for 10 min. Afterwards, samples were cooled under the water and 2 ml of sodium chloride (20% w/v) and 1 ml hexane were added. Mixing completely, the hexane layer which contained the fatty acid methyl esters was separated by centrifugation. The fatty acid methyl esters were analyzed by GC according to the method described by Azadmard-Damirchi and Dutta (2006). Glucosinolate was measured using a spectrophotometer (model UV2100, Company Unike, USA) equipped with a 50-m CP-Sil 88 capillary column, an inner diameter of 0.25, and 0.2 µm of a static thickness (Makkar et al. 2007). All materials used were manufactured by Merck, Germany.

Statistical Analysis

After conducting Bartlett's test and supplying the homogeneity of the test variance in each year, the combined analysis of variance was conducted using SAS software (version 9.2; SAS Institute, Cary, NC, US). The least significant difference (LSD) was applied to separate the means of the main effect and the significant interactions were assessed by the LSD test by the slicing method at $P < 0.05$. The significance of linear and quadratic regression models was tested with polynomial orthogonal contrasts. The regression models are given when orthogonal contrasts were significant at the $P \leq 0.05$ probability level.

Results

Combined analysis of variance revealed that the simple effect of year was significant for all studied traits, except oil content. Simple effects of irrigation regime and foliar spray of PGRs were significant for oil content. The interaction of year × irrigation regime was significant for number of siliques per plant, number of seeds per silique, seed yield, and stearic acid, linolenic acid, and glucosinolate content. Seed yield and palmitic acid were affected by the interaction of year × foliar spray of PGRs. Combined analysis showed that the interaction of irrigation regime × foliar spray was statistically significant for the number of siliques per plant, number of seeds per silique, thousand-seed weight, seed yield, and palmitic acid, stearic acid, linolenic acid, and glucosinolate content. Amounts of oleic, linoleic, and erucic acids were impacted by the three-way interaction of year × irrigation regime × foliar spray of PGRs (Table 3).

Number of Siliques per Plant

The means comparison of the two-way interaction of year × irrigation regime revealed that the highest number of siliques per plant was with optimal irrigation, averaging

Table 3 Combined analysis of variance for studied traits as affected by irrigation regime and foliar application of plant growth regulators over two years (2015–2016 and 2016–2017) in Dezful, Iran

S.O.V	Df	No. of siliques per plant	No. of seeds per silique	Thousand-seed weight	Seed yield	Oil content	Palmitic acid	Stearic acid	Oleic acid	Linoleic acid	Linolenic acid	Erucic acid	Glucosinolate content
Y	1	8595.34**	436.42**	1.46**	1,007,905.23**	3.09ns	6.94**	4.96**	1173.97**	140.94**	115.46**	0.03**	215.53**
Y (block)	4	36.51	20.12	2.14	453,255.45	10.2	0.03	0.09	7.5	0.82	2.94	0.0003	2.93
IR	2	9822.27**	106.75**	2.06**	30,149,900.5**	263.16**	15.85**	5.4**	4313.25**	191.88**	73.14**	0.0003**	246.63**
Y × IR	2	463.01**	51.94**	0.32ns	1,547,756.23**	7.03ns	0.04ns	0.18**	75.97**	7.93**	0.27**	0.000014**	2.87**
Ea	8	36.85	17.11	1.84	798,487	14.59	0.04	0.19	7.83	0.53	0.11	0.000012	3.99
PGR	9	496.36**	30.64**	9.17**	1,370,533.53**	16.05**	0.09**	0.1**	13.01**	2.51**	4.52**	0.00016**	4.04**
Y × PGR	9	43.44ns	0.55ns	0.00002ns	288,303.33*	0.18ns	0.18**	0.007ns	1.7*	1.02**	0.007ns	0.0000016ns	0.18ns
IR × PGR	18	62.54*	22.24**	21.01**	308,233.95**	2.22ns	0.07**	0.03**	6.14**	1.05**	0.39**	0.000008**	0.53**
Y × IR × PGR	18	17.3ns	0.55ns	0.00005ns	99,360.11ns	0.09ns	0.03ns	0.01ns	1.53*	0.49**	0.006ns	0.000013**	0.17ns
Eb	108	35.67	8.73	0.17	131,519.3	3.54	0.03	0.01	0.79	0.12	0.01	0.00002	0.18
CV %	–	11.94	10.46	11.34	11.86	4.44	4.51	6.33	1.83	1.75	1.85	3.51	7.61

ns not significant, * and ** significant at 5% and 1% probability level, respectively

Y year, *E* error, *IR* irrigation regime, *PGR* plant growth regulators, *CV* coefficient of variation, *df* degrees of freedom

51.1 and 70.8 in the first and second years, respectively. Rapeseed plants grown under restricted irrigation from the flowering stage had the lowest number of siliques per plant, averaging 46.5 and 59.6 in the first and second years, respectively (Table 4). Means comparison of the two-way interaction of irrigation regime × foliar application of PGRs showed that while the maximum number of siliques per plant (71) was detected in foliar spray of Sa200 under the full irrigation regime, the highest number of siliques per plant under restricted irrigation from flowering (46.83) and silique setting (63.16) stages was with foliar spray of AsA300 (Table 5).

Number of Seeds per Silique

The means comparison of the interaction of year × irrigation regime showed that the highest number of seeds per silique was obtained from the optimal irrigation regime in both years (27.0 and 32.3 in first and second years, respectively; Table 4). The means comparison of the two-way interaction of irrigation regime × foliar application of PGRs indicated that the highest number of siliques per plant (32.04) was observed with foliar spray of AsA100 when the rapeseed plants were fully irrigated, while the Sa300 and AsA200 foliar spray treatments produced the maximum number of seeds per silique, with averages of 32.88 and 29.59 under restricted irrigation from flowering and silique-setting stages, respectively (Table 5).

