

Canola quality and fatty acids composition as influenced by delayed cropping and late season drought stress

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ABSTRACT - To investigate the effects of drought stress and delayed planting on oil quality and fatty acids composition of canola cultivars, a factorial split-plot test was conducted in a randomized complete blocks design with three replications at Seed and Plant Improvement Institute, Karaj, Iran for two years (2015-2017). Planting date at two levels of timely planting (Oct. 27) and late planting (Nov. 27) and irrigation at two levels of normal irrigation (control) and restricted irrigation from pod formation stage on wards were considered the main factors. Canola cultivars including Kodiak, Traviata, Compass, Diffusion, Mehr2, Elvise and Tassilo were the variables. The simple effects of planting date, irrigation, and cultivar on all research parameters were significant. The interaction effect of planting date \times irrigation was significant on oil yield and oil content at the level of 5% and in terms of linoleic and palmitic acids, and stomatal resistance; it was significant at the level of 1%. The interaction effect of irrigation \times cultivar was significant on palmitic acid, glucosinolate and stomatal resistance at the level of 1%. Late season drought stress and late planting dramatically reduced oil yield (58%), oil content (8%) and qualitative characteristics of canola cultivars. Among the studied cultivars, with considering all conditions of this research, the Tassilo cultivar having the seed yield (4407 kg ha⁻¹), oil yield (2007 kg ha⁻¹), chlorophyll content (1.43 mg g⁻¹fw), high oleic and linoleic fatty acids, as well as low and standard erucic acid (0.41%) and glucosinolate (22.8 μ mol g⁻¹). was recommended as a favorable cultivar for planting in temperate cold regions with arid and semi-arid climates.

INTRODUCTION

Canola, along with other species of *Brassica* after soybeans, has been assigned the second position of producing oil between oily seeds. Also, the low concentration of canola oil-saturated fatty acids compared to other vegetable oils caused the use of canola oil as an useful edible oil (Scarath and Tang, 2006). Canola production is an important alternative for agricultural policy-makers in Iran to reduce dependency on the imported vegetable oils. Nevertheless, the canola planted area is only increasing at a slow pace, indicating a low willingness to accept of farmers (Zarafshani et al., 2017).

Drought is one of the most important factors limiting plant growth and agricultural crop production throughout the world (Sun et al., 2013). Drought stress can affect the reproductive mechanisms of canola seed yield such as flowering and pod formation, number of seeds per pod and seed filling.

However, the intensity of these effects has been reported to be the functions of cultivar, stress duration,

climate and growth stages (Sinaki et al., 2007; Farooq et al., 2008). It has been also reported that the level of saturated fatty acids of the canola seed oil decreases in drought stress conditions which can be correlated to the reduction in the growth stage (Shekari et al., 2015). The combination of canola seed oil fatty acids includes 7% saturated fatty acids, 66% unsaturated single fatty acids and 27% unsaturated multiple fatty acids and canola cultivars are significantly different in terms of the combination of seed oil fatty acids (Kadivar et al., 2010). The quality of canola seed oil is mainly determined by the oleic, linoleic and erucic fatty acids and is greatly influenced by environmental conditions (Enjalbert et al., 2013), cultivar type (Nasr et al., 2006, Javidfar et al., 2007) and the phenological stages (Pritchard et al., 2000). Safe limits for erucic acid and glucosinolate compounds have been described as less than 2% of erucic acid in oil and less than 30 μ mol g⁻¹ of glucosinolate in oil-free meals (Grombacher and Nelson, 1992).

Planting date is a crucial factor influencing seed yield, seed oil content and the combination of fatty acids (Shirani Rad et al., 2017). Appropriate cultural practices can reduce crop vulnerability to late-season drought stress, and also can contribute to increase seed yield (Bashir, 2010). Robertson and Holland (2004) also reported that the delay in the fall cultivation of canola led to a reduction in the flowering period due to the increase in the environmental temperature. Consequently, the late-season heat encounter with the seed filling period led to the reduction of seed yield and canola oil yield.

Canola cultivars that sustain higher-yielding ability during periods of late-season drought should have an important effect on the development of canola cultivation, especially in regions with late spring harvesting. Canola cultivation may be expanded into cold and temperate cold climates by selecting the appropriate cultivar and cultivation time in each region (Ul-Hassan et al., 2005).

