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The Performance of Two Species of Black Cumin (*Nigella sativa* L.) and (*Nigella arvensis* L.) Under Different Sowing Dates in Spring and Autumn at hallabja Governorate /Iraqi Kurdistan Region.

Adel M. A. Al-Zubaidy,

Bakhtiar S.Ghafoor,

Horticulture and Landscape Design Department, Technical College of Applied Sciences, Sulaiani Polytechnic University, Halabja, Kurdistan Region, Iraq.

Horticulture and Landscape Design Department, Technical College of Applied Sciences, Sulaiani Polytechnic University, Halabja, Kurdistan Region, Iraq.

Abdulsalam A. Rasul

University of Sulaimani, College of Agricultural Sciences

adel.adday@spu.edu.iq

Bakhtiarsaadey@gmail.com

abdulsalam.rasul@, nivsul.edu.iq

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Abstract

Two field experiments were performed to study the response of two species, Nigella sativa L. and Nigella arvensis L. within different sowing dates at spring and autumn seasons which included 1st and 20th March, 10th April and 1st May for spring season, while the sowing dates for the autumn season were, 2nd November, 21st November, 11th December, 31stDecember and 20thJanuary. Both experiments conducted according to the Completely Randomized Block Design (CRBD) within three replications at hallabja/Kurdistan Region, located [35°12'48.7"N; 45°57'34.4"E] and the altitude was 596 masl. Results showed that among the four different sowing dates of spring cultivations, both species were responded to only 1st and 20th March date with some superiority of the 1st March, otherwise the survival and their yield were not acceptable at sowing date later than 20th March, while at the autumn cultivation, both species responded to all sowing date, but not to be later than 31st December, the early dates 2nd November and 21st November were showed significant differences in most yield traits. The result of the study showed that, due to the favorable environmental condition and the longer period of growth at autumn cultivation, most of the yield traits were gained significant values as the average of both species and different sowing dates of autumn compared to that occurred at spring cultivations. Some chemical contents of both species under the different environment conditions showed significant differences. Generally, N. sativa compared to N. arvensis contained more significant chemical compounds in the term of carbohydrates and protein at autumn, fixed and volatile oils at spring.

Key words: Chemical contents, Nigella sativa, Nigella arvensis, Sowing dates, Yield components.



1. Introduction

Plants are considered to be one of the natural bases for the production of bioactive compounds, many of which are used to support health and fight against pathological conditions and many of them are marketed as food or herbal medicines [1]. Wild resources of medicinal plants are decreasing, but demand is increasing. Thus cultivation is the only solution to fill this gap, and to initiate systematic cultivation of medicinal and aromatic plants, many factors have to be selected [2,3]. The genus Nigella belongs to the family Ranunculaceae, many plants of this family have remarkable aromatic properties and medicinal values [4]. The important species, which belong to the Nigella are the common black cumin (Nigella sativa L.), damascus black cumin (Nigella damascena L.), oriental black cumin (Nigella orientalis L.) and field black cumin (Nigella arvensis L.) [5]. Nigella sativa L. cultivated widely in the mountains and also moisten lands of Mousl, while Nigella arvensis L. wildly grown in Mosul, Erbil, Sulaimani and west desert of Iraq, and there are other species such as Nigella deserti, Nigella oxypetala and Nigella assyriaca, described by [6]. Black cumin is intensively used as uncrushed in bakery products (bread, muffins, biscuits, etc.) and some cheese (brynza, cottage cheese, etc.), their seeds contain protein, alkaloids (nigellicines and nigelledine), saponin (α - hederin) fixed and essential oil [7]. The fixed and essential oil of black cumin contains various bioactive molecules such as thymoquinone, thymol, tocopherol, trans-retinol and selenium; these diversities are varied between different species especially between N. sativa and N. arvensis [8]. In various studies the black seeds have been shown to possess antioxidant, anti-inflammatory, anticancer, analgesic, antimicrobial activities and skin disease, cough, diuretic, digestive disorders, carminative, and immune-potentiating. Constant inhalation of fried seeds releases cold and catarrh. The seeds have also been used in mercury poisoning, sores and leprosy [9]. However, there are very few studies about adaptation and quality in our country specifically in hallabja province. The local population is generally used as seed production and breeding materials by breeders and farmers due to the lack of cultivars. The seeds of Nigella sativa, Nigella damascene and Nigella arvensis are used in folk medicine and as a spice and only the species of black cumin (Nigella sativa L.) are traded [10]. Genetic sources, climate and soil conditions are the main factors affecting the growth and development of plants and the physiological growth of the plants, being synthesized of the active substances, such as the amount of secondary metabolites. The effect of ecological factors on the yield and quality of medicinal plants is higher compared to the other plants. Because the quality in medicinal and aromatic plants is as important as yield, a certain quality cannot be cultivated even if they are with very high yield. Therefore, they should only be cultivated in convenient regions for the ecologies of these plants [11]. Successful production of medicinal and aromatic plants is under diverse physiological, biochemical metabolic and genetic regulation and can be manipulated by an alteration in the growing conditions and management of agricultural practices. Therefore, there is a need to investigate different agroclimate for the cultivation of a medicinal plant. Between the choices is selecting a successful species to be grown under suitable soil and environmental climate, which can be expressed in the term of different sowing dates, which can lead to gain this goal [12,13,14,15]. In hallabja province as well as in Kurdistan Region, there is a lack of scientific researches and information on Black Cumin cultivation in terms of sowing dates and species. These aspects necessitated this study and have given rise to the concept of our project to evaluate the performance of two species under different environmental condition (autumn and spring) and

