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INFLUENCE OF SEED RATE AND FERTILIZATION ON YIELD AND YIELD COMPONENTS OF *Nigella sativa* L. CULTIVATED UNDER MEDITERRANEAN SEMI-ARID CONDITIONS

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Abstract

A field experiment was conducted to determine the effect of seed rate and fertilization on yield and yield components of Nigella sativa crop. The experiment was laid out according to a split-plot design with three replicates, two main plots (seed rates: 50 kg ha^{-1} and 60 kg ha^{-1}) and four sub-plots (fertilization treatments: untreated, compost, sheep manure, inorganic fertilizer). Plants were higher in plots sown at a rate of 60 kg ha^{-1} (18.2-22.7 cm). The highest number of capsules per plant (5.0-5.8) were found in sub-plots subjected to inorganic fertilization. Moreover, there were significant differences between fertilization treatment regarding seed yield and biological yield. The highest seed yield (911-1066 kg ha^{-1}) and biological yield (3864-4063 kg ha^{-1}) were found in inorganic treatments. The number of branches per plant, number of seeds per capsule, thousand-seed weight, and Harvest Index was not affected neither by seed rate nor by fertilization. Finally, there was not any significant interaction between seed rate and fertilization.

Key words: *compost, inorganic fertilizer, Nigella sativa, seed rates, yield components.*

INTRODUCTION

Medicinal plants are used for treating many disorders that are either non-curable or rarely cured by modern systems of medicine (Abadi et al., 2015). Over the last three decades, the use of herbal medicinal products and supplements have grown to such an extent that about 80% of the world population relying on them for some part of primary healthcare. (Ekor, 2014). *Nigella sativa* L. is an annual medicinal plant belonging to the Ranunculaceae family. It is native to southern Europe, North Africa, South and West Asia (Tuncturk et al., 2012) and widely cultivated throughout Syria, Egypt, Saudi Arabia, Iran, Pakistan, India and Turkeyfor seed yield and oil production (Riaz et al., 1996).

The height of plant ranges from 20 to 30 cm and its leaves are finely divided and linear with grayish-green color. The flowers are of pale blue and white color with 5 to 10 pedals (2-2.5 cm). The fruit is an inflated capsule composed of 3 to 7 united follicles. Each capsule contains a various number of dark grey or black small sized (1-5 mg) seeds (Valadabadi, 2013).

Seeds of *N. sativa* are characterized for their unique chemical properties that may contribute to the improvement of human health. Various surveys have shown that the whole seeds or their extracts have diuretic, antihypertensive, antidiabetic, anticancer, immunomodulatory, anthelmintic, analgesic, antimicrobial, anti-inflammatory, spasmolytic, bronchodilator, hepatoprotective, gastroprotective, nephron-protective, antihypertensive and antioxidant effects (Al-Jassir, 1992; Riaz et al., 1996; Ahmad et al., 2013). Moreover, the seeds are rich in fats, fiber, minerals such as Fe, Na, Cu, Zn, P, Ca and vitamins such as ascorbic acid, thiamin, niacin, pyridoxine, and folic acid (Takruri and Dameh, 1998). *N. sativa* seeds contain 30-35% oil and 0.5-1.5% essential oil which have several uses for pharmaceutical and food industries (Üstun et al., 1990; Ashraf et al., 2006). One of the most important constituents of essential oil is thymoquinone that belongs to the chemical class of terpenoids and imports the plant under investigation about the ability to influence on important human diseases such as cancer (Banerjee et al., 2010) or the metabolic syndrome (Razavi and Hosseinzadeh, 2014).

The effective use of *N. sativa* for different therapeutic purposes is highly dependent on the yield and quality (Datta et al., 2012). Seed rate is one of the main key factors for obtaining high yield and quality in the production of crops. Several studies carried out in countries where systematically cultivated, have demonstrated that suitable seed rate can increase the growth and yield of *N. sativa* (Toncer and Kizil, 2004; Tuncturk et al., 2004). Talafih et al. (2007) reported that under Mediterranean semi-arid environment the highest seed yield (801.2 kg ha^{-1}) was obtain under 35 kg seed ha^{-1}.

The appropriate use of fertilizers increases the growth and quality of the medicinal plants (Mohamed et al., 2014). Many experiments have been conducted to investigate the effect of different amounts of nitrogen (Ashraf et al., 2006; Tuncturk et al., 2012) and phosphate (Kizil et al., 2008) fertilizers on different agronomic characteristics, yield and yield components in *N. sativa*. According to Rana et al. (2012), the maximum values of agronomic characteristic such as plant height and number of branches and the highest yield of seed were observed at a ratio of 60:120 kg NP ha^{-1}.