Although rainfall is usually sufficient in most temperate cold regions in Iran with arid and semi-arid climates during March and April, water management is crucial, especially during flowering, pod formation and seed-filling. Under controlled conditions at these stages, the number of irrigation can be reduced and the amount of water for irrigation can be saved between 1280 to 1920 m³ ha⁻¹ (Safavi Fard et al., 2018). Furthermore, for sustainable development of cultivation of canola, in addition to increasing production per area unit, it is essential to consider the main factor of the cultivation time limit to be able to get rid of the restriction of a fall cultivation time limit of 15-20-day for canola in cold temperate regions by providing appropriate strategies such as selection of cultivars with an optimal reaction in the new status of winter cultivation (Safavi Fard et al., 2018). Therefore, the present study aimed to examine the late cultivation and the effect of late cultivation and lack of water on the qualitative and quantitative properties of selected canola cultivars.

MATERIALS AND METHODS

Experimental Design

A factorial split-plot experiment was conducted based on the randomized complete block design (RCBD) with three replications in the cultivation years of 2015-2017 at Seed and Plant Improvement Institute, Karaj, Iran, (35° 59'N, 50° 75'E, and altitude of 1313 m) to investigate the effect of different sowing times on quantitative and qualitative characteristics of canola cultivars in late-season drought stress conditions. The average annual rainfall (for 25 years) has been reported to be 251 mm which was concentrated in the late fall and early spring (Iran Meteorological Organization, 2015). Fig. 1, shows the monthly precipitation rate of the experiment site during the two crop years.

In the present study, planting date at two levels of timely planting (Oct. 27) and late planting (Nov.

27) and irrigation at two levels of normal irrigation (control) and restricted irrigation from the pod formation stage onwards were considered the main factors. Canola cultivars including Kodiak, Traviata, Compass, Diffusion, Mehr2, Elvise and Tassilo were considered the variables (Table 1).

Experimental Procedure

Each experimental plot included six 6-meter lines with 30 cm distance. The distance between the plants along the line was 5 cm and 2 lateral lines were considered as margins. Fertilizers' applications based on the soil test were: 1) 150 kg ha⁻¹ ammonium phosphate and 150 kg ha⁻¹ potassium sulfate as a basis with preparing seedbed simultaneously, 2) 350 kg ha⁻¹ urea (100 kg at three-leaf stage, 150 kg at stem elongation stage and 100 kg at bud formation stage). All operations related to the harvesting except irrigation were carried out uniformly and according to the traditions of the area. Irrigation intervals were considered based on 80 mm evaporation from class A evaporation pan and 80% of consumed water at each irrigation was evaporated. The amount of water entering the farm was measured by the water meter (Safavi Fard et al., 2018). The number of irrigation frequencies in fall cultivation in control treatments, restricted irrigation from pod formation and flowering stages was 8, 6 and 5 times, respectively, and in winter cultivation it was 6, 4 and 3 times, respectively. Moreover, the amount of water consumed in these treatments was 5120 and 3840 m³ ha⁻¹, respectively.

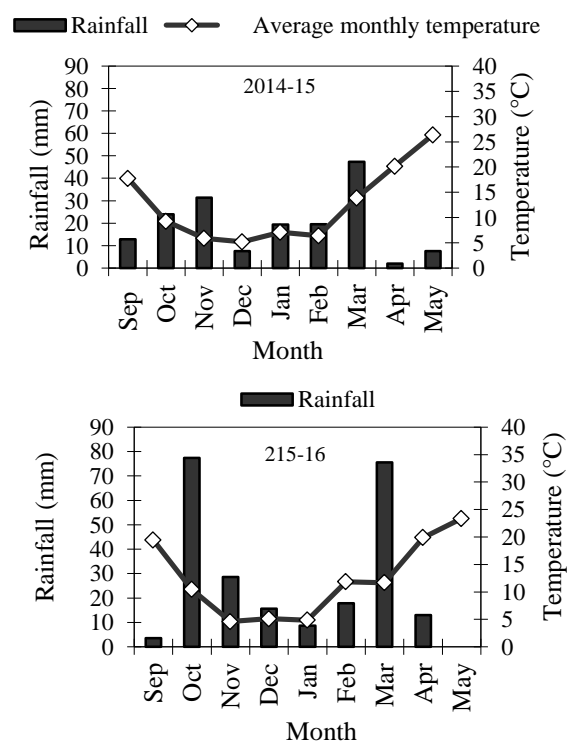


Fig. 1. Variation of temperature and rainfall in Karaj meteorology station during 2014-2016 growing seasons