to determine the optimum sowing date and its effect on some growth characters, yield and its components, and some chemical compounds. So this study is conducted to determine the seed yield, some yield components and some chemical compounds of the two species of black cumin *N. sativa* and *N. arvensis* by different sowing dates in spring and autumn in hallabja province/Iraqi Kurdistan Region.

2. Material and Methods

Two field experiments were performed to study the effect of two species of the genus Nigella, namely; Nigella sativa and Nigella arvensis within different sowing dates at spring and autumn seasons which included 1st and 20th March, 10th April and 1st May for spring season, while the sowing dates for the autumn season were, 2nd and 21st November, 11th and 31stDecember and 20thJanuary. Within three replications at hallabja/Kurdistan Region, located [35°12'48.7"N; 45°57'34.4E] and the altitude was 596 masl. The rainfall during the spring cultivation season was 87.6mm that tends to use supplementary irrigations whenever it was needed, while the amount of the rainfall during the autumn cultivation season was 607.6 mm that was sufficient without irrigations. The soil of the location of the experiment can be described as a clay texture with 2% organic matter. The study included two factors, first was using two species of black cumin which their seeds were obtained from an Iranian company of seed registration (PAKAN BAZR) and the second factor of this study was four and five different sowing dates at spring and autumn season respectively. A factorial experiment 2×4 for the 1st experiment and a factorial experiment 2×5 for the 2nd experiment, both were conducted according to completely Randomized-Block Design within three replicates. The land of the study prepared for cultivation by tilling the field two times using mold broad plow and harrow. Seeds of both species of Nigella were cultivated by hand in the 1st March, 20th March 10th April and 1st May for the spring season experiment, the dates2nd November,21st November, 11th December, 31st December and 20th January were appointed for the autumn season experiment. The distance between the rows was 20 cm and within the plants was 20 cm also. Each plot area at the spring experiment was 1×1mand at autumn was 2×3 m and consisted of 10 rows. Seeds were sowed at 1-3 cm depths. In the spring season, the experiment was irrigated at any growing stage when it was needed as supplemental to the precipitations, while in the autumn season the crop was grown without irrigation and completely depended on precipitation. The nitrogen fertilization was applied only in one dose, 20 g for each 1×1m at the time of plant height when 10-20 cm. All the necessary cultural practices were similarly applied to the plots during vegetation periods in both seasons. Weeds were controlled by hand when needed. The crop was attended as per routine cultural practices at each season.

2.1. Seed yield and yield components characters: Five plants randomly taken from each plot for measurements of, No. of Flowers. plant⁻¹, No. of Capsules. plant⁻¹, No. of seeds capsules⁻¹, No. of seeds. plant⁻¹, Weight of seeds. plant⁻¹(g), 1000 seed weight (g) and Seed yield (g/m^2) .