Because *N. sativa* is a plant that has been systematically studied only during last two decades, the available literature on plant growth and yield under organic fertilization is still quite limited. Data by other researchers (Efthimiadou et al., 2009; Bilalis et al., 2012) clearly proved the beneficial effects of organic fertilization on the yields ofs everal crops.

Limited data are available regarding the performance of *N. sativa* growth under Mediterranean semi-arid conditions and organic cropping system. Therefore, the aim of this study was to determine the effects of seed rate and organic and inorganic fertilization on growth and yield parameters of nigella crop.

MATERIALS AND METHODS

A field experiment was carried out in the organic experimental field of the Agricultural University of Athens (Latitude: 37°59′ 1.70″ N, Longitude: 23°42′ 7.04″ E, Altitude: 29 m above sea level) from April to July 2016. The soil was clay loam (29.8% clay, 34.3% silt and 35.9% sand) (Bouyoucos, 1962) with pH (1:1 H_2O) 7.29, nitrate-nitrogen (NO_3-N) 12.4 mg kg^{-1} soil, available phosphorus (P) 13.2 mg kg^{-1} soil, available potassium (K) 201 mg kg^{-1} soil, 15.99% $CaCO_3$ and 1.47% organic matter (Wakley and Black, 1934). The site was managed according to organic agricultural guidelines (EC 834/2007). Meteorological data (mean monthly air temperature and precipitation) during the experimental period as recorded by the weather station of Agricultural University of Athens are presented in Figure 1. The mean temperature during the crop cycle (April-July) was higher (24.5°C) than that observed in 35-year average (22.7°C). Accumulated precipitation of cultivation period was 26.8 mm, which means it was approximately two-thirds lower than 35-year average (62.1 mm).

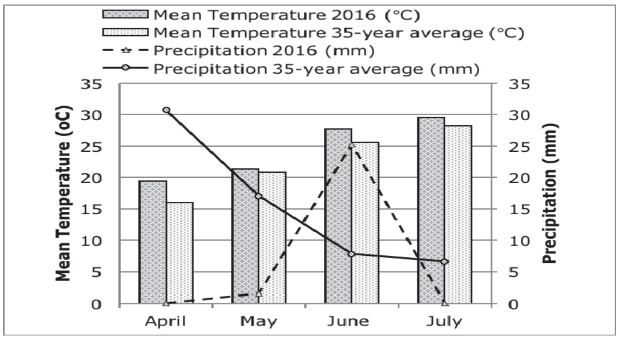

Figure 1. Meteorological data (mean monthly air temperature and precipitation) during the experimental period (April-July, 2016) and the 35-year average (1981-2015) in Athens, Greece

The experiment was set up on an area of 302 m^2 according to the split-plot design with three replicates, two main plots (seed rates: 50 kg ha^{-1} and 60 kg ha^{-1}) and four sub-plots [fertilization treatments: control (untreated), compost (2000 kg/ha), sheep manure (2750 kg/ha), inorganic fertilizer (15-15-15+5 S, 400 kg/ha)]. The main and sub-plot sizes were 42.25 m^2 (6.5 m X 6.5 m) and 9 m^2 (3 m X 3 m), respectively. There was a space of 0.5 m between sub-plots, 1 m between plots and 1 m between replications. Soil was prepared by ploughing at a depth of about 0.25 m. Fertilizers were applied by hand on the soil surface and then harrowed. *N. sativa* seeds were sown on 19th April by hand in rows 30 cm apart, at a depth of 1 cm. Drip irrigation system was installed in experimental plots. The first irrigation was applied immediately after sowing and the second irrigation was done two days later for uniform emergence. After that, plots were irrigated to field capacity every 3-4 days until the 10th day before harvest. The total quantity of water applied during the experiment was 735.2 mm. Throughout the experimental period, there was not observed any incidence of pest or disease on *N. sativa* crop. Weeds were controlled by hand hoeing when it was necessary. Finally, harvesting was done manually on 30th July 2016 after the stage of full maturity. Data were recorded on some growth and yield parameters including plant weight, number of branches and number of capsules per plant, number of seeds per capsule and thousand seed weight using five randomly selected plants from each sub-plot. Moreover, seed yield, biological yield and harvest index were assessed. For the determination of these parameters, the middle sub-plot area of 4 m^2 (2 m X 2 m) was used.