Table 1. Specifications of the studied cultivars

cultivar	Origin	Type	Growth type
Kodiak	Germany	Hybrid	Winter
Traviata	Germany	Hybrid	Winter
Compass	Germany	Hybrid	Winter
Diffusion	Germany	Hybrid	Winter
Mehr2	Iran	OP	Winter
Elvise	France	Hybrid	Winter
Tassilo	Germany	Hybrid	Winter

Measurement of Yield and Oil Content and Determination of Fatty Acid Composition

To determine the seed yield, the available shrubs were technically cut separately at an area of 4.8 m² of each plot test and were weighted and calculated using accurate scales. Stomatal resistance was measured using an AP₄ porometer (Delta-T Devices Ltd., Cambridge, UK) using an average of five leaves from each treatment. The youngest fully expanded leaves (third from the apex) were harvested 20 days after drought stress treatment (Shahsavari et al., 2014a). To determine the seed oil percentage, a 5 g sample was selected from each plot and its oil percentage was determined by NMR (Nuclear Magnetic Resonance, German Broker Brand minispec mq20 Model based on the International Standard ISO 5511., 1992). Having determined the seed oil percentage, the seed oil yield was calculated in kg ha⁻¹ by multiplying it by seed yield. The gas chromatography (GC) method was used to measure and determine the fatty acids in seed oils. Oil samples were extracted in triplicate from canola seed (100 g) according to the method described by Azadmard-Damirchi et al., (2005).

Fatty acid methyl esters (FAMES) were prepared from the oil samples according to the method reported by Savage et al. (1997). Briefly, 2 ml of 0.01 M NaOH in methanol was added to a tube containing the oil sample (ca. 10 mg) dissolved in 0.5 ml hexane and then held in a water bath at 60° C for 10 min. Thereafter, boron trifluoride in methanol (20% of BF₃ in methanol) was added and the samples were held an additional 10 min in a water bath at 60 °C. The sample was cooled under running water and 2 ml of 20% (w v⁻¹) of sodium chloride and 1 ml hexane was added. After mixing completely, the hexane layer that contained the FAMES was separated by centrifugation at 3000 rpm for 5 min.

The FAMES were analyzed by GC according to the method described by Azadmard-Damirchi and Dutta (2006). The GC instrument was equipped with a flame ionization detector and a split/splitless injector. A 50 m 9 0.22 mm, 0.25 lm film thickness fused-silica capillary column BPX70 (SGE, Austin, TX, USA) was used for analysis. Injector and detector temperatures were 230 and 250 °C, respectively. Oven conditions were 158 °C increased to 220 °C at a rate of 2 °C min⁻¹ and maintained for 5 min. Helium was used as the carrier gas and nitrogen as the make-up gas at a flow rate of 30 ml min⁻¹. The FAMES were identified by comparison of their retention times with standard FAMES and the peak areas reported as a percentage of

the total fatty acids.

Glucosinolate was analyzed using the FOSS Routine Near Measurement System (35RP- 3752F, TR-3657-C, Model 6500 (Maryland, USA).

The chlorophyll content of the tested leave was determined by a method described by Arnon (1967). The chlorophyll a, b and total contents in mg g⁻¹ of fresh leaf weight were determined for each treatment through using equations (1), (2) and (3), respectively.

$$\text{Chlorophyll a} = (19.3 * A_{663} - 0.86 * A_{645}) V/100W \quad (1)$$

$$\text{Chlorophyll b} = (19.3 * A_{645} - 3.6 * A_{663}) V/100 \quad (2)$$

$$\text{Total chlorophyll} = \text{Chl a} + \text{Chl b} \quad (3)$$

V = The volume of filtered solution (upper solution obtained from centrifugation)

A=The light absorbance at wavelengths of 663, 645, and 470 nm

W = The sample's fresh weight in grams

Statistical Analysis

SAS statistical software version 1.9 (Cary, NC, USA) was used to conduct the combined variance analysis after Bartlett's test and to determine the insignificant result of the test. A comparison of the means was made by LSD (Least Significant Difference, 2008) Test.

