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Responses of *Nigella damascena* L. and *Nigella sativa* L. to Drought Stress: Yield, Fatty Acid Composition and Antioxidant Activity

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ABSTRACT

***Nigella damascena* and *Nigella sativa* are two important species of the genus *Nigella* that have many medicinal and industrial applications. Both species are widely cultivated in arid and semi-arid areas, and are affected by abiotic stresses, especially drought stress. In the present study, the effect of water deficit stress on seed yield, oil percentage and yield, total phenolics and flavonoids content, and antioxidant activity of both species were investigated during two growing seasons (2018 and 2019). In this experiment, different levels of irrigation (severe stress, mild stress, and control) and two species (*N. sativa* and *N. damescena*) were studied as the main plots and subplots, respectively. Seed and oil yields decreased sharply due to drought stress in both species. Means of the seed and oil yields in *N. sativa* were 540.65 and 206.92 kg ha^{-1}, respectively, and seed and oil yields in *N. damascena* were 286.37 and 100.29 kg ha^{-1}, respectively. Mild stress increased the oil content in both species, but severe stress significantly decreased the oil content in *N. sativa*. Linoleic acid had the highest percentage in both species, followed by oleic and palmitic acids, in the order of their appearance. Drought stress decreased polyunsaturated fatty acids (linolenic acid and linoleic acid), but saturated ones (stearic and palmitic acids) increased under drought stress with increasing drought stress, and the amount of phenolics and flavonoids in both species increased (P< 0.05). Furthermore, the IC_{50} level decreased with increasing phenolics and flavonoids contents. In general, drought stress negatively affected seed and oil yields and edible oil quality in these species. Considering the acceptable seed and oil yields of *N. damascena* and its beneficial fatty acid compositions, it can be used in plant breeding programs and edible oil production.**

Keywords: Medicinal plants, Oil yield, Seed yield.

INTRODUCTION

Nigella damascena and *Nigella sativa* species are annual and herbaceous plants belonging to the Ranunculaceae family. *N. damascene*, commonly known as devil-in-the-bush, is grown as an ornamental plant in temperate regions of Europe, including the Mediterranean region (Jabbour *et al*., 2015). However, *N. sativa* is widely cultivated for various medicinal and industrial purposes in the Mediterranean Basin region and Asia (Rchid *et al*., 2004). Iran is one of the main centers of growth for *N. sativa* (Golkar and Nourbakhsh, 2019).The oil extracted from *N. sativa* seeds has been used in various therapeutic applications, including asthma, headache, dizziness, and blood pressure (Telci *et al*., 2014). Also, because of its high

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antioxidant properties, various medicinal properties such as antibacterial, antifungal, and anti-cancer have been stated for this plant (Majeed *et al*., 2020; Telci *et al*., 2014)

The seeds of *N. damascena* contains fatty acids (Telci *et al*., 2014), proteins and other biologically active compounds such as alkaloid compounds, different types of flavonoids, sesquiterpenes, sterols, and saponins (Fico *et al*., 2000; Telci *et al*., 2014).

Drought stress reduces the amount of water available to plants and leads to reduced yield and biomass, along with biochemical and physiological changes in the plant. Exposure of the plant to adverse drought conditions also leads to increased production of Reactive Oxygen Species (ROS). Plants use antioxidant defense systems to protect themselves against these ROS under environmental stresses (Bettaieb *et al*., 2012). Biosynthesis and accumulation of polyphenols are stimulated in response to abiotic stresses such as drought stress in plants (Navarro *et al*., 2006). Therefore, water-stressed plants may be a source of polyphenol compounds, through increasing the concentration of polyphenols inside the tissues and limiting biomass production (De Abreu and Mazzafera, 2005).

The fatty acid composition of the seed indicates how it is used for various food, industrial, or medicinal purposes. The mentioned species of *Nigella* (*N. sativa* and *N. damascene*) have a comparable composition in terms of their oil content, but there are differences between these species in view point of their quantitative values. Linoleic acid and oleic acid are the most important unsaturated fatty acids, whereas palmitic, stearic, and myristic fatty acids are the most important saturated fatty acids in these two species (Telci *et al*., 2014). The composition of fatty acids in crop seeds is mainly controlled by genetic effects (Ozer *et al*., 2020). Nevertheless, in some medicinal plants such as *Salvia officinalis* (Bettaieb *et al*., 2009) and *Carum carvi* (Laribi *et al*., 2009), the composition is partially affected by drought stress.

Regarding the commercial, medicinal, and multi-purpose values of products derived from *Nigella* species, much research is being done on this species. However, according to the literature review, few studies have been performed on the effect of drought stress on the changes of fatty acids and oil content of *Nigella* species. Bannayan *et al*. (2008) and Ghamarnia *et al*. (2010) reported that *N. sativa* has a relatively good tolerance to water deficit conditions. In other studies (Ghamarnia and Jalili, 2013; Shahbazi, 2019), it was reported that *N. Sativa* is sensitive to water stress. However, it is not clear how fatty acids composition will change under drought stress in these species, and presently, there is no study on the effect of drought stress on the fatty acid content of *N. damascena*. Therefore, this study aimed to investigate the effect of drought stress on oil content, fatty acids, and some secondary metabolites (total phenol content and total flavonoid content), as well as the antioxidant activity in the two species *N. sativa* and *N. damascena*.