2.2. Chemical components: Protein Content %: First the total nitrogen was determined using Macro-Kjeldahl apparatus [16]. And the protein was calculated from the formula: -

Protein% = $T.N\% \times 6.25$

Fixed oil % w/w: (10gm) of the harvested seeds used in each treatment combination which was powdered. Digital Soxhlet instrument used for oil distillation, with 150 ml Hexane

solvent, the instrument was adjusted at 50 $^{\circ}$, then oil content according to the method [17]. on the base of w/w calculated as follow:

Wt. of fixed oil = Wt. of the flask with oil - Wt. of the empty flask

Fixed oil % w/w =
$$\frac{\text{Wt. of the fixed oil}}{\text{Wt. of sample(gm)}} \times 100$$

Essential oil % v/w: The isolation of the essential oils from the 25g of powdered seeds was done by hydro distillation with Clevenger apparatus for 3h according to the method [18]. And the calculation was as follow:

Essential oil % v/w =
$$\frac{\text{Volume of essential oil in Clevenger (m1)}}{\text{Wt. of sample (25g)}} \times 100$$

Ash determination: The Ash determination by using Muffle Furnace in the 550C° the reference to method was [19].

Carbohydrates determination: The carbohydrates determination according to [20].

Statistical Analysis: Analysis of variance as a general test was done according to analysis of 2 factors in CRBD, and the means were tested according to least significant difference (LSD) using significant level of 0.05 and 0.01 confirmed by [21].

3. Results and Discussions

3.1. Spring Experiment

Table 1. Showed some yield traits of the two species of black seed grown in spring 2015. Data realized that *N. arvensis* showed significant values regarding some traits such as; No. of flowers/plant with 39.43, No. of capsules/plant with 23.67, No. of seeds/plant with 1031.65 compared to *N. sativa* that records only 8.08, 7.52 and 284.84 for the three mentioned traits respectively. Regarding to the traits of No. of seeds/plant *N. arvensis* significantly dominated *N. sativa*, while the seed weight/plant and seed yield g/m² also *N. arvensis* exceeded *N. sativa* but insignificantly, but oppositely the significant 1000 seed weight/plant (g) was recorded by *N. sativa* which was 2.64 g compared to 1.80 g in *N. arvensis* which mostly due to the longer duration of seed filling stage spend by *N. sativa* compared to the shorter time spend by *N. arvensis*.

Species	No.flower s. plant ⁻¹	No. of capsules. plant ⁻¹	No. of seeds. capsule	No. of seeds. plant	1000seed weight (g)	Seed weight. Plant (g)	Seed yield (g/m ²)
N. sativa	8.08	7.52	36.71	284.84	2.64	0.75	13.94
N. arvensis	39.43	23.67	40.78	1031.65	1.80	1.86	21.72
LSD (p≤0.05)	20.78	4.39	n.s	407.95	0.32	0.95	n.s
LSD (p≤0.01)	n.s	6.65	n.s	618.10	0.48	n.s	n.s

Table 1. Some yield and yield component traits of two species of black seed grown at spring 2015.

Regarding yield and some yield component of black seed in two different sowing dates at spring 2015, **Table 2.** Represents that except the seed yield which was dominant with 25.84 g/m^2 in the 1st March compared to 9.82 g/m^2 occurred in the 20th March, all other yield traits

were not significant. Recorded data of 1st March for some traits such as 44.67 seeds/capsule, 695.44seeds/plant, 2.35 g weight of 1000 seed and 1.42g weight of seeds/plant were insignificantly exceeded the 20th March date. Also, the 1st March date records 25.84 g/m² seed yield which was significantly surpassed 9.82 g/m²seed yield that gained on 20th March. Data in **Table 2.** Showed that only the No. of flowers/plant and the No. of capsules/plant were more on 20th March than the 1st March. Regarding the effect of the interactions on yield traits, the study results showed insignificant differences.

Table 2. Some yield and yield component traits of black seed grown within different sowing dates at spring2015.