RESULTS AND DISCUSSION

The effect of year on all research parameters (except erucic acid) and the effect of planting date and irrigation on all research parameters were significant at the level of 1%. The cultivars were significantly different in terms of all research parameters. The effect of planting date × irrigation was significant on oil yield and oil content at the level of 5% and in terms of linoleic and palmitic acids, and stomatal resistance, it was significant at the level of 1%. The interaction effect of irrigation × The interaction effect of irrigation × cultivar was significant on palmitic acid, glucosinolate and stomatal resistance at the level of 1%.

Seed Yield

The mean comparisons of the simple effect of cultivar on canola characteristics showed that the tested cultivars in this study are placed in statistically different groups in terms of seed yield (Table 2). The Tassilo cultivar with a mean of 4407 kg ha⁻¹ and Compass cultivar with a mean of 3711 kg ha⁻¹ had the most and the least seed yields, respectively.

The adverse impact of late planting on seed yield was determined with the 30% reduction in its amount compared to the normal planting (Table 3).

Late planting of canola has been reported to cause the ripening stage to encounter high temperature and increase pod respiration (Gan et al., 2004; Rafiei et al., 2011). The late fall planting of canola seems to lead the plant to start winter with weak rosette which leads to the plants being damaged due to cold weather.

Therefore, after the winter when the weather gets warm, the plants cannot use the environmental conditions appropriately for photosynthesis and producing enough phloem sap. Also, As a result of delayed cultivation, the seed filling takes place when the temperature is high and the high temperature prevents seed filling and the amount of metabolic substances reduces as the respiration is resonated (Ozer, 2003).

The average seed yield was different at different levels of irrigation, such that the average of this property in normal irrigation treatment (control)

was 4910 kg ha⁻¹ and in the restricted irrigation treatment, from the pod formation stage onwards, the seed yield was reduced to 59% (Table 4). Shahsavari et al. (2014b) reported that restricted irrigation from the pod formation stage onwards reduced canola seed yield by 195% compared to the normal irrigation. It seems that the reduction of photosynthetic materials under stress conditions and failure to sufficiently supply them for the pods and consequently their loss will ultimately result in reduced yields.

Table 2. Mean comparison of simple effect of cultivar on canola characteristics

	Tassilo	Elvise	Mehr2	Diffusion	Compass	Traviata	Kodiak
SY (kg ha ⁻¹)	3848b-d	3819b-d	3711cd	4064a-c	3515d	4145ab	4407a
OY (kg ha ⁻¹)	1733b-d	1708b-d	1657 cd	1833a-c	1564d	1876ab	2007a
OC (%)	44.61bc	44.43cd	44.34cd	44.57b	44.15d	44.87ab	45.09a
OLA (%)	65.44cd	65.44cd	65.34d	65.58bc	65.15e	65.67ab	65.74a
LINO (%)	16.44b-d	16.40b-d	16.25c-d	16.69bc	16.01d	16.80ab	17.23a
LINC (%)	6.06ab	6.03ab	6.11ab	5.91bc	6.32a	5.85bc	5.68c
PAL (%)	4.96c	4.94cd	4.90d	5.01b	4.83e	5.06b	5.14a
ERU (%)	0.32bc	0.32bc	0.33ab	0.30cd	0.35a	0.29de	0.27e
GLU (μmol g ⁻¹ dw)	18.99c	19.14bc	19.59b	18.21d	20.51a	17.92d	17.17e
CHL (mg g ⁻¹ fw)	1.33c	1.31cd	1.27de	1.37bc	1.22e	1.39ab	1.43a
RS (s cm ⁻¹)	20.15c	20.56c	21.25b	19.20d	22.41a	18.66e	17.36f

Any two means sharing a common letter do not differ significantly from each other at 5% probability

SY: Seed yield, OY: Oil yield, OC: Oil content, OLA: Oleic acid, LINC: Linolenic acid, LINO: Linoleic acid, PAL: palmitic acid, ERU: Erucic acid, GLU: Glucosinolate, CHL: Chlorophyll, RS: Stomatal resistance

Table 3. Mean comparison of simple effect of planting date on canola characteristics