MATERIALS AND METHODS

Experimental Design and Irrigation Treatments

This study was performed as split-plot in a Randomized Complete Block Design (RCBD) with four repetitions. Two genotypes of *N. sativa* and *N. damascena* were sown in the experimental farm of Shahrekord University, Shahrekord, Iran (32° 21′ N, 51° 49′ E, 2,083 m AMSL) in two growing seasons (2018 and 2019). The two genotypes and three irrigation regimes (supplying 100, 60, and 40% of the crop water requirements) were considered as the subplots and main plots, respectively. In the present experiment, the levels of 40, 60, and 100% were regarded as, respectively, severe and mild stresses, and the control. Each experimental plot consisted of furrows with 2 m length and 2 m wide, the rows were 25 cm apart. Buffering spaces of 1.5 m separated the plots. Irrigation regimens were specified by quantification of the MAWD (Maximum Allowable Water Depletions),

and the water stresses were applied at the onset of flowering and continued until maturity. Theta probes were utilized to measure alterations in soil moisture at various growth phases. After determination of crop water requirements, every subplot received irrigation (via an irrigation meter) upon reaching the relevant MAWD threshold. Moreover, the soil water quantity was expressed by the following equations (Allen, 1998):

$\theta_{irr}= (\theta_{FC}-\theta_{PWP})\times MAD$ (1)

$d= (\theta_F -\theta_{soil})\times D$ (2)

$V=d\times A\times 1000$ (3)

In the above equations, θ_{irr} and θ_{soil} are mean water in rhizosphere rooting depth at irrigation time (MAD threshold value) and mean water in plant rhizosphere, respectively. θ_{FC} and θ_{PWP} are volumetric soil contents, respectively, at FC (Field Capacity) and PWP (Permanent Wilting Point). D and d denote the plant rooting Depth and irrigation depth in m, respectively. A and V represents the subplot Area (m^2) and employed irrigation Volume (m^3), respectively.

The plant samples of every replication were dried in an aerated oven for 4 hours at 40°C and then pulverized by a mixer. The extraction of oil from powdered seeds (5 g) was done in a Soxhlet device utilizing petroleum ether as the dissolvent for 6 h based on the AOCS procedure (Cheikh-Rouhou *et al*., 2007), followed by calculation of oil contents in individual samples. Followed by extraction, separation of the solvent from the oil was performed by a vacuum evaporator.

Fatty Acids Profiling

To determine the fatty acid profile, by methylating the lipid extracts, fatty acid methyl esters (FAME) were produced for analyzing by Gas Chromatography (GC) (Goli *et al*., 2008). The gas chromatography of FAME was analyzed on an Agilent 6890N gas chromatograph fortified with a Flame Ionization Detector (FID). The utilized column was a HP-88 (0.25 mm id, 100 m, and film thickness of 0.2 μm).

Total Phenolics Content

Folin-Ciocalteu's reagent was used to evaluate the total phenolic of every extract based on the technique defined by Dewanto *et al*. (2002). The overall Total Phenolics Content (TPC) was presented as mg of GAE (Gallic Acid Equivalents) per gram of dry weight via the calibration curve of gallic acid.

Total Flavonoids Content

Total Flavonoids (TFD) content in methanolic extracts of *Nigella* was determined by the aluminum chloride procedure, according to Zhishen *et al*. (1999). Total flavonoids content in specimens was presented as mg Quercetin Equivalents (QE g^{-1}) dry weight.

Antioxidant Activity

The electron donation capability of the resultant methanolic extracts was determined according to Hatano *et al*. (1988). Inhibition Concentration (IC_{50}) of free radical DPPH was estimated as Equation (5):

$IC\ (\%)= [(A_{blank}-A_{sample})/A_{blank}]\times 100$ (5)

Where, A_{blank} and As_{ample} are the control reaction Absorbance and that using plant extract. The IC_{50} values were determined by the regression equation obtained from the inhibition percentage and the extracts' concentration. BHT was applied as a positive control.

Statistical Analysis

A split-plot design was employed on the basis of a randomized complete block design with four replications. Three irrigation levels (40%, 60% and 100%) and two *Nigella*

species (*N. sativa* and *N. damascena)* were considered as the main factor and sub-factor, respectively. Because the Bartlett test indicated homogeneity in the variance of data for the two years, the combined data of the two years were analyzed by the ANOVA with SAS statistical software (V.9.1) (SAS, 2004) and the mean comparisons were calculated using the Least Significant Difference ($LSD_{5\%}$) test.

RESULTS

Effects of Drought Stress on Seed and Oil Yields

Drought stress and species had significant effects on seed and oil yields, and oil content(%) ($P< 0.01$) (Table 1). The average of seed yield during the two years of study was recorded as 540.65 kg ha^{-1} for *N. sativa* and 286.37 kg ha^{-1} for *N. damascena (*Table 2). Drought stress significantly reduced seed yield by 41.4% and 80.73% under mild and severe stresses, respectively (Table 2). According to (Figure 1a), the seed yield decreased in both species with increase in drought intensity, but the decrease in yields was more severe under mild stress conditions in *N. sativa*, than *N. damascena*.