Thousand-Seed Weight

The means comparison of the simple effect of year showed that thousand-seed weight was 3.74 and 3.56 g in the first and second years, respectively (data not shown). The means comparison of the interaction effect of irrigation regime × foliar spray of PGRs illustrated that the AsA200, Me30, and Sa200 foliar spray treatments produced the maximum thousand-seed weight, averaging 4.22, 4.28, 4.20 g when rapeseed plants were grown under optimal water

Table 4 Means comparison of the two way-interaction of year × irrigation regime for study traits of rapeseed in 2015–2016 and 2016–2017

–	No. of siliques per plant		No. of seeds per silique		Seed yield (kg ha^{-1})		Stearic acid (%)	
–	2015–16	2016–17	2015–16	2016–17	2015–16	2016–17	2015–16	2016–17
IR1	51.1 ± 1.33a	70.8 ± 1.22a	27.0 ± 0.49 cd	32.3 ± 0.46a	3503 ± 100.9b	3931 ± 85.7a	1.90 ± 0.02b	1.45 ± 0.03d
IR2	31.6 ± 1.04c	40.2 ± 1.39c	27.2 ± 0.65 cd	29.1 ± 0.64b	2207 ± 92.1d	2409 ± 78.3d	2.16 ± 0.02a	1.92 ± 0.04b
IR3	46.5 ± 1.97b	59.6 ± 1.38b	25.7 ± 0.7d	28.2 ± 0.57bc	3036 ± 111.4c	3259 ± 82.4bc	1.6 ± 03c	1.30 ± 0.03e
–	Linolenic acid (%)		Erucic acid (%)		Glucosinolate (μmol g^{-1})		–	–
–	2015–16	2016–17	2015–16	2016–17	2015–16	2016–17	–	–
IR1	5.21 ± 0.07d	3.57 ± 0.07f	0.055 ± 0.00007b	0.026 ± 0.0006d	4.71 ± 0.12c	2.82 ± 0.14d	–	–
IR2	6.42 ± 0.13b	4.71 ± 0.12e	0.058 ± 0.001a	0.031 ± 0.0009c	9.14 ± 0.17a	6.45 ± 0.16b	–	–
IR3	7.32 ± 0.1a	5.87 ± 0.11c	0.059 ± 0.0008a	0.030 ± 0.0009c	6.37 ± 0.14b	4.38 ± 0.13c	–	–

Small-case letters refer to the means comparison for the overall interactions based on the slicing method at the 5% probability level according to LSMeans. The value after ± is standard error

IR1, IR2, and *IR3* are full irrigation, restricted irrigation from flowering, and restricted irrigation from silique-setting stages, respectively

Table 5 Means comparison of the two way-interaction of irrigation regime × plant growth regulators for study traits of rapeseed during two study years (2015–2016 and 2016–2017)

