

EFFECTS OF WEED CONTROL ON SEED YIELD AND FATTY OIL RATIO OF BLACK CUMIN (*Nigella sativa* L.)

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ABSTRACT

Nigella sativa, also known as black cumin, is one of the medicinal plants, its seeds show a wide therapeutic potential, and they are used as spice in different kind of foods. The effects of different herbicides on yield and some yield components of Cameli Black cumin cultivar under Adana ecological conditions were investigated in this experiment that conducted between 2015-2016, 2016-2017 and 2017-2018 growing seasons in the Research and Application Area of Field Crops Department of Agricultural Faculty of Cukurova University. Three different herbicides were applied in nine different applications. Plant height, number of branches, number of capsules in the plant, seed yield and oil ratio differed according to application periods. The highest seed yield was obtained from the treatment of hand hoeing. Malondialdehyde and proline analysis were performed to detect oxidative stress and it was found that hand hoeing caused less stress in plants compared to other treatments. According to the results of the study, it was concluded that hand hoeing instead of herbicide application for the control of weeds in black cumin is more appropriate for yield and ecological perspective.

Keywords: Black cumin, morphological, herbicide, stress, malondialdehyde, proline.

INTRODUCTION

Black cumin seeds have been used in folk medicine for many years and are one of the widely known spice plants besides their therapeutic properties, it is used in folk medicine because of its urinary and milk enhancing, appetizing, gas and menstrual expectorant properties. Black cumin seeds are the most important part of the plant, and the seeds contain about % 30-40 crude oil. The seeds also contain approximately 0.5-1.4% essential oil (Kucukemre 2009; Gutiérrez Prieto and Kirici, 2020). The seed essential oil and its main active constituent, thymoquinone, are reported to exhibited protective effect against many diseases depending on its high antioxidant activity (Mariod et al., 2009). Currently, black cumin seed-producing countries are: India, Pakistan, Sri Lanka, Bangladesh, Nepal, Egypt, Syria, Saudi Arabia, Turkey, Iran, and Iraq (Haq et al., 2015). Black cumin is generally cultivated under dry farming conditions. Growing black cumin under these conditions often results in low seed yield and quality losses due to limited rainfall, the requirement to compete with weeds (Can et al., 2021). The production area as well as amount of the production had been increasing in the years and years. In the Turkey, the annual production of black cumin was 3.322 metric tons in 2018 crop production season from a harvested area of 3386,4 ha (Kirici et al., 2020). The

demand of black cumin seed and its oil has also been increasing both in local and national markets for consumption purpose. Black cumin Seed Oil Market size was over USD 15 million in 2018 and industry expects significant gains by 2025 (Ahuja and Singh, 2019).

Recently, the chemical control against to weeds has brought some problems such as environmental pollution and resistance to herbicides. Weeds are in competition with the cultivated plants in terms of growing area, nutrients, water, light and carbon dioxide. When any of these substances is limited by weeds, the growth of the culture plant begins to slow down. Weeds cause a decrease in the amount of crop to be harvested and consequently a decrease in yield (Aldrich and Kremer, 1997). Plants are exposed to biotic and abiotic stresses in natural environments during their whole life circle. The frequently-occurring abiotic stresses includes extreme temperature, high salinity, excessive light, water deprivation, pollutants such as ozone and herbicides, high concentration of heavy metals, excessive ultra violet radiation and so on. Malondialdehyde (MDA) is a widely used marker of oxidative lipid injury caused by environmental stress (Kong et al., 2016). MDA accumulation increased with the increase of the herbicide dosage (Jin et al., 2010). In plants, Proline accumulation has been reported to occur after salt, drought, high temperature,

low temperature, heavy metal, pathogen infection, anaerobiosis, nutrient deficiency, atmospheric pollution and UV irradiation (Verbruggen and Hermans, 2008).