Plant weight was measured in centimeters at the stage of physiological maturity from the ground level to the tip of plant. Number of branches per plant was computed by counting the number of primary branches of five middle plants of the four middle rows. After that, the number of capsule per plant and number of seed per capsule on individual plant and capsule basis, respectively, were measured. For the computation of thousand seed weight, the seeds obtained from each of the ten selected plants were counted, weighed by analytical balance and expressed in grams. Biological yield was recorded by measuring the whole weight of plants from middle sub-plot areas after harvesting and drying for three days. Seed yield was determined by measuring the seed weight of plants used for biological yield. Harvest Index was calculated by divining seed yield by the biological yield.

The experimental data were checked for normality and subjected to statistical analysis according to the split-plot design. The statistical analysis was performed with JMP 9statistical software (SAS Institute Inc., Cary, NC, USA). The differences between means were compared using Least Significance Difference (LSD) test. All comparisons were made at the 5% level of significance.

RESULTS AND DISCUSSIONS

Results of variance analysis showed that the plant weight was significantly ($p<0.05$) affected by the different seed rates. In particular, the tallest plant (20.0 cm) recorded at the high seed rate (60 kg ha^{-1}), while, the lowest values (18.1 cm) were obtained from the low seed rate (50 kg ha^{-1}). These data are in accordance with previous studies reporting that increasing seed rates noticeably increased plant weight but with values lower than those of literature (Toncer and Kizil, 2004; Talafih et al., 2007).

These differences between our results and other studies could be explained by different variations in environmental conditions and the genetic potential of populations. Fertilization had not significant effect on plant weight. Despite the differences among fertilization treatments were not important, generally inorganic fertilizer gave slightly higher plant height (20.8 cm) and the minimum height obtained from control treatment (17.2 cm). Concerning number of branches per plant, the effect of different seed rates was found not to be statistically significant for the high and low seed rates. However, it was observed that the highest value (3.7) was obtained in the case of the low seed rate. This can be explained by the interplant competition in higher plant density which could induce plant growth reduction and produced the highest number of plants. Similar results were also reported by Tunctur et al. (2005). Fertilization treatments showed higher values than control in number of branches per plant but values were not statistically significant.

During this study, the highest number of branches per plant was achieved in inorganic fertilization treatment. Ali et al. (2015) mentioned that the number of primary branches per plant was significantly increased in the case of high nitrogen treatment, whereas number of secondary and tertiary branches per plant were not influenced.

The number of capsules per plant was only affected by different fertilization treatments. The highest value (5.4) was observed in inorganic fertilization treatment and the lowest was obtained in the untreated plots. Similar results concerning the positive response of nigella crop to inorganic fertilization were also recorded by other researchers (Rana et al., 2012; Khalid and Shedeed, 2015; Yiman et al., 2015).

Table 1. Plant height, number of branches per plant and number of capsules per plant and number of seeds per capsule of *Nigella sativa* as affected by different seed rates and fertilization

Fertilization	Seed Rate							
	Plant height (cm)		Number of branches per plant		Number of capsules per plant		Number of seeds per capsule	
	50 kg ha^{-1}	60 kg ha^{-1}	50 kg ha^{-1}	60 kg ha^{-1}	50 kg ha^{-1}	60 kg ha^{-1}	50 kg ha^{-1}	60 kg ha^{-1}
Control	16.2	18.2	3.0	2.2	3.0	1.7	37.0	36.6
Compost	18.7	19.4	3.5	3.3	5.4	4.3	42.5	40.8
Manure	18.5	19.5	3.7	2.3	4.7	4.6	41.6	37.9
Inorganic	18.9	22.7	4.6	2.7	5.8	5.0	42.6	42.0
$F_{seed\ rate}$	19.2417* ($LSD_{5\%}$= 0.4274)		5.4516^{ns}		0.7557^{ns}		1.0852^{ns}	
$F_{fertilization}$	1.7297^{ns}		1.8958^{ns}		4.8728* ($LSD_{5\%}$= 0.8672)		1.8025^{ns}	
$F_{seed\ rate\ X\ fertilization}$	0.3881^{ns}		1.2292^{ns}		0.9020^{ns}		0.1703^{ns}	

F-test ratios are from ANOVA. Significant at *p=0.05, **p=0.01, ***p=0.001, ns: not significant The LSD (p=0.05) for seed rate and fertilization are also presented.

Regarding number of seeds per capsule, the results have revealed that different seed rates had no significant effect on this trait. The results of number of seeds in capsule were lower than Toncer and Kizil (2004) demonstrated. Moreover, fertilization treatments resulted in values not statistically higher than control. However, the maximum number of seeds per capsule (42.3) was recorded after application of inorganic fertilizer. In addition, Hadi et al. (2015) found that application of manure in *N. sativa* crop did not affect the seed number per capsule.