Irrigation regime	Plant growth regulator	No. of siliques plant^{-1}	No. of seeds silique^{-1}	Thousand-seed weight (g)	Seed yield (kg ha^{-1})	Palmitic acid (%)	Stearic acid (%)	Linolenic acid (%)	Glucosinolate (μmol g^{-1})
IR1	AsA100	52.68 ± 4.74c	32.04 ± 1.71a	3.88 ± 0.09ab	3691.11 ± 129.67bc	3.44 ± 0.32b	1.66 ± 63.76b	4.38 ± 0.11c	4.05 ± 0.02bc
	AsA200	57.48 ± 4.11bc	29.96 ± 1.35ab	4.22 ± 0.11a	3801.66 ± 168.94b	3.46 ± 0.16b	1.75 ± 0.08ab	4.45 ± 0.39c	3.71 ± 0.45c
	AsA300	66.12±.24ab	30 ± 1.23ab	2.94 ± 0.09 cd	4493.33 ± 82.03a	3.3 ± 0.22b	1.79 ± 0.1a	4.8 ± 0.4a	3.51 ± 0.44 cd
	Sa100	57.72 ± 4.85bc	31.67 ± 1.53a	3.78 ± 0.26ab	4163.88 ± 142.88ab	3.61 ± 0.09ab	1.69 ± 0.11ab	4.24 ± 0.39d	4.28 ± 0.46b
	Sa200	61.66 ± 6.45b	28.93 ± 1.47ab	3.9 ± 0.12ab	3557.77 ± 207bc	3.56 ± 0.13b	1.65 ± 0.13b	4.44 ± 0.39c	3.89 ± 0.41bc
	Sa300	71 ± 4.23a	28.86 ± 1.37ab	2.57 ± 0.09d	3376.11 ± 338.67c	3.42 ± 0.19b	1.62 ± 0.15bc	4.61 ± 0.4b	3.62 ± 0.43 cd
	Me10	59.61 ± 4.56b	29.68 ± 1.28ab	3.57 ± 0.15b	3448.33 ± 182.16bc	3.68 ± 0.06ab	1.6 ± 0.11bc	4.13 ± 0.38d	3.5 ± 0.5 cd
	Me20	63.88 ± 4.99b	28.2 ± 1.67b	3.31 ± 0.1bc	3524.44 ± 205.28bc	3.76 ± 0.06ab	1.72 ± 0.1ab	4.45 ± 0.36c	3.19 ± 0.46d
	Me30	67.16 ± 4.24ab	29.04 ± 1.84ab	3.87 ± 0.14ab	3801.11 ± 139.78b	3.79 ± 0.1a	1.76 ± 0.1ab	4.65 ± 0.36b	3.07 ± 0.44d
	Control	52.16 ± 3.91c	27.84 ± 1.94b	3.05 ± 0.09c	3212.77 ± 138.47c	3.56 ± 0.13b	1.52 ± 0.13c	3.75 ± 0.34e	4.84 ± 0.66a
IR2	AsA100	33.48 ± 2.65bc	26.53 ± 0.44bc	3.77 ± 0.02b	2145.83 ± 216.01b	4.47 ± 0.06ab	1.97 ± 0.09b	5.54 ± 0.41d	8.11 ± 0.65b
	AsA200	36.47 ± 3.13b	27.85 ± 1.54bc	3.42 ± 0.03bc	2090.27 ± 162.29b	4.46 ± 0.12ab	2.01 ± 0.08b	5.72 ± 0.41c	7.76 ± 0.61bc
	AsA300	46.83 ± 3.01a	25.39 ± 0.5c	3.29 ± 0.1c	2369.44 ± 167.57ab	4.47 ± 0.15ab	2.15 ± 0.06a	6.44 ± 0.41a	7.58 ± 0.62c
	Sa100	36.69 ± 2.69b	26.07 ± 1.63c	3.45 ± 0.14bc	2377.5 ± 287.8ab	4.52 ± 0.11ab	2.1 ± 0.09ab	5.43 ± 0.38d	7.66 ± 0.68bc
	Sa200	37.98 ± 3.86b	27 ± 1.91bc	3.8 ± 0.07ab	2299.44 ± 241.25ab	4.54 ± 0.17ab	2.06 ± 0.11ab	5.69 ± 0.38c	7.27 ± 0.62 cd
	Sa300	39.36 ± 2.79b	32.88 ± 0.46a	3.65 ± 0.03bc	2513.61 ± 186.13ab	4.43 ± 0.2b	2.19 ± 0.08a	6.16 ± 0.38b	6.97 ± 0.69d
	Me10	27.57 ± 1.78c	29.46 ± 1.05b	3.36 ± 0.09bc	2204 ± 120.24b	4.36 ± 0.07b	1.81 ± 0.1c	4.5 ± 0.41e	8.5 ± 0.75ab
	Me20	36.85 ± 0.87b	30.42 ± 0.77ab	4.26 ± 0.09a	2441.94 ± 133.64ab	4.61 ± 0.04ab	2.08 ± 0.08ab	5.65 ± 0.41 cd	7.71 ± 0.75bc
	Me30	36.53 ± 2.83b	29.63 ± 2.15ab	4.28 ± 0.07a	2667.77 ± 165.37a	4.64 ± 0.06a	2.16 ± 0.09a	6.25 ± 0.41b	7.55 ± 0.72c
	Control	57.51 ± 1.12c	26.49 ± 0.5bc	3.33 ± 0.14bc	1968.88 ± 168.46b	4.58 ± 0.1ab	1.91 ± 0.09bc	4.32 ± 0.37f	8.83 ± 0.74a
IR3	AsA100	47.5 ± 3.11c	28.64 ± 1.45ab	3.37 ± 0.2bc	2687.5 ± 110.27c	3.65 ± 0.06ab	1.53 ± 0.06a	6.62 ± 0.35c	5.68 ± 0.47b
	AsA200	50.09 ± 6.74bc	29.59 ± 1.67a	4.12 ± 0.13ab	3014.16 ± 288.04bc	3.74 ± 0.07ab	1.51 ± 0.08a	7.07 ± 0.37b	5.31 ± 0.51bc
	AsA300	63.16 ± 3.29a	25.42 ± 1.5b	4.15 ± 0.16a	3884.72 ± 129.5a	3.73 ± 0.1ab	1.52 ± 0.11a	7.58 ± 0.35a	5.16 ± 0.46c
	Sa100	59.31 ± 4.12ab	23.64 ± 0.95b	3.06 ± 0.28c	3484.16 ± 224.14ab	3.7 ± 0.11ab	1.41 ± 0.09ab	6.32 ± 0.34d	5.18 ± 0.41c
	Sa200	52.25 ± 4.16bc	26.14 ± 1.37b	3.63 ± 0.1b	2981.66 ± 246.36bc	3.83 ± 0.1a	1.44 ± 0.08ab	6.57 ± 0.34c	4.89 ± 0.45c
	Sa300	54.52 ± 6.23b	27.45 ± 1.04ab	4.09 ± 0.28ab	2878.05 ± 123.77c	3.76 ± 0.14ab	1.51 ± 0.07ab	7.15 ± 0.34b	4.94 ± 0.28c
	Me10	51.16 ± 3.38bc	28.2 ± 0.81ab	3.92 ± 00.14ab	3352.83 ± 187.05b	3.55 ± 0.08b	1.24 ± 0.1c	6.05 ± 0.34e	5.52 ± 0.62bc
	Me20	55.16 ± 4.37b	27.73 ± 1.75ab	4.2 ± 0.37a	3134.44 ± 167.49bc	3.67 ± 0.09ab	1.39 ± 0.09b	6.39 ± 0.34d	5.28 ± 0.62bc
	Me30	54.92 ± 4.29b	29.44 ± 1.34ab	3.67 ± 0.3b	3378.05 ± 130.61b	3.68 ± 0.08ab	1.45 ± 0.08ab	6.62 ± 0.34c	5.09 ± 0.57c
	Control	42.66 ± 2.6c	23.31 ± 1.04b	3.48 ± 0.15bc	2676.11 ± 50.51c	3.59 ± 0.1b	1.49 ± 0.06ab	5.63 ± 0.34f	6.69 ± 0.54a

Small-case letters refer to the means comparison for the overall interactions based on the slicing method at the 5% probability level according to LSMeans. The value after ± is standard error

IR1, IR2, and *IR3* full irrigation, restricted irrigation from flowering stage, and restricted irrigation from silique setting stage, respectively; *AsA100, AsA200*, and *AsA300*: 100, 200, and 300 mg l^{-1} ascorbic acid, respectively; *Sa100, Sa200*, and *Sa300*: 100, 200, and 300 μM Salicylic acid, respectively; *Me10, Me20*, and *Me30*: 10, 20, and 30% (w/v) methanol, respectively; *control* distilled water

supply, restricted irrigation from flowering, and restricted irrigation from silique setting, respectively (Table 5).

Seed Yield

Averaged across irrigation regimes, rapeseed seed yield was 2915 and 3199 kg ha^{-1} in the first and second years, respectively; the highest seed yield was obtained from the full irrigation regime in both years (Table 4). The interaction of year × foliar spray revealed that the highest seed yield belonged to foliar spray of AsA300 in the first (3483.33 kg ha^{-1}) and second (3681.66 kg ha^{-1}) years (Table 6).

The results of means comparison illustrated that the rapeseed seed yield responded differently to foliar spray of PGRs under three irrigation regimes, so that the highest seed yield was found in AsA300, Me30, and AsA300 treatments (4493.33, 2667.77, and 3884.72 kg ha, respectively) under full irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting, respectively (Table 5).

Oil Content

The results showed that oil content was higher in the optimal irrigation regime (44.57%) compared to restricted irrigation from flowering (40.43%) and silique-setting (41.97%) stages (data not shown). The simple effect of foliar spray of PGRs on oil content showed that the maximum (43.61%) and minimum (40.64%) oil content belonged to the Sa300 and control treatments, respectively (Fig. 2).