In order to achieve the yield and quality targets, weed control is a necessity and we need to follow the ecological balance as much as possible in this process. The aim of this research was to determine effects of some of the weed control methods such as herbicide use and hand hoeing on the seed yield, yield components and oil content of the seeds as well as proline and malondialdehyde content of the plant in black cumin

MATERIALS and METHODS

Cameli cultivar which was developed and registered at 8 April 2014 by the Transitional Zone Agricultural Research Institute (Eskisehir) was used in this study.

Cultivating applications

The field experiments were conducted at research area of Department of Field Crops, University of Cukurova during winter season for three years. Experimental site was located in Southern part of Turkey, it has a Mediterranean climate zone. Monthly average temperature and rainfall during the experimental years are presented in Table 1. Soil samples were taken randomly from 0-30 and 30-60 cm depth. It has been determined that the soils of the experimental area have loamy (42.4 % and 46.3%) and clay (34.7% and 32.5%) texture, their pH (7.8 and 7.7) and salt (0.2 mmhos cm⁻¹) content were normal, the organic matter content (1.1 % and 0.9%) was low, and the phosphorus level (108 and 63 kg ha⁻¹) and potassium level (659 and 545 kg ha⁻¹) were sufficient. Fe, Zn Mn and Cu contents were 3.7 and 3.5, 0.3 and 0.3, 8.9 and 8.1 and 1.2 and 1.0 mg kg⁻¹, respectively.

Table 1. Monthly average temperature and rainfall values for the experiment area during the experimental

Years	November	December	January	February	March	April	May	June	
Average Temperature (°C)									Mean
2015-2016	17.5	11.8	8.7	13.9	15.7	20.5	21.6	27.1	17.1
2016-2017	15.6	9.0	8.7	10.7	15.2	18.5	21.8	26.2	15.7
2017-2018	15.9	12.6	10.5	13.5	16.8	20.1	24.4	26.4	17.5
Rainfall (mm)									Total
2015-2016	10.5	0.6	138.4	83.1	67.1	36.6	87.9	45.6	465.2
2016-2017	11.9	216.3	52	0.8	65.4	65.9	45.9	17.3	475.5
2017-2018	122.7	33.0	324.3	64.8	40.1	35.0	29.5	25.6	675.0

*Data were taken from Adana Meteorology Station

The experiment carried out in randomized complete block design with four replications. The plot size was 3x1.25 m (6 row per plot). Seeds were sown at the last week of November and harvest dates were varied according to years (Table 2). 15 kg ha⁻¹ seeds per hectare were sown by hand to a depth of 1-2 cm. Fertilization was carried out

as pure 40 kg ha⁻¹ P (Triple Super Phosphate) and 60 kg ha⁻¹ N (Ammonium Nitrate) during seed sowing. Seedlings were thinned out to 10 cm within row at two weeks after emergence. The first irrigation was carried out after sowing. Subsequent irrigations were done when it was needed.

Table 2. Some imported growing dates of black cumin experiment

	I. Year	II. Year	III. Year
Sowing date	26/11/2015	28/11/2016	15/11/2017
Emergence date	18/12/2015	03/01/2017	03/12/2017
Flowering date	01/04/2016	15/04/2017	04/04/2018
Harvest date	13/06/2016	19.07.2017	07/06/2018
Vegetation period	200 days	229 days	203 days

Hoeing and herbicide applications

In the experiment, Farmson super (active ingred is 50 g l⁻¹ Quizalofop-p-ethyl) as pre-sowing herbicide, Fusilade Forte (active ingred is 150 g l⁻¹ Fluzifop-P-Butyl) as pre-emergent herbicide and Challenge SC 600 (active ingred is 600 g l⁻¹ Aclonifen) as post-emergent herbicide were tested. The weed control treatments were as following 1) application of pre sowing herbicide 2) application of pre emergent herbicide 3) application of post emergent herbicide 4) application of pre-sowing herbicide+ pre-emergent herbicide 5) application of pre-sowing herbicide+ post emergent herbicide 6) application of pre emergent

herbicide +post emergent herbicide 7) application of pre sowing herbicide +pre-emergent herbicide +post emergent herbicide 8) hand hoeing 9) control with without weed control. Biomass of weeds was weighted as total aboveground dry weight for each plot at harvesting time of black cumin.