The mean of oil content was 37.95 and 35.37% for *N. sativa* and *N. damascena*, respectively (Table 2). With increase in the intensity of water tension, the oil yield (kg ha^{-1}) showed a significant decrease in both species. The severity of reduction in oil yield was greater in *N. sativa* rather than *N. damascena* (Figure 1b).

Effects of Drought Stress on Fatty Acids Content

Saturated Fatty Acids

The mean of palmitic acid were 15.75 and 12.71% in *N. sativa* and *N. damascene*, respectively (Table 3). The mean of palmitic acid was 13.22 %, 14.22 and 15.25% under normal, mild stress and severe stress conditions, respectively (Table 3). According to (Figure 2a), responses of the two species were almost similar in terms of palmitic acid changes, but the amount of palmitic acid content in *N. damascena* increased more strongly than *N. sativa* due to severe stress.

The effect of irrigation and year×irrigation was significant on stearic acid (Table 1). The means of stearic acid in *N. sativa* and *N. damascena* were 4.06%- and 4.23%, respectively (Table 3). With increase in the intensity of drought stress, the content of stearic fatty acid increased, but there was no significant difference between mild and control stress conditions (Table 3).

Unsaturated Fatty Acids

The effects of irrigation, species, and interaction effects of species × irrigation showed significant difference for unsaturated fatty acids (oleic acid and linoleic acid) (Table 1). The mean comparison demonstrated a higher content of oleic acid in *N. damascena* (30.73%) than *N. sativa* (24.45%) (Table 3). The percent of oleic acid was 26.29, 27.51 and 28.98 (%) under control, mild-stress and severe-stress conditions, respectively (Table 3). Although the response trends of the two species to drought stress conditions were almost similar for oleic acid content, the amount of oleic acid increased more sharply in *N. damascena* in severe stress (Figure 2b). The mean of linoleic acid was 51.99 and 55.5% in *N. damascena* and *N. Sativa*, respectively (Table 3). The mean comparison between different levels of irrigation showed a significant decrease under normal conditions, mild stress and severe stress, as 56.42%, 54.09% and 50.73%, respectively (Table 3). Comparing the trend of responses to drought stress in the two species showed that with increase in the level of stress, the amount of linoleic

Table 1. Combined analysis of variance for different traits under irrigation regimes (control, mild and sever stress) in *N. sativa* and *N. damascena* at 2018 and 2019.

Source of variation[a]	DF	Seed yield	Oil content	Oil yield	Total phenolic content	Total flavonoid content	IC50	Palmitic acid (C16:0)	Stearic acid (C18:0)	Oleic acid (C18:1)	Linoleic acid (C18:2)	Linolenic acid (C18:3)	Saturated Fatty Acids (SFA)	Unsaturated Fatty Acids (USFA)	SFA/USFA
Y	1	6724.8^{ns}	2.26^{ns}	1102.4^{ns}	14.21^{ns}	12.77^{ns}	0.347^{ns}	2.66^{ns}	10.69	31.40^{ns}	16.61^{ns}	0.0015^{ns}	2.22^{ns}	2.22^{ns}	0.104^{ns}
Rep/Y	6	17296.0	1.37	2321.6	3.82	2.74	0.088	0.44	0.33	6.16	2.76	0.0008	0.87	0.87	0.093
I	2	947277.3**	15.4**	126855.6**	298.58**	89.36**	1.730**	12.28**	4.10**	21.81**	98.13**	0.0432**	29.68**	29.68**	2.774**
Y×I	2	4162.9^{ns}	1.72^{ns}	660.9^{ns}	4.07^{ns}	2.26^{ns}	0.032^{ns}	1.79**	1.01**	0.93^{ns}	10.17**	0.0034^{ns}	5.40**	5.40**	0.575**
E_a	12	2645.8	0.48	349.1	3.14	1.71	0.013	0.16	0.04	0.28	0.49	0.0022	0.18	0.18	0.021
S	1	581917.2**	59.98**	102317.4**	51.19*	0.01^{ns}	0.058*	83.24**	0.27^{ns}	354.44**	110.57**	0.0831**	73.96**	73.96**	7.189**
Y×S	1	587.2^{ns}	6.06^{ns}	149.3^{ns}	4.29^{ns}	0.34^{ns}	0.511**	0.04^{ns}	0.02^{ns}	1.24*	1.84^{ns}	0.0930**	0.01^{ns}	0.01^{ns}	0.021^{ns}
I×S	2	67640.3**	9.35^{ns}	13813.0**	145.16**	84.20**	1.605**	1.84**	0.31^{ns}	1.37*	4.50**	0.0136*	1.47^{ns}	1.47^{ns}	0.289**
Y×I×S	2	148.3^{ns}	5.03^{ns}	77.3^{ns}	0.49^{ns}	2.97^{ns}	0.011^{ns}	0.78^{ns}	0.16^{ns}	0.14^{ns}	0.35^{ns}	0.0172*	0.57^{ns}	0.57^{ns}	0.109^{ns}
E_b	18	8459.1	2.42	1539	7.17	3.05	0.007	0.20	0.11	0.21	0.47	0.002	0.37	0.37	0.037

[a] Y: Year, Rep: Replication, I: Irrigation, E_a: Pooled Error a, S: Species, E_b: Pooled Error b. ns: Non-significant, * and **: Significant at 5 and 1% probability levels, respectively.