Table 6 Means comparison of the two way-interaction of year × plant growth regulators for seed yield and palmitic acid of rapeseed

	Seed yield (kg ha^{-1})		Palmitic acid (%)	
Plant growth regulator	2015–2016	2016–2017	2015–2016	2016–2017
AsA100	2714.81 ± 218.58c	2968.14 ± 286.67b	4.03 ± 0.14b	3.67 ± 0.17bc
AsA200	2688.88 ± 269.74c	3248.51 ± 295.62b	4.12 ± 0.14ab	3.64 ± 0.16bc
AsA300	3483.33 ± 333.66a	3681.66 ± 327.21a	4.16 ± 0.57ab	3.5 ± 0.21c
Sa100	3403.7 ± 290.63ab	3280 ± 337.68b	4.16 ± 0.14ab	3.73 ± 0.14b
Sa200	2720.36 ± 189.06c	3172.22 ± 291.58b	4.26 ± 0.15a	3.69 ± 0.15bc
Sa300	2746.29 ± 165.07c	3098.88 ± 254.62b	4.2 ± 0.15ab	3.53 ± 0.17c
Me10	3112.4 ± 235.69b	2957.7 ± 255.54b	3.98 ± 0.13b	3.73 ± 0.13b
Me20	2996.29 ± 164.94bc	3070.92 ± 243.37b	4.13 ± 0.13ab	3.9 ± 0.17a
Me30	3333.33 ± 135.6ab	3231.29 ± 249.54b	4.09 ± 0.13b	3.98 ± 0.19a
Control	2625.92 ± 155.11c	2612.59 ± 248.35c	3.99 ± 0.16b	3.82 ± 0.2ab

Small-case letters refer to the means comparison for the overall interactions based on the slicing method at the 5% probability level according to LSMeans. The value after ± is standard error

AsA100, AsA200, and *AsA300*: 100, 200, and 300 mg l^{-1} ascorbic acid, respectively; *Sa100, Sa200*, and *Sa300*: 100, 200, and 300 μM Salicylic acid, respectively; *Me10, Me20*, and *Me30*: 10, 20, and 30% (w/v) methanol, respectively; *control* distilled water

Palmitic Acid

The response of rapeseed to foliar spray of PGRs in both years was different in term of palmitic acid content, so that the Sa200 and Me30 treatments had the highest values of palmitic acid in the first and second years, with averages of 4.26 and 3.98%, respectively (Table 6). The means comparison of the interaction of irrigation regime × foliar spray of PGRs revealed that foliar spray of Me30 had the highest content of palmitic acid under optimal irrigation and restricted irrigation from flowering (3.79 and 4.64%, respectively), while the Sa200 foliar spray treatment produced the highest palmitic acid content (3.83%) when irrigation was restricted from the silique-setting stage (Table 5).

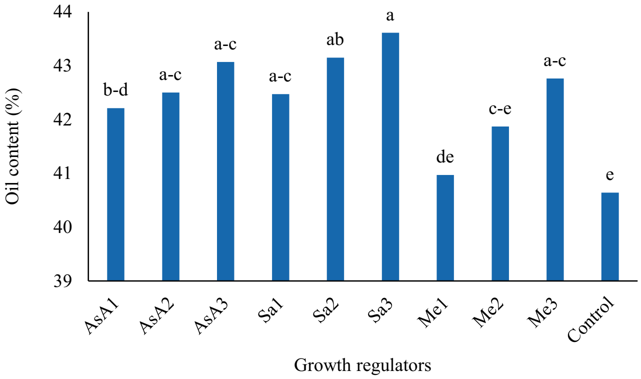

Fig. 2 Means comparison of foliar spray of plant growth regulators for rapeseed oil content. *AsA1*, *AsA2*, and *AsA3*: 100, 200, and 300 mg l^{-1} ascorbic acid; *Sa1*, *Sa2*, and *Sa3*: 100, 200, and 300 μM salicylic acid; Me1, Me2, and Me3: 10, 20, and 30% (w/v) methanol; control distilled water

Stearic Acid

The results of means comparison indicated that stearic acid content was higher in the first year as compared to the second year under three irrigation regimes; the highest value was found in rapeseed plants that experienced restricted irrigation from the flowering stage in the first and second years (2.16 and 1.92%, respectively; Table 4). The foliar spray of PGRs had different influences on stearic acid content under three study irrigation regimes; the AsA300, Sa300, and AsA100 treatments produced the highest stearic content, averaging 1.79, 2.19, and 1.53%, when full irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting regimes were applied, respectively (Table 5).

Oleic Acid

The oleic acid content responded differently to foliar spray of PGRs under three irrigation regimes in two study years. Averaged across irrigation regimes and foliar spray of PGRs, the oleic acid content was 46.76 and 51.04% in first and second years, respectively (Table 7). The maximum oleic acid content (averaged across years and foliar sprays of PGRs) was obtained from the well-watered irrigation regime, averaging 57.53%, and decreases of 29 and 18% were detected under restricted irrigation from flowering and silique setting, respectively (Table 7). Averaged by years, spraying Sa300, Sa200, and AsA300 had the highest oleic acid content, averaging 58.69, 41.71, and 49.54% under full irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting, respectively (Table 7).