Determination of yield and morphological properties

When the fruit peels began to darken and crackings were seen, plants were harvested by hand. Ten plants were selected at maturity randomly from each plot for recording plant height (cm), number of branches per plant, number of

capsules per plant. Two samples of 1000 seeds were taken from each plot to record 1000-seed weight. Seed yield was recorded with middle four row on per plot basis and was converted to kg ha⁻¹. Fatty oil ratio was determined according to The American Oil Chemists Society (AOCS) method (AOCD, 1993). 5 grams of whole seeds were milled and the extraction was by soxhlet apparatus with n-hexan for 6 hours, after extraction, the solvent was evaporated, than weighed. The seed oil content was reported as the mass percent (crude oil weight (g)/ground seeds weight (g) × 100).

Proline analyse

Proline content was determined under herbicide applying and hoeing stress according to the method of (Bates 1973) on total harvested vegetative parts of plants at 2018. In the case, 500 mg of fresh plant material was homogenized with 3% sulpho salicylic acid and filtered at blue band filter paper. Then 2 ml sample was put in a glass tube, then 2 ml ninhydrin solution (which contains ninhydrin, ortophosphoric acid and acetic acid) and 2 ml acetic acid were added into the tube. After that it was incubated in boiling water for one hour, samples put into ice bath. Then, 4 ml toluene put on the samples and read at 520 nm spectrophotometrically. Proline content of samples was calculated according to standard curve which drowns with L-proline.

Malondialdehyde analyse

Lipid peroxidation of membranes was estimated by measuring malondialdehyde content according to modified Hodges et al.'s (1999) protokol. Malonialdehyde content was determined at 2018. 500 mg dry leaf material was

extracted with 80% ethyl alcohol and santrifuged 10 minutes at 3000 g. After this, treatment was conducted as two stage. For first stage 2 ml TBA (-) solution which contains 20% TCA and 0.01% BHT in it; samples incubated in 95°C waterbath for 25 minutes; after ice bath santrifuged again 15 minutes at 3000 g. Distinctively from first stage, (+) TBA solution was used instead of (-) TBA solution, which had also 0,65% in it, used in second stage. First samples read at 532 and 600 nm; second stage samples read at 440, 532 and 600 nm spectrophotometrically. Absorbans values put into formulas below and MDA content calculated.

$$1. [(ABS_{532+TBA})-(ABS_{600+TBA})-(ABS_{532-TBA})-(ABS_{600-TBA})]=A$$

$$2. [(ABS_{440+TBA}-ABS_{600+TBA})/0.0571]=B$$

$$3. \text{nmol MDA / ml} = (A-B)/157\ 000 / 10^6$$

Statistical Analyses

The data obtained from the study were analyzed for variance by using the MSTAT – C statistical package program according to the randomized complete block design, and standard deviation values were calculated for Proline and MDA analysis. The differences between the mean values were compared by using Duncan's new multiple range test (MRT) at 5% significance level (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Results of statistical analysis for the effects of experimental factors (yaer and treatments) on yield and some yield components of black cumin with P values were given in Table 3.

Table 3. ANOVA results for some yield and quality traits of black cumin.

Experimental Factors	DF	Weed Weights (kg ha ⁻¹)	Plant height (cm)	Number of branches	Number of capsules	Seed yield (kg ha ⁻¹)	Thousand Seed W. (g)	Fatty oil Ratio (%)
Mean Squares								
Year	2	898.14**	6059.79**	0.62**	429.06**	444519.01**	1.84**	12.21*
Error	6	0.49	3.69	0.02	0.05	7036.43	0.03	1.26
Treatment	8	107.14**	160.01**	0.82**	120.19**	591131.54**	0.15**	43.38**
Year x Treatment	16	34.73**	41.14**	0.41**	19.68**	27673.03**	0.03	6.93*
Error	48	0.60	8.24	0.14	0.29	8597.09	0.02	3.20
C.V.(%)		9.97	6.32	13.38	7.17	36.40	7.48	5.26

* and ** are significant at p=0.05 and p=0.01, respectively.