Table 2. Mean comparisons of main effects of species and irrigation on different studied traits averaged over two years of study (2018 and 2019).[a]

		Seed yield (kg ha^{-1})	Oil content (%)	Oil yield (kg ha^{-1})	Total phenolics content (mg GAE g^{-1} DW)	Total flavonoids content (mg QE g^{-1} DW)	IC50 (µg mL^{-1})
Species							
	N. sativa	540.65^{a}	37.95^{a}	206.92^{a}	60.67^{b}	24.45^{a}	2.43^{b}
	N. damascena	286.37^{b}	35.37^{b}	100.29^{b}	63.06^{a}	24.49^{a}	2.51^{a}
Irrigation							
	Control	696.05^{a}	35.82^{b}	254.4^{a}	57.78^{c}	22.04^{c}	2.84^{a}
	Mild stress	410.33^{b}	37.95^{a}	157.54^{b}	60.39^{b}	23.95^{b}	2.5^{b}
	Severe stress	134.15^{c}	36.21^{b}	48.88^{c}	67.43^{a}	27.42^{a}	2.08^{c}

[a] Means with the same letters have no significant difference, in each column (LSD, P< 0.05).

Table 3. Mean comparisons of main effects of species and irrigation on fatty acids profiles averaged over two years of study (2018 and 2019).[a]

		Palmitic acid (%)	Stearic acid (%)	Oleic acid (%)	Linoleic acid (%)	Linolenic acid (%)	Saturated Fatty Acids (SFA) (%)	Unsaturated Fatty Acids (USFA) (%)	SFA/USFA
Species									
	N. sativa	15.75^{a}	4.06^{a}	24.45^{b}	55.5^{a}	0.24^{b}	19.81^{a}	80.19^{b}	4.07^{b}
	N. damascena	12.71^{b}	4.23^{a}	30.73^{a}	51.99^{b}	0.33^{a}	16.94^{b}	83.06^{a}	4.96^{a}
Irrigation									
	Control	13.22^{c}	3.72^{b}	26.29^{c}	56.42^{a}	0.35^{a}	16.94^{c}	83.06^{a}	4.96^{a}
	Mild stress	14.22^{b}	3.91^{b}	27.51^{b}	54.09^{b}	0.28^{b}	18.13^{b}	81.87^{b}	4.58^{b}
	Severe stress	15.25^{a}	4.81^{a}	28.98^{a}	50.73^{c}	0.23^{c}	20.06^{a}	79.94^{c}	4.01^{c}

[a] Means with the same letters have no significant difference, in each column (LSD, P< 0.05).

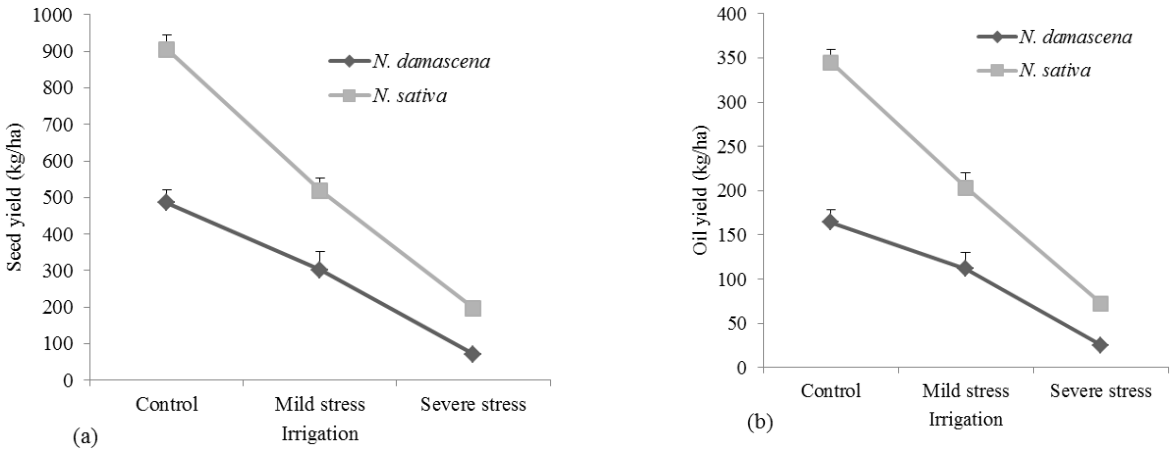

Figure 1. Response of *N. sativa* and *N. damascena* to different levels of irrigation for seed yield (a) and oil yield (b) averaged over two years of study (2018 and 2019).