Planting date	Seed yield (kg ha ⁻¹)	Oil yield (kg ha ⁻¹)	Oil content (%)	Oleic acid (%)	Linoleic acid (%)	Linolenic acid (%)	Palmitic acid (%)	Erucic acid (%)	Glucosinolate (μmol g ⁻¹ dw)	Chlorophyll (mg g ⁻¹ fw)	Stomatal resistance (s cm ⁻¹)
Oct.27	4445a	2022.5a	45.1a	65.8a	17.2a	5.6b	5.2a	0.26b	16.8b	1.45a	17.1b
Nov.27	3415b	1514.7b	44.1b	65.1b	15.9b	6.4a	4.8b	0.36a	20.8a	1.2b	22.8a

Any two means sharing a common letter do not differ significantly from each other at 5% probability

Table 4. Mean comparison of simple effect of irrigation on canola characteristics

Irrigation	Seed yield (kg ha ⁻¹)	Oil yield (kg ha ⁻¹)	Oil content (%)	Oleic acid (%)	Linoleic acid (%)	Linolenic acid (%)	Palmitic acid (%)	Erucic acid (%)	Glucosinolate (μmol g ⁻¹ dw)	Chlorophyll (mg g ⁻¹ fw)	Stomatal resistance (s cm ⁻¹)
Normal irrigation (control)	4910a	2254.4a	45.8a	66.1a	17.9a	5.2b	5.3a	0.21b	14.8b	1.6a	13.8b
Restricted irrigation (after pod formation stage)	2910b	1282.9b	43.4b	64.8b	15.2b	6.8a	4.6b	0.41a	22.8a	1b	26.1a

Any two means sharing a common letter do not differ significantly from each other at 5% probability

Oil Yield

The mean comparison of the simple effect of cultivar on canola characteristics showed that the oil yield of the tested cultivars in this research were significantly different (Table 2). The Tassilo cultivar had the highest seed oil yield (2007 kg ha⁻¹). The comparison of the interaction effect of means of planting date × irrigation showed that the highest oil yield was related to the timely planting date (Oct. 27) which had a mean of 2566 kg ha⁻¹ in normal irrigation conditions. While the least oil yield was related to the late planting (Nov.27) which had a mean of 1078 kg ha⁻¹ in restricted irrigation treatment from the pod formation stage onwards (Table 5). Safavi Fard et al. (2018) and Miri and Bagheri (2013) also reported a reduction in oil yield due to late planting. In the study conducted by Soleymani et al. (2011), drought stress reduced the oil yield from 2138 kg ha⁻¹ in the control group to 877, 1141 and 1265 kg ha⁻¹ in restricted irrigation after stem elongation stage, flowering stage and pod formation stage in canola, respectively. It was observed in a study that seed yield and yield increase by an increase in the moisture after the pollination stage (Sinaki et al., 2007).

Oil Content

The average seed oil percentage was significantly different between the timely planting date and the late planting date (Table 3). The late planting date caused a 2.3% reduction in seed oil content compared to the normal planting date. It seems that there is limited opportunity to increase the percentage of seed oil in late planting because of the shorter duration time of growth. The average percentage of seed oil was different at different levels of irrigation (Table 4). Restricted irrigation at the pod formation stage reduced the percentage of oil by 5.2 % compared to the normal irrigation. The mean comparisons of the simple effect of cultivar on canola characteristics showed that oil content of the tested cultivars in this research were significantly different (Table 2). Tassilo cultivar had the highest seed oil content (45.09%). In general, it has been reported that genetic factors are the main parameters determining the percentage of canola seed oil, whereas the effect of environmental factors on seed oil content is less (Robertson and Holland, 2004). Sinaki

et al. (2007) reported that drought stress at the end of the growing season reduced the percentage of seed oil.

The different in seed oil content of the tested cultivars in this study was statistically significant (Table 3). The Tassilo cultivar had the highest percentage of seed oil (45.09%). Mean comparison of the interaction effect of planting date and irrigation showed that the highest oil content was obtained on the timely planting date (Oct. 27) and under normal irrigation conditions with an average of 46.51%. While the lowest oil content was due to the late planting date (Nov. 27) and restricted irrigation from the pod formation stage with an average of 43.01% (Table 5). The cause of oil loss in late planting dates can be related to the temperature changes in the seed filling stage and the reduction of photosynthesis. In this case, a lower percentage of the substances is created and carbohydrates are converted into oil. On favorable sowing date, with the high seed yield and oil content, canola produces the highest seed oil yield, while delay in sowing date reduces seed yield, seed oil content and, finally, the seed oil yield due to reduced plant growth, contact of the seed-filling stage with high temperature, increased respiration and reduced photosynthetic materials

Oleic Acid

The average percentage of oleic acid was significantly different between timely planting date and late planting date (Table 3). The late planting reduced the amount of oleic acid by 1.1% compared to the normal planting date. The average percentage of oleic acid was different at different levels of irrigation (Table 4). Restricted irrigation at the pod formation stage reduced the percentage of oleic acid by 2 % compared to the normal irrigation.