Weed species encountered in the experimental plots

Weed species encountered in the experimental plots are given in Table 4. As seen in the table, totally 23 species of weeds from 15 plant families were determined in the experimental plots during the experimental years. *Cyperus*

rotundas, *Portulaca oleracea*, *Amaranthus retrqflexus*, *Prosophis farcta*, *Convolvulus arvensis*, *Xanthium strumarium*, *Cynodon dactylon* were found the most intense in cotton, corn and soybean fields in Cukurova region (Gonen, 1999).

Table 4. Determined weed species of black cumin in experiment years.

Family	Species	Family	Species
Alliaceae	<i>Allium</i> sp.	Brassicaceae	<i>Sinapis arvensis</i>
Poaceae	<i>Alopecurus myosuroides</i>	Boraginaceae	<i>Echium</i> sp.
	<i>Avena sterilis</i>	Lamiaceae	<i>Lamium purpureum</i>
	<i>Avena fatua</i>	Polygonaceae	<i>Polygonum aviculare</i>
Asteraceae	<i>Centaurea</i> sp.	Ranunculaceae	<i>Ranunculus</i> sp.
	<i>Matricaria</i> sp.	Ranunculaceae	<i>Ranunculus</i> sp.
	<i>Sonchus</i> sp.		<i>Lathyrus annuus</i>
Amaranthaceae	<i>Chenopodium album</i>	Fabaceae	<i>Medicago</i> sp.
Convolvulaceae	<i>Convolvulus arvensis</i>		<i>Medicago scutellata</i>
Apiaceae	<i>Daucus carota</i>		<i>Vicia narbonensis</i>
Geraniaceae	<i>Geranium rotundifolium</i>	Papaveraceae	<i>Papaver rhoeas</i>
			<i>Fumaria officinalis</i>

Weeds are one of the factors which negatively affect the quality and yield of crops. Continuous changes in weed flora takes place either by climatic change or through the introduction of new weed species into the agro ecosystem. Therefore, comparative weed surveys were carried out in Cukurova Region, across three provinces namely Adana, Mersin and Osmaniye, and determined the most troublesome weed species in corn fields, as a result, total 42 weed species belonging to 19 families were identified (Hancerli and Uygur, 2017). Among the identified weed species, *Chenopodium album*, *Convolvulus arvensis*, *Papaver rhoeas*, *Avena sterilis*, *Polygonum aviculare*, *Medicago* sp. and *Vicia* sp. were common plants with our research in spite of different growing period. Sterile wild oat (*Avena sterilis* L.) and wild

mustard (*Sinapis arvensis* L.) were determined as the main harmful weeds in wheat fields in Cukurova region (Tunk and Uygur, 2020). The herbicide doses should be established for weeds and recommended according to the their economic threshold level in a wheat field in the region for environmental protection, food safety, herbicide resistance problem.

Weed Biomass yield in the experimental plots

Weed biomass yield determined in the experimental plots treated with different weed control treatments in the experimental years are given in the Table 5. Weed control method significantly affected the weed biomass yield in all the experimental years (Table 3).

Table 5. Weed biomass yields (t/ha) in the experimental plots treated with different weed control methods in experiment years (Pr.S= pre-sowing, Pr.E=pre-emergence, Po.E= post-emergence).

Treatments	Weed Weights (kg ha ⁻¹)			
	2016*	2017**	2018*	Means of years**
Control	5.89 b*	6.37 a	24.52 a	12.26 a
Pr.S.	5.82 b	4.77 cd	13.90 d	8.16 de
Pr.E.	4.97 c	4.80 cd	14.13 d	7.94 e
Po. E.	4.51 d	4.88 c	20.92 b	10.10 b
Pr.S+Pr.E.	6.65 a	4.80 cd	13.95 d	8.46 cde
Pr.S+Po.E.	3.48 e	4.67 de	7.81 e	5.32 f
Pr.E.+Po.E.	3.39 e	5.70 b	17.31 c	8.82 cd
Pre.S.+Pre.E.+Po.E.	4.93 c	4.60 e	17.48 c	9.00 c
Manuel hoeing	0	0	0	0
Means	4.40 b	4.51 b	14.45 a	