Table 7 Means comparison of the two way-interaction of irrigation regime × plant growth regulators for study traits of rapeseed

Irrigation regime	Plant growth regulator	Oleic acid (%)		Linoleic acid (%)		Erucic acid (%)	
		2015–2016	2016–2017	2015–2016	2016–2017	2015–2016	2016–2017
IR1	AsA100	54.75 ± 0.08b	57.91 ± 0.41b	20.55 ± 0.2b	22.44 ± 0.25b	0.0577 ± 0.002b	0.0283 ± 0.009ab
	AsA200	55.25 ± 0.02b	59.55 ± 0.32ab	20.4 ± 0.17b	22.26 ± 0.36b	0.0560 ± 0.002c	0.0266 ± 0.008b
	AsA300	56.1 ± 0.09ab	60.18 ± 0.48a	21.4 ± 0.11a	22.72 ± 0.23b	0.0547 ± 0.003e	0.0248 ± 0.092bc
	Sa100	56.1 ± 0.07ab	59.19 ± 0.44ab	20.45 ± 0.08b	22.9 ± 0.15ab	0.0592 ± 0.002a	0.0300 ± 0.084ab
	Sa200	56.3 ± 0.05ab	59.55 ± 0.39ab	21.35 ± 0.02a	23.46 ± 0.21a	0.0550 ± 0.002d	0.0285 ± 0.092ab
	Sa300	57.6 ± 0.17a	59.79 ± 1.06ab	21.8 ± 0.05a	23.21 ± 0.35ab	0.0544 ± 0.004f	0.0264 ± 0.075bc
	Me10	55.4 ± 0.11b	58.74 ± 0.42b	20.55 ± 0.02b	22.69 ± 0.14b	0.0534 ± 0.004g	0.0246 ± 0.068bc
	Me20	55.35 ± 0.21b	59.21 ± 0.6ab	20.6 ± 0.11b	22.97 ± 0.08ab	0.0533 ± 0.002h	0.0232 ± 0.048bc
	Me30	56.55 ± 0.21ab	59.19 ± 0.57ab	21.5 ± 0.23a	23.24 ± 0.38ab	0.0512 ± 0.006i	0.0227 ± 0.075c
	Control	56.4 ± 0.17ab	57.63 ± 0.27b	20.5 ± 0.05b	21.42 ± 0.09c	0.0550 ± 0.003d	0.0310 ± 0.008a
IR2	AsA100	38.25 ± 0.14a	43.41 ± 0.57ab	18.15 ± 0.08a	19.24 ± 0.08ab	0.0592 ± 0.002e	0.0329 ± 0.084bc
	AsA200	38.15 ± 0.02a	43.66 ± 0.54ab	18.3 ± 0.05a	19.45 ± 0.11ab	0.0584 ± 0.003f	0.0319 ± 0.006bc
	AsA300	38.35 ± 0.14a	42.73 ± 0.32b	18.15 ± 0.08a	19.78 ± 0.17a	0.0581 ± 0.002g	0.0309 ± 0.067bc
	Sa100	38.25 ± 0.14a	42.47 ± 0.31b	17.55 ± 0.14b	18.76 ± 0.17bc	0.0633 ± 0.002c	0.0350 ± 0.079ab
	Sa200	39.45 ± 0.29a	43.98 ± 0.11a	18.25 ± 0.08a	18.93 ± 0.24b	0.0645 ± 0.004b	0.0338 ± 0.058b
	Sa300	39.3 ± 0.11a	44.52 ± 0.31a	18.4 ± 0.17a	19.18 ± 0.37b	0.0656 ± 0.003a	0.0334 ± 0.59b
	Me10	38.25 ± 0.31a	42.12 ± 0.21bc	16.35 ± 0.2c	18.01 ± 0.2c	0.056 ± 0.004h	0.0297 ± 0.082c
	Me20	38.5 ± 0.14a	42.76 ± 0.33b	17.4 ± 0.05b	18.22 ± 0.11c	0.0529 ± 0.008i	0.0276 ± 0.028c
	Me30	39.45 ± 0.02a	43.86 ± 0.12ab	17.5 ± 0.17b	18.14 ± 0.13c	0.0517 ± 0.009j	0.0255 ± 0.035c
	Control	36.1 ± 0.08b	41.01 ± 0.49c	17.4 ± 0.11b	17.87 ± 0.16c	0.0594 ± 0.007d	0.0379 ± 0.082a
IR3	AsA100	45.35 ± 0.14a	51.47 ± 0.41ab	18.45 ± 0.02bc	20.49 ± 0.21c	0.0614 ± 0.006c	0.0317 ± 0.063b
	AsA200	46.2 ± 0.05a	52.16 ± 0.54a	18.5 ± 0.11bc	20.88 ± 0.13bc	0.0594 ± 0.091d	0.0302 ± 0.075b
	AsA300	46.6 ± 0.05a	52.48 ± 0.46a	19.5 ± 0.14a	20.98 ± 0.25bc	0.0584 ± 0.081f	0.0290 ± 0.084bc
	Sa100	41.8 ± 1.5bc	50.98 ± 0.14b	18.1 ± 0.46c	20.46 ± 0.28c	0.0628 ± 0.009b	0.0359 ± 0.073a
	Sa200	43 ± 1.9b	51.68 ± 0.21ab	18.85 ± 0.43b	20.63 ± 0.15c	0.0613 ± 0.008c	0.0345 ± 0.086ab
	Sa300	43.15 ± 2.33b	51.53 ± 0.53ab	19.05 ± 1.12ab	21.37 ± 0.23b	0.0593 ± 0.12e	0.0350 ± 0.046ab
	Me10	39.85 ± 2.4c	48.51 ± 0.15d	17.65 ± 0.54d	21.31 ± 0.12b	0.0575 ± 0.081g	0.0280 ± 0.057bc
	Me20	41.1 ± 1.73c	49.65 ± 0.25c	18.25 ± 0.54c	21.85 ± 0.16ab	0.056 ± 0.005h	0.0261 ± 0.064c
	Me30	42.15 ± 1.52bc	50.55 ± 0.45bc	18.35 ± 0.66bc	22.28 ± 0.19a	0.0555 ± 0.008i	0.0250 ± 0.091c
	Control	39.85 ± 2.14c	50.87 ± 0.29b	19.2 ± 0.71ab	20.29 ± 0.09c	0.0657 ± 0.094a	0.0313 ± 0.081b

Small-case letters refer to the means comparison for the overall interactions based on the slicing method at the 5% probability level according to LSMeans. The value after ± is standard error

IR1, IR2, and *IR3* full irrigation, restricted irrigation from flowering stage, and restricted irrigation from silique setting stage, respectively; *AsA100, AsA200*, and *AsA300*: 100, 200, and 300 mg l^{-1} ascorbic acid, respectively; *Sa100, Sa200*, and *Sa300*: 100, 200, and 300 µM Salicylic acid, respectively; *Me10, Me20*, and *Me30*: 10, 20, and 30% (w/v) methanol, respectively; *control* distilled water

Linoleic Acid

Different responses were found for the three studied irrigation regimes when PGRs were sprayed on rapeseed plants. Averaged by irrigation regime and foliar sprays of PGRs, linoleic acid content was 19.08 and 20.84% in the first and second years, respectively (Table 7). The highest linoleic acid content (averaged across years and foliar sprays of PGRs) was recorded in the well-watered irrigation regime, averaging 21.82%, and decreases of 16 and 9% were observed under restricted irrigation from flowering and silique setting, respectively (Table 7). Averaged across years, spraying Sa300, AsA300, and Me30 produced the highest linoleic acid content, averaging 22.50, 18.96, and 20.31%, under full irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting, respectively (Table 7).