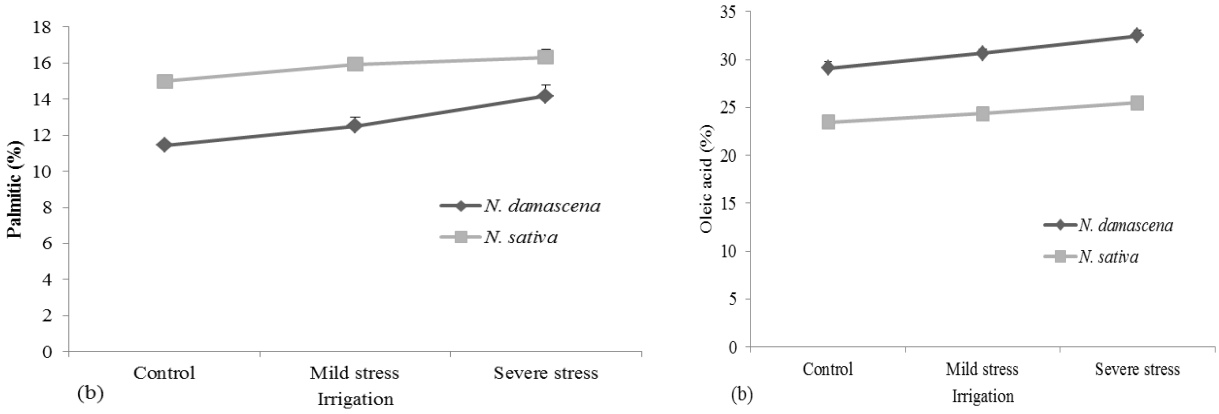

Figure 2. Response of *N. sativa* and *N. damascena* to different levels of irrigation for palmitic acid (a) and oleic acid (b) averaged over two years of study (2018 and 2019).

acid decreased, but its decrease was more severe in *N. damascena*. (Figure 3a). The linolenic content was lower than all other studied fatty acids. It was 0.24 and 0.33% in *N. sativa* and *N. damascena*, respectively (Table 3). The content of linolenic acid was 0.35, 0.28 and 0.23% under normal, mild-stress and severe-stress conditions, respectively (Table 3). The responses trends of the two species for linolenic acid were different under drought stress (Figure 3b). In this regard, mild stress conditions resulted in decrease of linolenic acid in *N. damascena* species, but no significant decrease was observed in *N. sativa* species (Figure 3b).

Content of Total Saturated and Unsaturated Fatty Acids

Total saturated fatty acids in *N. Sativa* and *N. damascena* were 19.81 and 16.94%, respectively (Table 3). The mean of saturated fatty acids under normal irrigation, mild stress and severe stress were considered as 16.94, 18.13, and 20.06%, respectively (Table 3). With increase in intensity level of drought stress, total unsaturated fatty acids showed a significantly decrease in both species (Table 3).