*, Significant at p=0.05, **: Significant at p=0.01. Mean values with the same letter are not statistically significant different from each other according to the Duncan test at P ≤0.05

At the harvest of the black cumin, the highest weed biomass yield was determined in the plots treated with pre-sowing+pre-emergent herbicide application in the first year. The treatments of pre-sowing+post-emergent herbicide application and pre-emergent+post emergent herbicide application resulted in significantly lower weed biomass yield than the other herbicide treatment. Application of pre-sowing herbicide did not provide any weed control because averaged value of weed biomass yield for this

herbicide treatment was not statistically significant different than the control treatment with without weed control. In the second year, the most effective weed control treatment among the herbicide treatments was the application of pre-sowing+pre-emergent+ post emergent herbicide no weed biomass was determined in the manually hoed plots. It was reported that different climate conditions had affected herbicide activity and availability at lower doses, by Kudsk and Kristenden (1992).

Morphological properties, seed yield and oil ratio

In the years of 2016, 2017 and 2018, mean values of plant height, number of branches and capsules and seed yield in black cumin grown under different weed control treatment are shown in Table 6.

In the first year experiment, averaged value of plant height in the hand hoed plots was significantly higher than those in the plots treated with other weed control treatments. Application of pre-sowing+pre-emergent+post emergent herbicides resulted in a plant height being significantly higher than the control treatment and other herbicide treatments with the exception of the treatment of pre-sowing+post emergent herbicides. In the second year, the plants in the manually hoed plots showed significantly higher averaged value of plant height than the other weed control treatments. In the third year, the averaged plant heights in the herbicide treated plots with the exceptions of the applications of alone pre-emergence, pre-sowing herbicide +pre-emergent herbicide and pre-emergent +post emergent herbicides were not statistically significant different from that in the manually hoed plots. Plant height values determined in the study are compatible with those reported by Kızıl et al. (2008) and Tunçturk et al. (2005).

The effects of the treatments on the number of branches of black cumin were statistically significant in the first and third years, but not in the second year. In the first year, the treatment of manually hoeing gave significantly higher branch number per plant than all of the other treatments with the exception of the treatment of pre-emergent+ post emergent herbicides application. In the third year, the treatment of manually hand hoeing gave significantly higher branch number than all of the other weed control treatments with the exception of the treatment of pre-sowing+pre-emergent+post-emergent herbicide application. The results of the study showed that black cumin could not compete with weeds in plots which had dense weed population and therefore the number of branches decreased. It was stated that the number of branches in black cumin was significantly affected by weed competition periods (Hussain et al., 2009). The values of branch number per plant determined in the study are compatible with those reported by Ozel et al. (2002 and 2009).

Analyses of the data related to the number of capsules per plant showed that the weed control treatments significantly affected the mentioned characteristic of black cumin in all of the experimental years (Table 3). Manuel

hoeing resulted in significantly higher capsule number per plant than all of the other weed control treatments in all of the experimental years. In the first year, all of the herbicide treatments with the exception of the treatment of pre-sowing herbicide application gave statistically significant higher capsule number per plant than the control treatment. In the second year, alone applications of pre-sowing and pre-emergent herbicides were not statistically significant different from control treatment while post-emergent herbicide application resulted in significantly higher capsule number per plant than the control treatment. In the third year, application of post emergent herbicide and combined applications of pre sowing+pre-emergent herbicides, pre-emergent+post-emergent herbicides and pre-sowing+pre-emergent+post-emergent herbicides resulted in higher capsule number per plant than control treatment.