Linolenic Acid

The results of comparing means showed that the highest linolenic acid content belonged to restricted irrigation from the silique-setting stage in the first (7.32%) and second years (5.87%), respectively (Table 4). Although the rapeseed plants responded differently to foliar spray of PGRs under irrigation regimes in term of linolenic acid content, the AsA300 foliar spray treatment maximized the linolenic acid content under full irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting c, averaging 4.8, 6.44, and 7.58%, respectively (Table 5).

Erucic Acid

Means comparison of the three-way interaction of year × irrigation regime × foliar spray of PGRs revealed that the erucic acid content was different for PGRs foliar spray treatments under irrigation regimes in both years (Table 7). Averaged by irrigation regimes and foliar spray of PGRs, the erucic acid content was 0.057 and 0.029% in the first and second years, respectively (Table 7). Averaged across years and foliar spray of PGRs, the lowest erucic acid content was recorded in the optimal irrigation regime, averaging 0.0408%, and decreases of 11.02 and 10.78% were observed under restricted irrigation from flowering and silique setting, respectively (Table 7). Averaged across years, spraying Me30 produced the minimum erucic acid content, averaging 0.036, 0.038, and 0.040% under full irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting, respectively (Table 7).

Glucosinolate

The data presented in Table 4 show that glucosinolate content was significantly affected by the two-way interaction of year × irrigation regime; the highest glucosinolate content belonged to the restricted irrigation regime from flowering stage, averaging 9.14 and 6.45 μmol g^{-1} in the first and second years, respectively (Table 4). When compared to the full irrigation regime, the restricted irrigation from flowering and silique-setting regimes boosted glucosinolate content; by contrast, foliar spray of PGRs significantly decreased glucosinolate content as compared to the control treatment. The lowest glucosinolate content was achieved from the Me30, Sa300, and Sa200 foliar spray treatments under full irrigation, restricted irrigation from flowering, and restricted irrigation from silique setting, with averages of 3.07, 6.97, 4.89 μmol g^{-1}, respectively (Table 5).

Discussion

The lower rapeseed seed yield in the first year than the second year could be attributed climatic parameters such as rainfall and minimum and maximum temperatures. In other words, different climatic conditions during the flowering and seed-filling periods in the two years of the experiment (Fig. 1) and their impacts on yield components led to a significant difference in seed yield between the two study years. Crop seed yield is a function of yield components, which are influenced by environmental factors (i.e., temperature and rainfall) as well as agronomic practices (i.e., irrigation, sowing date, plant nutrition, etc.). Drought stress caused by restricted irrigation leads to an increase in the ratio of respiration to photosynthesis (Aliyari Rad et al. 2021) and drought stress in the reproductive stages such as flowering and silique setting leads to falling flowers and siliques due mainly to the lower supply of photosynthetic materials. Obviously, the harmful effects of drought stress on growth, yield components, and grain yield depend on the intensity and time at which the stress occurs (Eyni-Nargeseh et al. 2022; Mokhtassi-Bidgoli et al. 2022). Applying PGRs could mitigate adverse effects of environmental stresses and improve crop growth and yield (Ijaz et al. 2019; Ganj-Abadi et al. 2021). Increased growth caused by applying ascorbic acid under stress and non-stress conditions is due to increased cell division and elongation (Athar et al. 2008), and considering this, it seems that the positive effect of ascorbic acid on growth led to an increase in the number of siliques (Kamal et al. 2017). The current findings specified that increasing the salicylic acid concentration under the optimal irrigation regime resulted in an increase in the number of siliques per plant, while a low concentration of salicylic acid was more effective under restricted irrigation from the silique stage (Table 5). Exogenous application of salicylic acid improves growth, physiological traits, yield components, and seed yield (Aftab et al. 2011; Fariduddin et al. 2003). In some studies, it was reported that applying methanol caused an increase in rapeseed seed yield by increasing the number of siliques (Zbiec et al. 2003). Methanol increases the number of siliques by influencing different metabolic pathways such as growth and development and activation of genes involved in jasmonic acid biosynthesis (Gout et al. 2000), and improvement of remobilization leads to an increase in seed weight (Paknejad et al. 2009).

Water supply is important during flowering and early growth of siliques, and drought stress during these stages affects carbohydrate reserves and seed development, and causes seed abortion (Mohammad et al. 2007). Occurrence of drought stress in the reproductive stage of rapeseed decreases seed yield and yield components (Jamshidi Zinab et al. 2022). Basically, the reduction in the number of seeds per silique can be attributed to the negative effect of water-limited conditions on pollination and the fertility of flowers. Nevertheless, application of foliar spray of PGRs such as salicylic acid, ascorbic acid, and methanol under water-limited irrigation positively affects crop growth and yield.