Sowing Dates	No. of flowers/ plant ⁻¹	No. of capsules/ plant ⁻¹	No. of seeds/ capsule plant ⁻¹	No. of seeds/ plant ⁻¹	1000seed weight (g)	Seed weight/ Plant (g)	Seed yield (g/m²)
1 st March	23.08	14.07	44.67	695.44	2.35	1.42	25.84
20 th March	24.43	17.12	32.82	621.05	2.09	1.19	9.82
LSD (p≤0.05)	n.s	n.s	n.s	n.s	n.s	n.s	13.05
LSD (p≤0.01)	n.s	n.s	n.s	n.s	n.s	n.s	n.s

3.2. Autumn Experiment

The effect of the two species of black seed on some yield and yield component traits shown in Table 3. The two species of black seed showed different significant over all the yield and yield component traits. Similar pattern as in the growth characters had been observed, N. arvensis gave the upper significant limits compared to N. sativa in all yield and yield component traits except the 1000 seed weight (g) which records 2.42 g by N. sativa and it was significant to 1.67 g recorded by N. arvensis may be due to the longer seed filling duration in N. sativa compare to the N. arvensis. The N. arvensis gave the upper limits of the traits; No. of flowers/plant 104.88, No. of capsules/ plant 57.12, No. of seeds /capsule 54.55, ultimately increase the No. of seeds/plant which was 3142.00 seeds in compare to N. sativa which were 27.53, 25.61, 42.91 and 1256.39 for the mentioned traits respectively. These improvements may be due to the reflection of the relatively higher performance of vegetative growth in N. arvensis compared to N. sativa. The 5.24 g seed weight gained by N. arvensis was significant in comparison to the seed weigh/plant recorded by N. sativa which was only 3.00 g. Finally, the seed yield (g/m^2) was 106.73 g/m² obtained by *N. arvensis* and significantly predominated 68.31 g/m^2 seed yields recorded by N. sativa, this predominated seed yield of N. arvensis compare to the N. sativa.

Table 3. Some yield and yield component traits of two species of black seed grown at autumn 2015-2016.

Species	No. of flowers. plant	No. of capsules. Plant	No. of seeds. capsule	No. of seeds. plant	1000 seed weight (g)	Seed weight. plant	Seed yield (g/m²)
N. sativa	27.53	25.61	42.91	1256.39	2.42	3.00	68.31
N. arvensis	104.88	57.12	54.55	3142.00	1.67	5.24	106.73
LSD (p≤0.05)	13.14	8.24	5.12	432.97	0.11	0.79	23.49
LSD (p≤0.01)	18.00	11.28	7.01	593.20	0.15	1.08	32.18

Data in Table 4. Represent some yield and yield component traits of black seed grown within different sowing dates at autumn (2015-2016), as it is. D1 and D2 predominated significantly all other sowing dates. Regarding the No. of flowers, it was recorded the highest value 101.33 flowers/plant in D2 followed by 100.73 flowers/plant in D1 they were not significant between themselves, but they have predominated all other dates significantly, while the lowest value 35.17 flowers/plant recorded in the latest date D5. The 1st date D1 records higher significant value which was 64.07capsules/plant predominated significantly on all other dates even the D2. Although the No. of flowers/plant in D2 was more than in D1, but the No. of capsules/plant exactly took an opposite direction which showed more capsules/plant in D1 compared to D2. This difference confirmed that the fertility in D1 which was 63.60% exceeded the fertility in D2 which was 47.59%, [22]. Were also noticed that different sowing dates significantly affected the No. of umbrella plant⁻¹ in black cumin. As a result of higher fertility ratio in D1 the higher significant No. of seeds also recorded in D1, which were 59.53 seeds/capsule and 3822.87 seeds/plant and the lowest values were 37.80 seeds/capsule and 989.13 seeds/plant recorded in D5, good seed setting condition in early sowing as compared to late sowing was founded in research of [23]. Regarding the 1000 seed weight, D2 gained the highest value 2.20g, insignificantly followed by 2.14 g in D1, they were not significant between themselves, but they were significant compared to other dates, the minimum value was 1.94 equally occurred in D3 and D5. The highest significant value of seed weight/plant was 7.60 g and seed yield 184.43 g/m^2 were obtained in D1 compared to all other dates without exception. The minimum seed weights were 1.63 g/plant and 25.47 g/m² recorded in D5. It can be summarized that the early dates especially D1 followed by D2 play a great role in the enhancement of growth and yield production of black seed in the autumn period compared to later sowing dates. On the other hand, sowing at the winter period may be succeeded but because it companied low temperature, caused a weak growth of plants and ultimately decreased seed yield. This agrees with that founded by [24]. Regarding the effect of the interactions on yield traits, the study results showed insignificant differences.