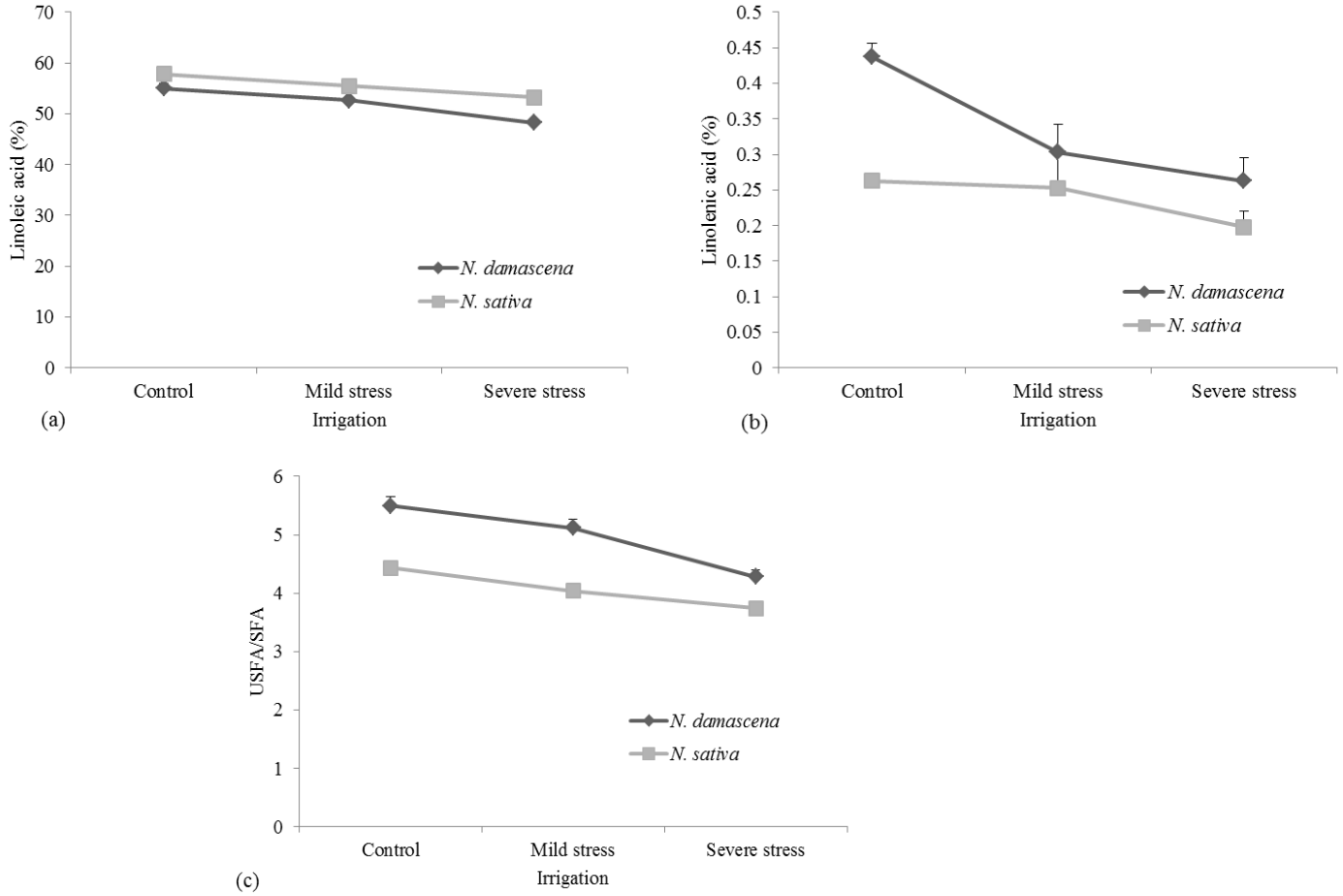

Figure 3. Response of *N. sativa* and *N. damascena* to different levels of irrigation for linoleic acid (A), linolenic acid (B) and Saturated Fatty Scids (SFA)/Unsaturated Fatty Acids (USFA) (C) averaged over two years of study (2018 and 2019).

A significant difference was observed for ratio of total unsaturated fatty acids to total saturated fatty acids (Table 1). This ratio was 4.07 and 4.96 for *N. sativa* and *N. damascena*, respectively (Table 3). This ration was reduced under drought stress in both species, but the reduction was more severe in *N. damescana* (Figure 3c).

Effects of Drought Stress on Total Phenolics and Flavonoids Content

The effects of drought stress, and species × irrigation was significant on TPC at $P < 0.01$ (Table 1). The mean comparison between different levels of water stress indicates a significant increase in TPC (Table 2). The trend responses of the studied species were different to drought stress for TPC, such that in mild stress, TPC did not show significant changes in *N. sativa* compared to *N. damascena* (Figure 4a). In severe stress condition, TPC increased sharply in *N. sativa*, while in *N. damascena* it did not have significant changes (Figure 4a). On average, TPC (63.06 mg GAE g^{-1} DW) was higher in *N. damascena* than *N. sativa* (60.67 mg GAE g^{-1} DW) (Table 2). The effects of irrigation and irrigations × species interaction were significant for TFD content ($P < 0.01$) (Table 1). On average, drought stress increased TFD content (Table 2). Mild stress resulted in increase in TFD