Thousand-seed weight was not influenced neither by seed rate nor by fertilization. Despite the seed rates had no significant effect on thousand seed weight the high seed rate produced higher results (2.70 g). Talafih et al. (2007) supported that the increase in plant density resulted in an increase in the thousand-seed weight that could be related to the competition between plants on available soil moisture, nutrients and light. This result is in agreement with Ali et al. (2015) and Yimam et al. (2015) who found that the increase in levels of inorganic fertilizer did not affect the thousand-seed weight.

The results of experiment indicated that the effect of different seed rates on seed yield was insignificant. Despite that, the study data showed that the application of high seed rate led to declines in seed yield. Toncer and Kizil (2004) and Tuncturk et al. (2005) explained that increasing seed rate over 20 kg ha^{-1}, plant population density increased, which might cause higher interplant competition, thus reducing seed yield. Fertilization effect on this trait was statistically significant. The highest value (988 kg ha^{-1}) was recorded from inorganic treatment. This result is in agreement with Tuncturk et al. (2012) reported that yield components such as branches and capsules affects seed yield. In this study as mentioned above, the highest values of number of branches and capsules noted in inorganic treatment. Moreover, Yimam (2015) stated the maximum seed yield (1337 kg ha^{-1}) obtained from application of 60:40 kg NP ha^{-1} and Seyyedi et al. (2015) reported than increase in amount of phosphorus might significantly increase seed and biological yield.

The biological yield was only influenced by fertilization. The maximum yield (3963 kg ha^{-1}) was achieved in the inorganic treatment followed by compost (3347 kg ha^{-1}) and manure (3263 kg ha^{-1}). Biological yield was actually affected by the combined supply of nitrogen and phosphorus.

The increase in these nutrients might be resulted in a more vigorous plant growth with greater plant height, number of branches, number of leaves and number of capsules producing a greater total plant biomass, thereby resulting in higher biological yield (Rana et al., 2012; Ali et al., 2015). This result is in full accordance with Yimam et al. (2015) reported that the highest biological yield was observed after application of 60 and 40 kg of N and P ha^{-1}, respectively.

The results obtained from variance analysis indicated that Harvest Index was not affected by different seed rates and fertilization.

Table 2. Thousand seed weight, seed yield, biological yield and Harvest Index of *Nigella sativa* crop as affected by different seed rates and fertilization

Fertilization	Seed Rate							
	Thousand-seed weight (g)		Seed yield (kg ha^{-1})		Biological yield (kg ha^{-1})		Harvest Index (%)	
	50 kg ha^{-1}	60 kg ha^{-1}	50 kg ha^{-1}	60 kg ha^{-1}	50 kg ha^{-1}	60 kg ha^{-1}	50 kg ha^{-1}	60 kg ha^{-1}
Control	2.65	2.50	601	497	2471	2387	24.53	20.83
Compost	2.67	2.73	881	828	3455	3239.	25.50	25.57
Manure	2.58	2.79	820	773	3279	3247	25.00	23.81
Inorganic	2.68	2.76	1066	911	4063	3864	26.23	23.57
$F_{seed\ rate}$	0.1555^{ns}		3.5316^{ns}		0.5280^{ns}		3.9431^{ns}	
$F_{fertilization}$	1.1637^{ns}		6.7781** ($LSD_{5\%}$= 100.57)		9.3441*** ($LSD_{5\%}$= 291.73)		1.2908^{ns}	
$F_{seed\ rate\ X\ fertilization}$	1.5043^{ns}		0.3414		0.0465^{ns}		1.1118^{ns}	

F-test ratios are from ANOVA. Significant at *p=0.05, **p=0.01, ***p=0.001, ns: not significant The LSD (p=0.05) for seed rate and fertilization are also presented.

Despite the no significant effect of fertilization, the highest value (25.54%) was found in compost treatment followed by inorganic (24.90%) and manure (24.41%) treatments. Harvest Index values in our study were higher than Yimam et al. (2015) and lower than Rana et al. (2012) who studied the influence of various nitrogen and phosphorus rates in *N. sativa*.

CONCLUSIONS

The results of the present study indicate that nigella crop was not affected neither by seed rate nor by fertilization. The highest plant height was found under 60 kg ha^{-1} seed rate. Moreover, it was observed that plants received the inorganic fertilizer had higher number of capsules per plant which is a key yield component. Finally, there were significant differences between fertilization treatments regarding the seed yield and biological yield with highest values found under inorganic treatment.

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