Sowing Dates	No. of flowers. plant	No. of capsules. Plant	No. of seeds. capsule	No. of seeds. plant	1000 seed weight (g)	Seed weight. plant	Seed yield (g/m ²)
D1	100.73	64.07	59.53	3822.87	2.14	7.60	184.43
D2	101.33	48.23	51.87	2636.13	2.20	5.17	110.58
D3	49.53	38.43	51.12	1934.43	1.94	3.45	64.66
D4	44.27	34.73	43.35	1613.40	2.00	2.74	52.46
D5	35.17	21.37	37.80	989.13	1.94	1.63	25.47
LSD (p≤0.05)	20.77	13.02	8.09	684.58	0.17	1.25	37.13
LSD (p<0.01)	28.46	17.84	11.09	937.93	n.s	1.71	50.88

 Table 4. Some yield and yield component traits of black seed grown within different sowing dates at autumn 2015-2016.

Sowing dates; D1 (2nd November, 2015), D2 (21st November, 2015), D3 (11th December, 2015), D4 (31st December, 2015) and D5 (20th January, 2016) obtained data in **Table 5.** Confirmed that all the yield and the yield components traits were not significantly different, but insignificant higher values recorded by the plants sown and grown at Autumn season compared to spring season except for the 1000 seed weight which showed opposite direction,

may be due to the more growth performance by the plants of Autumn, higher values recorded by Autumn seasons were 66.21 flowers/plant, 41.37 capsules/plant, 48.73 seeds/capsule, 2199.19 seeds/plant, 4.12 g seed weight/plant and 87.52 seed yield/m², whereas lower values recorded by plants at spring season were 23.76 flowers/plant, 15.60 capsules/plant, 38.74 seeds/capsule, 658.27 seeds/plant, 1.31g seed weight/plant and 17.83 seed yield/m². Oppositely, the higher 1000 seed weight was 2.22 g recorded by plants of the spring season and a lower 1000 seed weight was 2.04 g recorded by the autumn season.

Growing seasons	No. of flowers. plant	No. of capsules.plant	No. of seeds. capsule	No. of seeds. plant	1000 seed. weight (g)	Seed weight. Plant (g)	Seed yield (g/m ²)
Spring	23.76	15.60	38.74	658.27	2.22	1.31	17.83
Autumn	66.21	41.37	48.73	2199.19	2.04	4.12	87.52
Stat.t	1.46 ^{n.s}	2.16 ^{n.s}	1.60 ^{n.s}	2.03 ^{n.s}	0.68 ^{n.s}	2.15 ^{n.s}	2.15 ^{n.s}

Table 5. Some yield and yield component traits of black seed cultivated at spring 2015 and autumn 2015-2016.

t(12)0.05=2.18, * significant at 0.05 level, ^{n.s} not significant.

Table 6. Revealed that there were highly significant differences existed between the two species of black seed that cultivated under different sowing dates in spring and autumn seasons and generally, N. sativa record higher levels of the chemical contents at both seasons especially at autumn compared to N. arvensis, while the ash content observed at higher levels in N. arvensis especially at spring compared to N. sativa. These results were related to that point that the production of secondary metabolites such as essential oil within the seeds of aromatic plants was under diverse physiological, biochemical metabolic and genetic regulation and can be manipulated by alteration in the growing conditions and management of agricultural practices and confirmed by [12,13.14,15]. Regarding to results in Table 6. Carbohydrate% recorded maximum value by N. sativa that cultivated in autumn season that was 58.30% followed by N. sativa grown in spring which was 51.00%, they were highly significantly exceeded N. arvensis at both seasons autumn and spring, which indicated that the soil and environment of hallabja more suitable for growth of N. sativa compared to N. arvensis. Concerning the levels of carbohydrate of black seed that recorded by several researchers were ranged between 24.9-40.0% [25,26]. The confidence interval of this study showed higher levels and ranged between 40.95-55.10%. Similarly, protein content records highly significant level that was 20.60% by N. sativa grown at autumn compared to the rest, but the confidence interval showed the range 14.08-18.76% which was lower compared to the referenced range 20.85-31.20% recorded by [25, 26]. Fixed oil content obtained by N. sativa in the spring season and autumn season were 29.66% and 28.31% respectively, followed by Oil content of N. arvensis grown at spring that was 27.36%, were not significant between themselves but they predominated significantly the oil content that recorded by N. arvensis which was only 20.37%. And regarding the lower level and upper level of fixed oil content in this study represented by confidence interval which was 22.99-29.86% it shows a lower range compared to the range recorded by [27, 28, 29, 30]. Which was 31-35%. Volatile oil content that recorded by both species were high and acceptable values to the reference records and the higher levels also were observed in *N. sativa* at Spring and autumn seasons which were 0.82%