In all of the experimental years, manually hoeing treatment gave higher seed yield than all of the other weed control treatments. In the first and second year, combined application of pre-sowing+pre-emergent+post-emergent herbicides resulted in significantly higher seed yield than the other herbicide treatment as well as control treatment while combined application of pre-sowing+post-emergent herbicides gave higher seed yield than the other herbicide treatment and control treatment in the third year. Seed yield negatively effected with weed biomass, because lowest one obtained from high biomass weight according to mean of years. Meena et al. (2014) have determined that the application of herbicides before emergence and removal of weeds by hand 45 days after planting is the best way both economically and in terms of high yields. Hussain et al. (2009) stated that weed control in black cumin provided 69.41% increase in seed yield and weed competition caused significant decreases in seed yield. Nadeem et al. (2013) stated that, maximum seed yield was obtained from the plots where weeds were removed, there was a linear decrease depending on weed competition duration, it was necessary to control in 40 days after emergence which is critic stage to avoid degradation in black cumin. Besides, the herbicide treatments had a negative impact on photosynthesis (Saladin et al, 2003). Seed yields obtained in this study were higher than those reported by D'antuono et al (2002) and Ozel et al (2002). Weed competition for total growing season reduced the grain yield of black seed by 87%, compared with entire weed-free treatment (Seyyedi et al., 2016). Grain yield losses due to uncontrolled weed growth, it had direct correlation with weed competition (Korav et al., 2018).

Table 6. Morphological properties and seed yield in black cumin grown under different weed control treatments in experiment years (Pr.S= pre-sowing, Pr.E=pre-emergence, Po.E= post-emergence).

Treatments	Plant Height (cm)				Number of Branches (per plant)			
	2016**	2017*	2018*	Mean of Years**	2016**	2017	2018*	Mean of Years**
Control	24.1 d	48.9e	49.0bc	40.7 d	2.1 c	2.6	2.9 bc	2.5 cd
Pr.Sowing	23.5 d	53.0d	57.8a	44.7 c	2.3 bc	2.8	2.9 bc	2.7 bcd
Pr.Emergence	24.4 d	55.3b	45.1c	41.6 d	2.4 bc	3.0	1.8 d	2.4 d
Post Emergence	25.2 d	54.5bc	58.2a	45.9 c	2.6 bc	3.2	2.4 cd	2.7 bcd
Pr.S+Pr.E	24.6 d	50.1e	50.4b	41.7 d	2.7 bc	3.1	2.7 bcd	2.8 bc
Pr.S+Po.E	28.6 bc	54.2bc	57.7a	46.8 bc	2.7 bc	3.1	2.6 bcd	2.8 bc
Pr.E+Po.E	28.0 c	53.1cd	52.3b	44.5 c	2.9 ab	3.0	2.3 cd	2.7 bcd
Pr.S+Pr.E+Po.E	31.1 b	54.7bc	61.2a	49.0 b	2.8 b	2.9	3.4 ab	3.0 b
Manual hoeing	44.0 a	57.7a	60.5a	54.1 a	3.5 a	3.0	3.8 a	3.4 a
Mean	28.2 b	53.5 a	54.7 a		2.6 b	2.9 a	2.7 b	
	Number of Capsules (per plant)				Seed Yield (kg ha ⁻¹)			
	2016**	2017*	2018*	Mean of Years**	2016**	2017*	2018*	Mean of Years**
Control	5.5 g	5.1 d	1.9 c	4.2 f	13.7 g	94.0 g	59.3 d	77.7 d
Pr.Sowing	6.9 fg	6.5 cd	2.3 c	5.2 e	35.3 f	244.0 f	146.5 c	141.9 cd
Pr.Emergence	7.6 ef	7.2 bcd	2.3 c	5.7 e	34.3 f	237.0 f	154.0 c	142.1 cd
Post Emergence	10.0 cd	9.4 b	3.2 b	7.4 c	62.8 d	332.0 cd	163.6 c	186.1 bc
Pr.S+Pr.E	8.8 de	6.7 cd	3.4 b	6.3 d	50.5 e	267.0 ef	140.3 c	152.6 c
Pr.S+Po.E	10.9 c	8.3 bc	2.6 c	7.3 c	143.5 c	368.0 c	279.7 b	263.7 b
Pr.E+Po.E	10.4 cd	7.9 bc	3.2 b	7.1 c	69.8 d	300.0 de	125.8 c	165.2 c
Pr.S+Pr.E+Po.E	14.3 b	7.8 bc	3.7 b	8.6 b	199.0 b	435.0 b	174.0 c	269.3 b
Manual hoeing	26.9 a	15.8 a	7.4 a	16.7 a	767.6 a	1013.0 a	666.7 a	915.8 a
Mean	11.2 a	8.3 b	3.3 c		153.1 c	398.9 a	212.2 b	