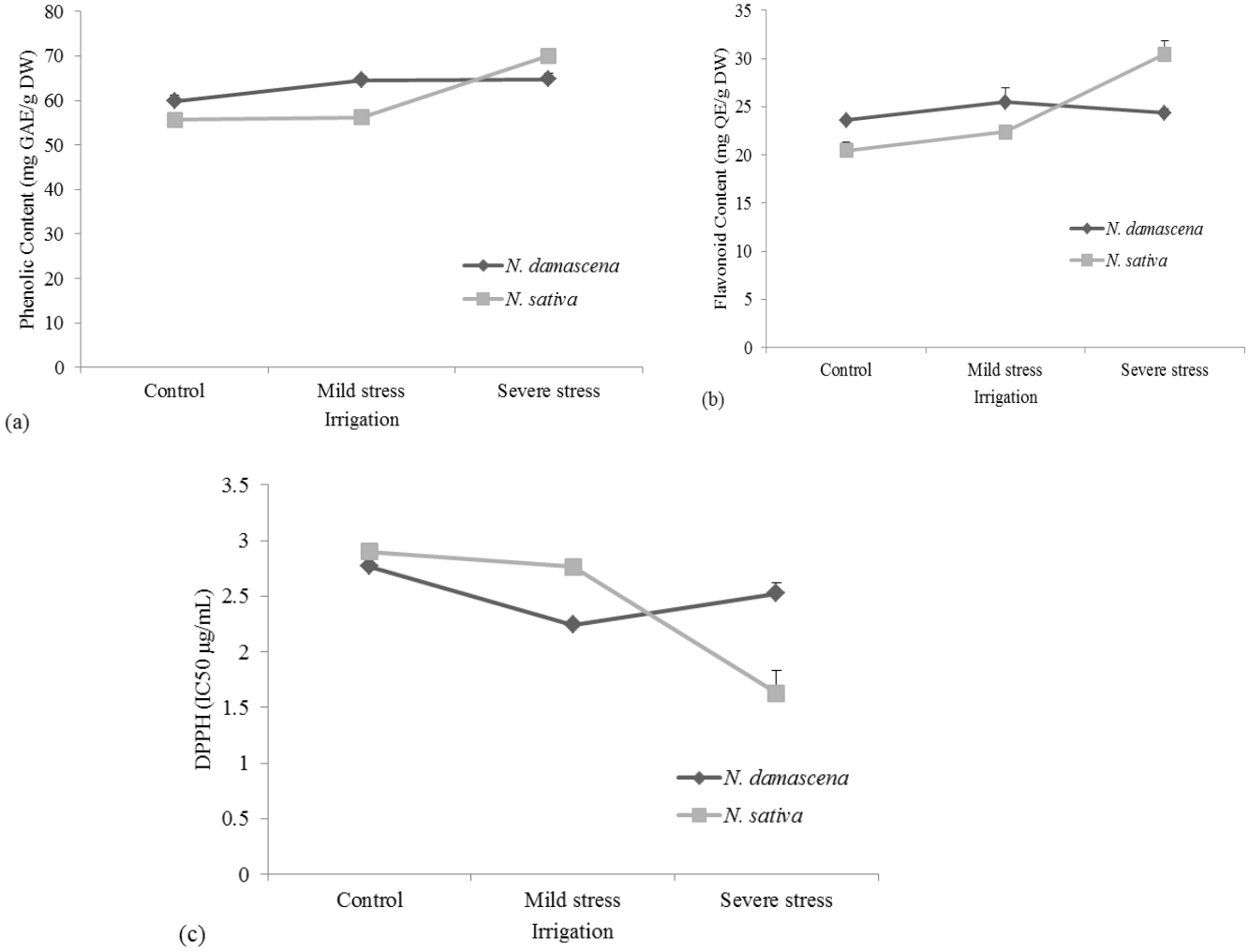

Figure 4. Response of *N. sativa* and N. *damascena* to different levels of irrigation for total phenolic content (A), total flavonoid content (B) and IC_{50} value (C) averaged over two years of study (2018 and 2019).

content in both species, but under severe-stress condition response of species was different for TFD content. It showed an increasing trend in *N. sativa*, but a decreasing one in *N. damascena* (Figure 4b). The means of TFD content were recorded as 27.42, 23.95, and 22.02 mg QE g^{-1} DW under severe-stress, mild-stress and control conditions, respectively (Table 2).

Drought Stress Effects on Antioxidant Activity

All the main and interaction effects were significant on IC_{50} value, except for year, and interaction effects of year×irrigation and year×irrigation×species (Table 1). The IC_{50} value was 2.43 and 2.51 µg mL^{-1} in *N. sativa* and *N. damascena*, respectively (Table 2) .The mean comparison between different irrigation levels showed a significant increase in DPPH scavenging activity (decrease in IC_{50}) with increasing stress levels (Table 2). The IC_{50} value was recorded as 2.84, 2.5, and 2.08 µg mL^{-1} under non-stress conditions, mild-stress and sever- stress, respectively (Table 2). The trend responses of the two species under drought stress were completely different to drought stress (Figure 4c). In *N. damascena*, The IC_{50} value decreased initially due to

mild stress but it then increased with increase in stress intensity (Figure 4c). On the other hand, in *N. sativa*, no significant change was observed in mild stress, but it decreased significantly with increase in drought tension (Figure 4c).

DISCUSSION

Drought stress is considered as a restricting factor affecting growth rate, seed yield, and morpho-physiological traits in plants (Lamaoui *et al*., 2018). How plants respond to water deficit depends on the stress intensity, the period of stress, phonologic stage of the plant, genotype, and plant species (Farooq *et al*., 2012). In this study, the effects of water deficit on grain and oil content, fatty acid composition, and some secondary metabolites were investigated in *N. sativa* and *N. damascena*.

Seed yield and oil yield in both species decreased sharply due to stress, indicating the adverse effect of drought stress on these traits. Moreover, it suggests that oil yield is more affected by seed yield than oil percentage (Table 2). Previous studies have also reported a decrease in grain yield in *N. sativa* due to drought stress (Bannayan *et al*., 2008; Shahbazi, 2019). The effect of drought stress on the flowering and grain filling stage is very important because drought stress in these stages reduces the number of fertile flowers, the number of seeds, and seed weight (Merajipoor *et al*., 2020; Ozer *et al*., 2020). This reduction can be due to physiological conditions such as reduced photosynthesis and enzymatic activities, closed stoma, and reduced carbohydrates (Merajipoor *et al*., 2020; Ozer *et al*., 2020). Mild stress increased the oil content, but it decreased with increasing stress (severe stress). Similar to these findings, the increase in the intensity of drought stress led to a significant decrease in oil yield in other oilseeds such as sesame (Eskandari *et al*., 2015) and soybeans (Ghassemi-Golezani and Lotfi, 2013).

The ratio of unsaturated fatty acids to saturated ones is an important index for the strength of the nutritional value of plant oils. Both species are rich in unsaturated fatty acids (Table 3). The ratio of unsaturated to saturated fatty acids was more in *N. damascena* than *N. sativa* (Table 3). In previous studies, linoleic acid has been reported as the most important fatty acid in *N. sativa* and *N. damascena* (Aitzetmuller *et al*., 1997). According to the reports of Telci *et al*. (2014), the percentage of unsaturated fatty acids in *N. damascena* and *N. sativa* was 79 and 81%, respectively. The highest contents for saturated fatty acid in *N. sativa* (12.5%) and *N. damascena* (9.7%) were reported as palmitic acid (Telci *et al*., 2014).

In some studies, small amount of lauric fatty acid has been reported in *N. damascena* (0.6-0.05%) (Hendawy *et al*., 2012) but lauric acid was not reported in *N. sativa*. However, the difference in the content of fatty acids in these species can be due to genetic differences of these species, climatic conditions (Telci *et al*., 2014), and intenal factors that affect fatty acids synthesis in seeds of oily crops (Harwood, 1996).

The degree of fatty acids saturation is the most selective factor in conserving membrane fluidity, which provides a suitable environment for membrane function and plant adaptation processes under environmental stresses conditions. In this study, drought stress decreased linolenic and linoleic fatty acids and increased oleic, stearic, and palmitic fatty acids. In other words, drought stress decreased polyunsaturated fatty acids and increased saturated fatty acids. This response might be due to accelerated lipid accumulation and/or shorter duration of all enzymatic activities, including Δ_{12} desaturase (Xu and Beardall, 1997). This similar trend was also reported under drought stress in *Cuminum cyminum* L. (Bettaieb *et al*., 2011) and *Salvia officinalis* (Bettaieb *et al*., 2009).