and 0.71% respectively followed by 0.65% gained by *N. arvensis* grown at spring while the 0.62% was the lowest level recorded by *N. arvensis* grown at autumn season. The confidence interval for volatile oil in this study showed the range 0.62-0.78% has exceeded the range that recorded by [27, 28, 29, 30]. which was 0.4-0.7%. Ash content in black seeds as shown in **Table 6.** The higher level was 5.30% followed by 4.90% both occurred in *N. arvensis* grown in spring and autumn respectively, while 2.84% was the lowest level recorded by *N. arvensis* grown at spring. The range of 3.20-5.17% that shown by the confidence interval of ash content in the seeds of both variety was the following, the range 3.7-4.7% recorded by [25, 26].

			1		1	
Treatments		Carbohydrate %	Protein %	Fixed Oil %	Volatile Oil %	Ash
						%
N. sativa	Spring	51.00	15.30	29.66	0.82	2.84
	Autumn	58.30	20.60	28.31	0.71	3.70
<i>N</i> .	Spring	38.60	14.69	27.36	0.65	5.30
arvensis	Autumn	44.20	15.10	20.37	0.62	4.90
LSD	p≤0.01	9.38	3.55	4.34	0.17	2.29
	1-					
C.I	99%	$40.95 \le \mu \le 55.10$	$14.08 \le \mu \le$	$22.99 \le \mu \le$	$0.62 \leq \mu \leq 0.78$	3.20 ≤
			18.76	29.86		$\mu \leq$
						5.17

 Table 6. Chemical contents of *N.sativa* and *N. arvensis* grown under different sowing dates at spring and autumn seasons.

Conclusions

Based on the obtained results of this study, we can conclude the following: Due to the favorable environmental and the soil condition of Hallabja, both N. sativa and N. arvensis can be cultivated successfully, in the autumn season they can be considered as rainfed crops, while in spring season they need some supplementary irrigations. The early sowing dates always at the both seasons showed superiority in most growth and yield characters, which is due to the longer life period of the crops in early sowing dates. In spring cultivation, the higher yield (25.84) g/m² gained by 1st March sowing date compared to (9.82) g/m² of the sowing date in 20th March, hence, every day delaying in seeding decrease seed yield by (0.8) g/m^2 , this shortage may be due to that at late dates the crop blooming and seed filling stages companied with unfavorable conditions especially, high temperature and low humidity. Also, in autumn cultivation it was concluded that the early sowing dates namely; 2nd and 21st November gained 6.24 fold and 3.34 fold increasing in seed yield respectively compared to the lowest yield recorded by 20th January. Regarding to the difference between the two species, N. arvensis surpassed N. sativa in all characters except the 1000 seed weight that oppositely record higher value in N. sativa compared to N. arvensis. The interaction between species and the sowing dates were insignificantly affected on the traits of the study. Autumn cultivation surpassed significantly in growth characters compared to the spring cultivation as averages of the both species and different sowing dates. Also, the seed yield gained in autumn cultivation was significantly exceeded by 3.9 fold compared to the seed yield in spring cultivation. The seeds of N. sativa that produced at autumn contained the highest level of carbohydrates and protein, which were 58.30% and 20.60% respectively. While, they were gained high levels of fixed and volatile oils in spring cultivation, which were 29.66 and 0.82

respectively. The *N. arvensis* seeds that grown in autumn records lowest levels of the chemical contents.

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