Increasing seed weight can be due to enhanced photosynthesis and distribution of assimilates entering seeds along with a reduction in drought stress because of ascorbic acid. Dolatabadian et al. (2010) declared that foliar application of ascorbic acid (150 mg l^{-1}) decreased the adverse effects of drought stress and improved growth and seed yield under optimal and drought-stress conditions. Furthermore, appli-

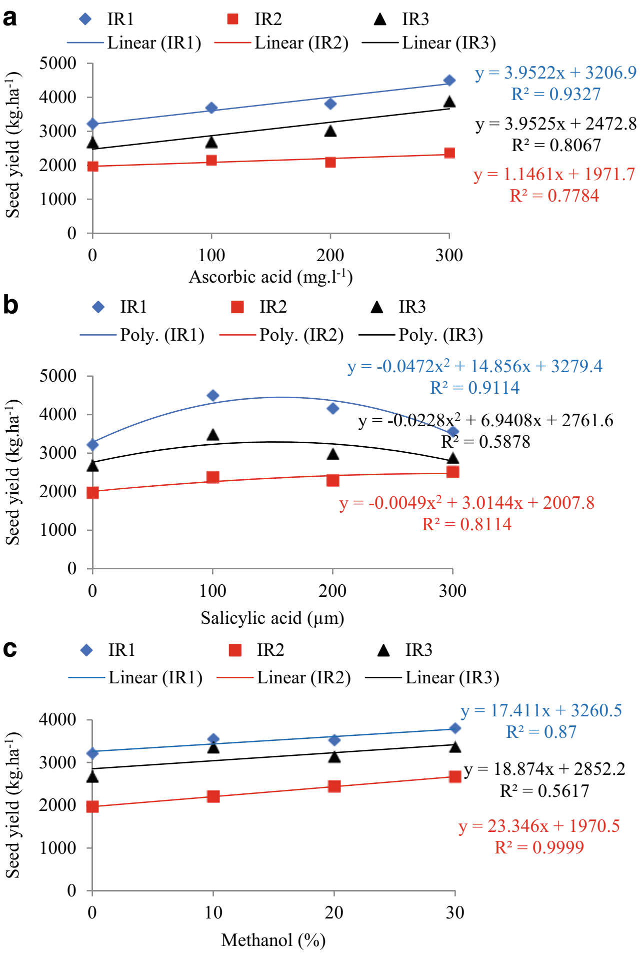

Fig. 3 Regression analysis of the response of rapeseed seed yield to foliar application of plant growth regulators (**a** ascorbic acid, **b** salicylic acid, and **c** methanol) at different concentrations under three irrigation regimes (*IR1* optimal water supply, *IR2* restricted irrigation from flowering, and *IR3* restricted irrigation from silique setting) in two study years (2015–2016 and 2016–2017)

cation of salicylic acid had a positive effect on thousand-seed weight under different irrigation regimes and this beneficial influence may be due to more remobilization to seeds during the seed-filling period. On the other hand, closure of stomata and reduced transpiration are induced by salicylic acid under drought-stress conditions (Zhang et al. 2020). The reaction of thousand-seed weight to methanol was positive under restricted irrigation from flowering and silique stages, and this result is in agreement with other research (Mirakhori et al. 2009; Rehman 2018).

Foliar spraying of AsA300 led to increased seed yield under the well-watered irrigation regime and restricted irrigation from the silique stage by 40 and 45%, respectively. It seems that reducing stress intensity and an increased rate of application of ascorbic acid can prevent a further decrease in seed yield (Fig. 3a). Ascorbic acid would increase crop seed yield through a positive effect on respiration activities, cell division, growth, and enzyme activities (Smirnoff 2011). Application of salicylic acid at low concentration (Sa100 treatment) was more effective in terms of seed yield compared to a high concentration (Sa200 and Sa300 treatments) under optimal irrigation and restricted irrigation from the silique stage, while rapeseed plants reacted positively to a high concentration of salicylic acid in restricted irrigation from flowering stage and showed an increasing trend upon increasing the salicylic acid concentration from 100 to 300 µm (Fig. 3b). Although the use of salicylic acid in high concentrations can have an inhibitory effect on plant growth (Fariduddin et al. 2003), an appropriate concentration boosts the seed yield by improving growth and physiological processes and remobilization of photosynthetic materials (Grown 2012). The regression analysis results showed that increasing the concentration of methanol from 10 to 30% linearly increased the rapeseed seed yield and this increase was higher with restricted irrigation from flowering than in the other two irrigation regimes (Fig. 3c). The increase in seed yield caused by application of methanol can be attributed to its positive role in enhancing chlorophyll concentration and plant photosynthesis. Moreover, increasing sucrose production improves plant turgor pressure and therefore reduces the sensitivity of methanol-treated plants to water-limited stress (Zbiec et al. 2003).

Oil content in oilseed crops is a key trait that is influenced by environmental and management factors (Kalantarahmadi and Daneshian 2023). High oil accumulation in seeds is related to expression of genes involved in fatty acid biosynthesis (Liu et al. 2014). Drought stress at flowering and silique-setting stages negatively affects oil content due to reducing seed capacity for absorbing assimilates and converting them into oil. Overall, the results of this experiment are in line with previous studies based on reduction of oil content due to a decrease in the photosynthesis assimilation rate and remobilization (Elferjani and Soolanayakanahally 2018; Safavifard et al. 2018). Foliar spray of Sa300 led to an increase in oil content by 7.3% compared to distilled water. As previously discussed, PGRs improve various parameters of plant growth under environmental stress and non-stress conditions and ultimately positively affect crop quantity and quality. Regression analysis shows that the application of all three PGRs increased rapeseed oil content with a linear trend under the three irrigation regimes (Fig. 4a, b, c).

Foliar spray of salicylic acid increases chlorophyll content, leaf area (Ghassemi-Golezani et al. 2018), and raises transportation of photosynthetic materials (Grown 2012), leading to higher seed yield and oil content (Mabudi Bilasvar et al. 2022; Ullah et al. 2012). Salicylic acid also enhances the accumulation of oil by increasing the proteins involved in the transcription of synthetic oil genes (Wu et al. 2014). The current findings clearly indicate an increase in