Due to this fact that oleic acid forms the highest amount of canola seed oil fatty acids and has a significant role in increasing the quality of canola seed oil, The Tassilo cultivar with 65.74% contained the highest oleic acid (ω9) (Table 2). Flagella et al. (2002) emphasized the effect of different environmental conditions on the content of oleic acid in different regions. It has been shown that the quality of seed oil is determined mainly by the amount of oleic and linoleic fatty acids (Ul-Hassan et al., 2005).

Table 5. Interaction effects of planting date and irrigation on characteristics of canola

Mean	Treatment			
	Normal irrigation		Restricted irrigation after pod formation stage	
	Oct. 27	Nov. 27	Oct. 27	Nov. 27
OY (Kg ha ⁻¹)	2566a	1942b	1479c	1078d
OC (%)	46.51a	45.11b	43.79c	43.01d
LINO (%)	18.61a	17.20b	15.73c	14.63d
PAL (%)	5.62a	5.06b	4.75bc	4.47c
RS (s cm ⁻¹)	10.13d	17.39c	24.03b	28.22a

OY: Oil Yield, OC: Oil content, LINO: Linoleic acid, PAL: palmitic acid, RS: Stomatal resistance

Linoleic Acid

The different linoleic acid percentages of the genotypes tested in this study were statistically significant (Table 2). The Tassilo cultivar with an average of 17.23% had the highest percentage of linoleic acid ($\omega 6$). The reaction of the cultivars to the irrigation treatments was different in terms of linoleic acid content in both the timely planting date and the late planting date (Table 4).

The highest percentage of linoleic acid was obtained on the timely planting date (Oct. 27) and in the normal irrigation conditions with an average of 18.61%. While the lowest percentage of linoleic acid was related to the late planting date (Nov. 27) and irrigation was restricted from the pod formation stage with an average of 14.63% (Table 5). Shahsavari et al. (2014c) reported a significant decrease in the amount of linoleic acid under stress conditions in the study of the reaction of canola seed oil fatty acids to late-season drought stress.

Linolenic Acid

Significant differences in the average percentage of linolenic acid in both the timely planting (Oct. 27) and the late planting date (Nov. 27) showed that the late planting resulted in an increase of 14.2% linolenic acid compared to the timely planting date (Table 3). The average percentage of linolenic acid ($\omega 3$) was significantly different at different levels of irrigation, so that drought stress caused a 30% increase in linolenic acid percentages compared to that at normal irrigation conditions. The average percentages of this fatty acid in normal irrigation (control) and restricted irrigation at the pod formation stage was 5.2% and 6.8%, respectively (Table 4). An increase of 1.7 to 2% of unsaturated fatty acids, such as linolenic acid, with a 3.8% reduction of oleic acid in canola seed oil through drought stress was reported at the Mediterranean climate conditions (Aslam et al., 2009). Shahsavari et al. (2014c) also pointed to a 35% increase in linolenic acid percentage under drought stress conditions in the canola plant. Since linolenic acid is an essential fatty acid for the activity of plant photosynthesis and the development of canola seed pollen, it seems that increasing its percentage through late planting conditions is necessary to achieve maximum seed yield.

Palmitic Acid

The canola cultivars, tested in this study, had statistically significant differences in palmitic acid content (Table 2). The highest percentage of palmitic acid (5.14%) was seen in the Tassilo cultivar. Cultivars' reaction to the irrigation treatments was different in terms of palmitic acid content in both the timely planting date and the late planting date (Table 4). The highest percentage of palmitic acid was obtained on the timely planting (Oct. 27) and in normal irrigation conditions with an average of 5.62%. However, the lowest percentage of palmitic acid was related to the late planting (Nov. 27) and

restricted irrigation at the pod formation stage with an average of 14.63% (Table 5). Safavi Fard et al. (2018) reported a 12% decrease of palmitic acid in Delgan cultivar in the late planting date of canola. Garsid (2004) showed that the levels of palmitic and oleic fatty acids decreased due to the coincidence of oil synthesis with hot summer weather.