*: Significant at $p=0.05$, **: Significant at $p=0.01$. Mean values with the same letter are not statistically significant different from each other according to the Duncan test at $P \leq 0.05$

The effects of treatments on thousand seed weight was only found significant in second year (Table 7). In that year, highest thousand seed weight (2.65 g) was obtained from the treatment of pre-emergent herbicide application. However, the values of thousand seed weight for the treatments of pre-sowing herbicide applications, pre-sowing+post-emergent herbicide application, pre-emergent+pot-emergent herbicide applications as well as manually hoeing were not statistically significant different from that of pre-emergent herbicide application. Hussain et al. (2009) have also found that thousand grain weight was significantly influenced by weed competition period. The values of thousand grain weight determined in this study results were compatible with those reported by Tuncturk et al. (2005) and Ozel et al. (2009) while the values were lower than determined by Ozel et al. (2002) and Abdolrahimi et al. (2012) results. Differences in thousand seed weight of black cumin in different researches can be due to differences in genotype, ecological conditions and growing technique among the researches.

The effect of weed control treatments on fatty oil content of black cumin seeds was found statistically significant different in all years (Table 7). Fatty oil content of seeds was changed between 28.5% and 36.2% in first year, 32.7% and 36.1% in second year and 26.7% and

36.5% in third year (Table 7). In the first year, seeds from the plots with the treatment of pre-sowing+pre-emergent herbicide application showed significantly lower oil content than all of the other weed control treatment with the exception of control treatment. In the second year, treatments of pre-emergent herbicide application and pre-sowing+pre-emergent herbicide application resulted in significantly lower oil content than the other weed control treatments with the exception of control treatment. In the third year, the treatments of pre-sowing+pre-emergent herbicide application and pre-emergent+post emergent herbicide application caused significantly lower oil content than the other herbicide treatments. Hussain et al. (2009) emphasized that, oil content of seeds in black cumin was decreased with weed competition and this situation may be resulted from the competition between crop plant and weed for environmental conditions and nutrient elements, thus the crop had lower photosynthesis activity and reduced oil content. Gutiérrez Prieto and Kırıcı (2020) stated that oil content of black cumin seeds which had been changed between 32.0% and 36.33% in Cukurova. Kalcın (2003) reported that oil content of *Nigella sativa* and *Nigella damascena* L had been changed between 28.08% and 34.29% in Ankara conditions.

Table 7. Thousand seed weight and fatty oil ratio (%) in black cumin grown under different weed control treatments in experiment years (Pr.S= pre-sowing, Pr.E=pre-emergence, Po.E= post-emergence).

Treatments	Thousand Seed Weight (g)				Fatty Oil Ratio (%)			
	2016	2017**	2018	Mean of Years**	2016*	2017*	2018*	Mean of Years*
Control	2.08	2.32 d	1.67	2.02 d	28.5 c	32.6 c	26.7 c	29.3 c
Pr.Sowing	2.38	2.58 ab	2.17	2.37 abc	36.2 a	36.1 a	36.5 a	36.3 a
Pr.Emergence	2.40	2.65 a	2.12	2.39 ab	34.8 a	32.3 c	36.0 a	34.4 a
Post Emergence	2.31	2.49 bc	2.17	2.43 a	35.3 a	34.2 b	34.6 a	34.7 a
Pr.S+Pr.E	2.19	2.41 c	2.00	2.20 c	32.6 b	32.7 c	29.5 b	31.6 b
Pr.S+Po.E	2.37	2.54 ab	2.25	2.39 ab	34.8 a	35.5 a	35.3 a	35.2 a
Pr.E+Po.E	2.23	2.64 a	1.75	2.21 c	35.4 a	36.0 a	31.9 b	34.4 a
Pr.S+Pr.E+Po.E	2.30	2.49 bc	2.00	2.24 bc	35.3 a	35.6 a	34.9 a	35.3 a
Manual hoeing	2.35	2.52 abc	2.08	2.29 abc	35.7 a	35.8 a	34.0 ab	35.2 a
Mean	2.28 b	2.54 a	2.0 c		34.3 a	34.5 a	33.3 b	