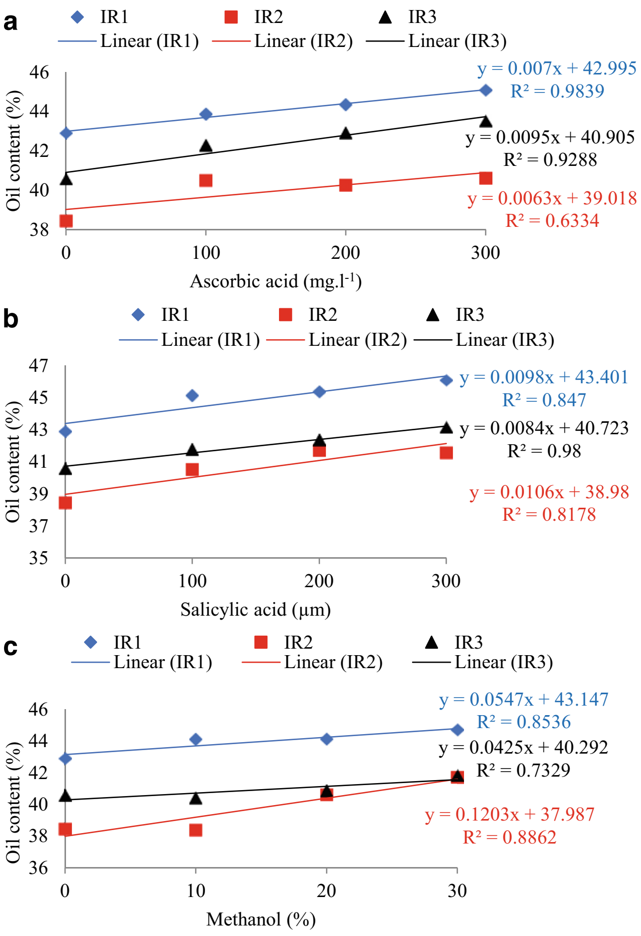

Fig. 4 Regression analysis of the response of rapeseed oil content to foliar application of plant growth regulators (**a** ascorbic acid, **b** salicylic acid, and **c** methanol) at different concentrations under three irrigation regimes (*IR1* optimal water supply, *IR2* restricted irrigation from flowering, and *IR3* restricted irrigation from silique setting) in two study years (2015–2016 and 2016–2017)

oil content following the use of methanol (30%), ascorbic acid (300 mg l^{-1}), and salicylic acid (200 µm). Also, by using PGRs, it was observed that the harmful effects of drought stress were somewhat neutralized, particularly by methanol at 30% (Fig. 4). In this regard, Pastori et al. (2003) declared that ascorbic acid is an important soluble antioxidant and acts as a coenzyme in reactions by which carbohydrates, fats, and proteins are metabolized. Kalantar Ahmadi et al. (2016) showed that foliar spray of methanol, salicylic acid, and ascorbic acid improved the oil content of canola under drought-stress conditions. Methanol increases turgor pressure, photosynthesis, sugar content (Zbiec et al. 2003), and oil content (Yazdi Far et al. 2015).

Oil composition is determined by genetic and environmental conditions (Shirani Rad et al. 2021) and agronomic operations such as drought stress (Mokhtassi-Bidgoli et al. 2022; Feizabadi et al. 2021; Shiranirad et al. 2023), fertilization (Amiri-Darban et al. 2020), and application of PGRs (Ijaz et al. 2019; Estaji and Niknam 2020). Water-deficit stress would cause a shortening of the lipid accumulation phase, some detriments to enzymatic activities including oleate desaturase (Ebrahimian and Bybordi 2012), and decrease unsaturated acids (Flagella et al. 2004). In a study by Safavifard et al. (2018), oleic acid content was reported to be decreased when irrigation was restricted from flowering and silique-setting stages. Similarly, Ebrahimian and Bybordi et al. (2012) declared that oleic acid and linoleic acid contents were reduced when sunflower plants were subjected to drought stress.

PGRs such as salicylic acid (Ullah et al. 2012) and ascorbic acid (Ali-Shahhat et al. 2014) can improve the oil quality of rapeseed under drought-stress conditions. Foliar spray of ascorbic acid, salicylic acid, and methanol significantly reduces lipid peroxidation and malondialdehyde of rapeseed under drought-stress conditions (Kalantar Ahmadi et al. 2015a). The enhancement in linolenic acid with application of ascorbic acid might be related to acceleration of the biosynthesis pathway of linolenic acid (Ali-Shahhat et al. 2014). It seems that the antioxidant properties of ascorbic acid reduce the negative effects of drought stress and improve the conditions necessary to improve the accumulation of linoleic acid. The role of salicylic acid in alteration of unsaturated fatty acids can be mentioned as it increases the activity of oleoyl-phosphatidylcholine D12 desaturase. The current findings are in agreement with Ganj-Abadi et al. (2021), who reported that use of salicylic acid led to increased palmitic acid, while Noreen and Ashraf (2010) declared that palmitic and stearic acids were not affected by salicylic acid. The present findings are in line with Ali-Shahhat et al. (2014) regarding increased stearic acid as a result of using ascorbic acid. Emam et al. (2011) declared that ascorbic acid caused a decrease in palmitic and stearic acids. Increasing linolenic acid content under drought-stress conditions and application of salicylic acid is supported by a report from Mabudi Bilasvar et al. (2022).

The contents of erucic acid and glucosinolate are considered two important traits to evaluate the quality of rapeseed oil and cake press. In this research, although drought stress resulted in a slight increase in the amount of these two traits and foliar spraying of PGRs reduced their contents, the contents of both traits were within a safe range. Eyni-Nargeseh et al. (2022) concluded that drought stress increases the amount of erucic acid and glucosinolate for rapeseed genotypes, while Ganj-Abadi et al. (2021) reported a decrease in these traits under salicylic acid application.

Conclusion

The results showed that seed yield and fatty acid composition reacted differently to irrigation regimes and foliar spray of PGRs. Foliar spray of salicylic acid (300 µm) in the optimal irrigation regime and restricted irrigation from flowering as well as ascorbic acid (300 mg l^{-1}) in restricted irrigation from the silique setting stage were more effective in increasing the oleic acid content. Foliar spray of salicylic acid (300 µm) in the optimal irrigation regime, and ascorbic acid (300 mg l^{-1}) and salicylic acid (300 µm) in restricted irrigation from the flowering stage were more effective for increasing linoleic acid content. Overall, the increase of unsaturated fatty acids in reaction to growth regulators improves the nutritional value of oil. Further experiments with a combination of growth regulators under different irrigation regimes are required to investigate the response of rapeseed in terms of seed yield and oil quality.

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Conflict of interest S.A. Kalantar Ahmadi, and H. Eyni-Nargeseh declare that they have no competing interests.

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Seyed Ahmad Kalantar Ahmadi works at Safiabad Agricultural and Natural Resources Research and Education Center. His research interest is crop ecophysiology, especially the effects of abiotic environmental stresses on development and growth of canola and soybean