Erucic Acid

Significant differences in the average percentage of erucic acid in both the timely planting (Oct. 27) and the late planting (Nov. 27) showed that the late planting date resulted in an increase of 38% erucic acid compared to the timely planting date (Table 3). The average percentage of erucic acid in different levels of irrigation was significantly different, and drought stress caused a 95% increase in erucic acid content compared to that in normal irrigation conditions. The average percentage of this fatty acid in normal irrigation (control) and in restricted irrigation at the pod formation stage was 0.41% and 0.21%, respectively (Table 4). Shahsavari et al. (2014c) and Safavi Fard et al. (2018) also reported an increase in erucic acid percentage as a result of late season drought stress and the canola late planting date. Given that the amount of erucic acid is an important indicator for the edible consumption of canola oil (Gecgel et al., 2007), in this study, the Tassilo cultivar had the lowest average of erucic acid with 0.27% (Table 2). Also, in all watering conditions, the quantity of erucic acid was been within the standard range (less than 2%). According to European Union standards, it should be below two percent) Ullah et al., (2012) reported that water stress increased canola erucic acid and conversely reduced oleic and linolenic acids.

Glucosinolate

The canola cultivars tested in this study were statistically different in terms of having different levels of glucosinolate. The Tassilo cultivar with an average of 17.17 $\mu\text{mol g}^{-1}$ had the lowest glucosinolate content (Table 2). The reaction of cultivars to the irrigation treatments was different in terms of glucosinolate content. The lowest rate of glucosinolate was obtained under normal irrigation conditions with an average of 14.8 $\mu\text{mol g}^{-1}$ and the highest rate of glucosinolate was obtained in restricted irrigation at the pod formation stage with an average of 22.8 $\mu\text{mol g}^{-1}$ (Table 4). Glucosinolate levels in tested canola cultivar were significantly different between the normal planting date compared to the late planting date. The late planting date increased the glucosinolate content to 23.8% more compared to that in the normal planting date (Table 3).

Safe limits for erucic acid and glucosinolate compounds have been already described as less than 2% of erucic acid in oil and less than 30 $\mu\text{mol g}^{-1}$ of glucosinolate in oil-free meals (Grombacher and Nelson 1992). To describe the quality of canola, two factors of oil and protein content of seed are used, which are applied for human or industrial use depending on the composition of fatty acids, (Rathke et al., 2005). It has been reported that increasing glucosinolate reduces the quality and nutritional value

of canola press cake (Sulisbury et al., 1987), which is influenced by hereditary and environmental factors (Fieldsend et al., 1991).

Chlorophyll Content

The cultivars had statistically significant differences in terms of chlorophyll content. The Tassilo cultivar had the highest chlorophyll content with an average of 1.43 mg g^{-1} (Table 2). The average chlorophyll content was significantly different at different levels of irrigation so that the average chlorophyll content under restricted irrigation conditions at the pod formation stage reduced to 62% of the chlorophyll content under normal irrigation conditions (Table 4).

Significant differences in the average chlorophyll content in both the timely planting (Oct. 27) and the late planting (Nov. 27) showed that the late planting date caused a 17.2% reduction in chlorophyll content compared to the timely planting date (Table 3). These results showed that climatic conditions had great effects on the synthesis and accumulation of the different pigments, as an indicator for playing a role in the formation of the secondary products. In this matter, Abou-Dahab et al. (2014) found that early planting dates resulted in an increase in chlorophyll a and chlorophyll b contents in the leaves while delaying the planting dates decreased them.

Stomatal Resistance

The cultivars had statistically significant differences in stomatal resistance. The Tassilo cultivar with an average of 10.13 s cm^{-1} had the lowest stomatal resistance and Mehr2 cultivar with an average of 22.42 s cm^{-1} , had the highest stomatal resistance (Table 2). The reaction of the cultivars to the irrigation treatments was different in the timely planting date and the late planting date in term of stomatal resistance (Table 5). The least stomatal resistance was obtained on the timely planting (Oct. 27) and under normal irrigation conditions with an average of 10.13 s cm^{-1} . The highest stomatal resistance was observed in the late planting (Nov. 27) and under restricted irrigation at the pod formation stage with an average of 28.22 s cm^{-1} . Several investigators reported the negative effects of drought stress on stomatal

conductance (Kausar et al., 2006, Shahsavari et al., 2014a; Shahsavari and Dadrasnia, 2016). Similarly, Pasban Eslam (2011) reported that water stress during the pod formation stage significantly decreased relative water content, stomatal conductance and increased canopy temperature. A decrease in stomatal conductance indicates the inverse for stomatal resistance.