*: Significant at p=0.05, **: Significant at p=0.01. Mean values with the same letter are not statistically significant different from each other according to the Duncan test at P ≤ 0.05

Proline and Malondialdehyde (MDA) Content Result

Proline contents of the plants of black cumin in different plots treated with different weed control applications are given in the Table 8. Proline content of the black cumin plants ranged from 28.696 to 116.850 mM depending on the weed control treatment. The lowest proline content was determined in the plants grown with without weed control while highest proline content was determined in the plants grown with the post-emergent herbicide to control weeds. The plants in the plots with manually hoeing showed the proline content not so much different from those grown in the control plots with without weed control. Proline is a kind of amino acid, of which content is important to determine the stress condition and takes part to stabilise cell homeostasis in stressful conditions (Szabados and Savoure, 2010). In the studies on black cumin, the proline content of control application was found between 0-20 $\mu\text{M g}^{-1}$ (Fazeli et al., 2018; Fallah et al., 2018). Similarly, proline content of herbicide treated soyabean plants was markedly

increased in comparison with that of untreated plants (Fayez, 2000).

MDA contents of the leaves of black cumin grown with different weed control treatments are given in the Table 8. The values of MDA content changed between 0.46 nmol.g^{-1} and 1.14 nmol.g^{-1} depending on the weed control treatment. The lowest value of MDA was determined in the leaves of the plants grown with without weed control while highest value of MDA content was determined in the leaves of the plants grown with the treatment of pre-sowing+pre-emergent+post emergent herbicide application. MDA is a result of lipid peroxidation and its elevation indicates stress-induced cell destruction (Buege and Aust, 1978). The MDA contents of control plants were recorded between 0.1-0.2 nmol.g^{-1} (Rozita et al., 2012; Fazeli et al., 2018). In our study, MDA content of control plants was found lower than those treated with different weed control applications.

Table 8. The Proline and MDA content of black cumin plants grown with different weed control treatment in the year of 2018 (Pr.S= pre-sowing, Pr.E=pre-emergence, Po.E= post-emergence).

Treatments	Proline (mM)		Malondialdehyde (nmol.g ⁻¹)	
	Average	Standard Dev.	Average	Standard Dev.
Control	22.820	7.50	0.46	0.11
Pre-emergence	37.211	20.81	0.59	0.15
Post-emergence	116.856	27.26	0.49	0.02
Pr.S+Pr.E	29.996	12.88	0.86	0.54
Pr.S+Po.E	38.784	8.03	0.67	0.00
Pr.E+Po.E	55.307	19.70	1.02	0.25
Pr.S+Pr.E+Po.E	34.364	8.69	1.14	0.39
Manual hoeing	28.696	9.85	0.60	0.41

CONCLUSION

Weeds compete with the crops and affect negatively the yield and quality of the crop. Therefore, weeds must be controlled with proper weed control method. Chemical control or control with herbicides is a widely applied weed control method. It is an important factor to select suitable herbicide and dose in accordance with dominated weeds for controlling them (Kudsk, 2008), and also soil humidity at

application time and irrigation after weed control are important factors for herbicide activities. Effective minimum dose applications' activity can be optimized by considering climate conditions and weed species' sensitivity, growing high competitive crops. Optimum sowing density can be controlled weed biomass and needed further studies (Izquierdo et al., 2003; Rastegar et al., 2018). Thus, an environmentally friendly and economical

weed control system can be constituted. At the end of study, result of this study showed that herbicides used to control up weeds was not effective to control weeds, because of weeds' high density at trial period, so the development of black cumin was influenced negatively.

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