Under drought stress conditions, a lower conversion of oleic acid to linoleic acid could result from a diminished time for seed filling in early maturity condition (Liu and

Guan, 2016). This phenomenon could be the main reason for reduction of linoleic acid ratio under drought-stress condition. Limitations due to drought stress may cause an earlier embryo development, which subsequently stimulates enzymatic activities related to the biosynthesis of fatty acids and determine the final composition of fatty acids (Baldini *et al*., 2002; Bettaieb *et al*., 2012).

In this study, with increasing drought stress, the amount of phenolics and flavonoids compounds in both species increased (Table 2). This result indicates that *Nigella* species uses the biosynthesis of phenolics and flavonoids compounds in response to oxidative stress caused by drought stress. An increase in TPC under drought stress in various plants including black seed (Kabiri *et al*., 2014), cumin (Bettaieb *et al*., 2011), pepper (Navarro *et al*., 2006), and *Achillea* species (Gharibi *et al*., 2016) was also reported, which is consistent with the results of this study.

In the present study, a correlation was observed between the phenolics and flavonoids content with DPPH scavenging activity, indicating that with increase in the contents of total phenolics and flavonoids, the value of IC_{50} decreased. The activity of antioxidants with a low molecular weight such as polyphenols (Sreenivasulu *et al*., 2000) and flavonoids (Sgherri *et al*., 2003) can lead to the elimination of free radicals generated during stress.

CONCLUSIONS

According to these findings, drought stress decreased the seed and oil yields of *Nigella* and edible oil quality and its unsaturation degree. In other hand, water deficit reduced the proportion of the polyunsaturated fatty acids, which are of great importance for human nutrition. Moreover, drought stress was found to significantly influence the phenolic and flavonoid contents. Antioxidant activity of *Nigella* seeds was positively correlated with their phenolics and flavonoids contents, which increased under water-deficit condition. This finding indicated that genus *Nigella* uses the accumulation mechanism of phenolics and flavonoids compounds in response to oxidative stress due to drought stress. Regarding the acceptable yield of *N. damascena* and its higher beneficiary unsaturated fatty acid compositions, this species can be used in plant breeding programs and edible oil production.

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پاسخ *Nigella damascena* L. و *Nigella sativa* L. به تنش خشکی: عملکرد، ترکیبات اسیدهای چرب و فعالیت آنتی‌اکسیدانی

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چکیده

گونه‌های *N. sativa* و *N. damascena*، دو گونه مهم از جنس Nigella هستند که دارای کاربردهای داروئی و صنعتی بسیاری می‌باشند. هر دو گونه در مناطق خشک و نیمه خشک که تحت تاثیر تنش خشکی می‌باشد، کشت و کار می‌شوند. در مطالعه حاضر تاثیر تنش خشکی روی عملکرد دانه، درصد و عملکرد روغن، محتوای کل فنول و فلاونوئید و همچنین فعالیت آنتی‌اکسیدانی هر دو

گونه در طی دو فصل رشد زراعی (۱۳۹۷ و ۱۳۹۸) مورد مطالعه قرار گرفت. در این آزمایش، سطوح مختلف آبیاری (شاهد، تنش ملایم و تنش شدید) و دو گونه (*N. sativa* و *N. damascena*) به ترتیب به عنوان کرت‌های اصلی و فرعی مورد مطالعه قرار گرفتند. عملکرد دانه و روغن در هر دو گونه با شرایط تنش خشکی کاهش نشان داد. میانگین عملکرد دانه و روغن در *N. sativa* به ترتیب ۵۴۰/۶۵ و ۲۰۶/۹۲ کیلوگرم درهکتار بود. همچنین عملکرد دانه و روغن در *N. damascene* به ترتیب ۲۸۶/۳۷ و ۱۰۰/۲۹ کیلوگرم درهکتار بود. تنش خشکی ملایم منجر به افزایش محتوای روغن در هر دو گونه شد، اما تنش شدید به صورت معنی‌داری باعث کاهش محتوای روغن در *N. sativa* شد. در هر دو گونه لینولئیک اسید دارای بیشترین درصد و به دنبال آن اولئیک اسید و پالمتیک اسید قرار گرفتند. تنش خشکی منجر به کاهش اسیدهای چرب غیر اشباع (لینولنیک اسید و لینولئیک اسید) و افزایش اسیدهای چرب اشباع (استئاریک اسید و پالمتیک اسید)، محتوای کل فنول و فلاونوئید شد. مقدار IC_{50} با افزایش محتوای کل فنول و فلاونوئید کاهش نشان داد. به صورت کلی، تنش خشکی تاثیر منفی بر روی عملکرد دانه و روغن و کیفیت روغن خوراکی در این گونه‌ها داشت. با توجه به عملکرد دانه و روغن قابل قبول و ترکیب اسیدهای چرب مطلوب گونه *N. damascena* این گونه می‌تواند در برنامه‌های اصلاح نباتات به منظور تولید روغن خوراکی مورد استفاده قرار گیرد.