Canola grown under drought stress conditions exhibited lower stomatal conductance (higher stomatal resistance) to conserve water. Consequently, CO_2 fixation and the photosynthetic rate decreased, resulting in lower production of assimilates for growth and plant yield (Mafakheri et al., 2010).

CONCLUSIONS

Drought stress, especially in late-season, is one of the most important and limiting factors in the growth of canola, especially in temperate cold regions with arid and semi-arid climates. The appropriate planting time provides the necessary growth rate for canola plants and reduces their vulnerability to late season drought stress and leads to a significant increase in seed yield and oil quality. In this study, late season drought stress (restricted irrigation at the pod formation stage onwards) and late planting dramatically reduced seed yield, oil yield and qualitative characteristics of canola cultivars. Among the studied cultivars, The Tassilo cultivar is recommended for planting, as this cultivar showed maximum seed yield, oil yield, chlorophyll content, oleic and linoleic fatty acids, as well as low and standard erucic acid and glucosinolate under conditions applied in this study.

Author Contribution

Zahra Biyouk, carried out the experiment, collected the data; Nasser Shahsavari, Performed the statistical analysis and assisted in manuscript writing and other contribution.

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تغییرات کیفی و ترکیب اسیدهای چرب کلزا تحت تاثیر کاشت تاخیری و تنش خشکی آخر فصل

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کاشت

تنش رطوبتی

چکیده - به منظور بررسی اثر کاشت تاخیری و تنش خشکی بر کیفیت روغن و ترکیب اسیدهای چرب هیبریدهای کلزا، آزمایشی به صورت فاکتوریل اسپلیت پلات در قالب طرح بلوکهای کامل تصادفی با سه تکرار به مدت دو سال (۱۳۹۳-۱۳۹۵) درموسسه تهیه نهال و بذر کرج، ایران، اجرا شد. تاریخ کاشت در دو سطح شامل کاشت به موقع (۵ مهر ماه) و کاشت تاخیری (۵ آبان ماه)، آبیاری نیز در دو سطح شامل آبیاری معمول (شاهد) و قطع آبیاری از مرحله خورجیندهی به بعد به عنوان عوامل اصلی و ژنوتیپ های کلزا شامل Kodiak، Traviata، Compass، Diffusion، Elvise، Mehr2 و Tassilo به عنوان عوامل فرعی بودند. اثرات ساده تاریخ کاشت، آبیاری و رقم بر تمام صفات آزمایش معنی دار بود. اثر متقابل تاریخ کاشت×آبیاری بر صفات درصد و عملکرد روغن دانه در سطح ۵ درصد، و بر درصد اسیدهای لینولئیک و پالمیتیک، و مقاومت روزه ای در سطح یک درصد معنی دار گردید. اثر متقابل آبیاری×رقم نیز بر میزان اسید پالمیتیک، گلوکوزینولات و مقاومت روزه ای تاثیر معنی داری را در سطح یک درصد نشان داد. تنش خشکی آخر فصل و کاشت تاخیری، به میزان چشمگیری عملکرد روغن (۵۸٪)، درصد روغن (۸۰٪) و ویژگی های کیفی ارقام کلزا را کاهش داد. در بین ژنوتیپ های مورد مطالعه و با در نظر گرفتن کلیه شرایط این تحقیق، هیبرید Tassilo با میزان بالای عملکرد دانه (۴۴۹۷ کیلوگرم در هکتار)، عملکرد روغن (۲۰۰۷ کیلوگرم در هکتار)، محتوای کلروفیل (۱۴۳ mg g⁻¹fw)، اسیدهای چرب اولئیک و لینولئیک و داشتن اروسیک اسید (۴۱٪) و گلوکوزینولات دانه (۲۲.۸ μmol g⁻¹) به میزان استاندارد میتواند در توسعه کشت کلزا در مناطق معتدل سرد با اقلیم خشک و نیمه خشک موثر و قابل توصیه باشد.