EFFECT OF THREE VARIETIES OF BASIL AND THE NITROGEN SOURCE IN ESSENTIAL OIL CONTENTS

Zainb H. Alakashy*[1] and Nidhal A. H. Al-Bedairi [2]

[1]Dept. of Horticulture and Landscaping, Faculty of Agriculture/University of Kufa, Njaf, Iraq

[2]Dept. of Biology, Faculty of education for Girls/University of Kufa, Njaf, Iraq

E-mail: zainab.alakaishi@uokufa.edu.iq

ABSTRACT

The study was conducted during the 2018 growing season in a field belong to Horticulture and Forestry Division/ Iraqi Ministry of Agriculture / Al-Najaf Agricultural Directorate. Three varieties of basil were tested as the first factor including *Ocimum basilicum* var. basilicum (sweet basil), *O. basilicum* var. purple (red basil) and *O. basilicum* var. citriodorum. (lemon basil). The second factor was the effect of three sources of nitrogen fertilizers including chemical using Urea at the level of 400 mg. L^{-1}, Organic fertilizer using a liquid organic nitrogen at 4 ml. L^{-1} and Bio- fertilizer using *Azotobacter chroococcum* at 5 gm. bacteria $.m^{-2}$ was applied to the soil while the control treatment was the spraying with distilled water. Randomized Complete Block Design (R.C.B.D.) was used as a factorial experiment in a split-plot system with three replicates for each treatment. The L.S.D. test was adopted to compare mean with a probability of 0.05 level ($P < 0.05$).The Results showed superiority of red basil cultivar increased the active constituents content of Linalool to 64.96 $mg.gm^{-1}$ Myercene to 29.87 $mg.gm^{-1}$. While the Nitrogen source with Bio-fertlizer was superior increased Camphor ,Linalool, Limoine and Myercene to 7.82,77.92,18.45and 35.86 $mg.gm^{-1}$ respectively, compared with control treatment.

Key Words: Basil varieties, Urea, Organic Fertilizer, Bio-fertilizer, Essential oil.

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*The research is taken from the thesis of the first researcher

INTRODUCTION

Ocimum pp.L. is a one of the important leafy vegetable plants which belong to Lamiaceae family, that's includes many aromatic plants that are spread all over the world, especially in the countries of East Asia, the Mediterranean basin and the African continent (14). It is also one of the most important sources of volatile essential oils, which are characterized by the strong aromatic smell because they contain terpenes, which collect inside specialized glandular trichomes (7). Myercene and Linalool Therefore, fresh and dry green basil leaves are used, as well as their content of volatile oils and its various types in the fields of medicine, preventive and complementary therapies and pharmaceutical preparations due to their biological activity and the lack of their side effects compared to the pain medications chemically (4). Dry basil leaves are also used as strong spices or flavors in preparing food. Wierdak (18) find the Plants belonging to the *Ocimum* genus are a rich source of essential oils that have expressed many biological activities and their use in therapeutic areas widely used, there is a large variety of chemical patterns varying in type and concentration within the same cultivar basil. Some studies showed a significant heterogeneity in morphological and chemical properties of basil varieties and indicated the importance of the *O.basilicum* var. citriodorum. (lemon basil) for the accumulation of the essential oil content in its leaves by 1.09% and high (20.5%) geranial, neral (15.8%), (10.7%) E-caryophyllene, (10.1%) bisabolene and (9.8%) linalool and

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nerol 7.8%. So,the aim of this study is to know the effect of the source of nitrogen on content of some active constituents for three cultivars of basil plant under the environmental conditions of the Iraqi Middle Euphrates region.

MATERIALS AND METHODS

A field experiment, arranged in a randomized complete blocks design with three replications, was conducted in the Experimental field of Horticulture and Forestry Division of the Directorate of Agriculture Najaf province, during the growing season of 2018 , Soil analysis tests were carried out before planting and fertilization by taking ten samples , to be analyzed in the analysis laboratory of the Department of Soil and Water Resources in the Faculty of Agriculture / University of Kufa, as shown in Table (1).

Table 1: Physical and chemical properties of the experimental soil

Physical analysis		Chemical analysis			
Sand	200 gm.kg^{-1}	pH	7.3	Mg^{++} (m.equ.L^{-1})	2.71
Silt	350 gm.kg^{-1}	EC mmohs/cm	1.98	Na^{+} (m.equ.L^{-1})	9.24
Clay	450 gm.kg^{-1}	Total N%	0.72	$HCO3^{-}$ (m.equ.L^{-1})	0.32
Soil texture	Clay loam	Total P%	0.32	Cl^{-} (m.equ.L^{-1})	4.39
Organic matter (O.M)	4.9 gm.kg^{-1}	Total K%	0.75	$SO4^{--}$ (m.equ.L^{-1})	19.72
field capacity %	28.32 %	Ca^{++} (m.equ.L^{-1})	3.75	Fe (mg.L^{-1})	0.40

The experiment included two factors, which included three varieties of basil, *Ocimum basilicum* var. *basilicum* (sweet basil) , *O. basilicum* var. purple (red basil) and *O. basilicum* var. *citriodorum*. (lemon basil) , Its symbol (V1,V2 and V3)respectively and the second factor, it was using different sources of nitrogen fertilization, as it included both the nitrogenous fertilizer urea CO (NH2)2 containing 46%N which sprayed on the vegetative at a concentration of 400 mg. L $^{-1}$ (5) , Symbolize it (N1). The second was the liquid organic nitrogen (plus Optimus) prepared with nano-technology, which contains organic nitrogen at a concentration of 4% and the product was produced by the company Agri. Sciences, as it was sprayed on the vegetative growth at the level of 4 ml. liters $^{-1}$ according to the recommendation of the producing company has code (N2) and third source is addition of the soil applied *Azotobacter chroococccun*, with a concentration of CFU106 ml^{-1} at 5 gm. bacteria. m^{-2}, The symbol has N3 (8), the control treatment (without addition) that is sprayed with distilled water only and has a symbol of N0. All necessary cultural practices and plant protection were followed uniformly for all the plot during the entire period of experimentation fifteen plants were randomly selected from each plot for extraction essential oil from basil leaves. Randomized Complete Block Design (R.C.B.D.) was used as a factorial experiment (3×4) in a split-plot design with three replicates for each treatment. The comparison was done using L.S.D. test to compare means with a probability of 0.05 level ($P < 0.05$).

Essential oil extraction

According method Kmiecik *et al.* (9) essential oils were extracted from air dried plant material (leaves) by hydro-distillation, using a modified Clevenger type apparatus and lasted 3 h. The distilled essential oils had been dehydrated with 25 ml $C_4H_{10}O$ and stored at 4 °C in dark airtight bottles until further .

Measuring some active constituents in volatile oil

Some of the active compounds from the oil extracted were measured by the GC

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chromatography device to estimate the concentration of each of the following compounds: Camphor, Linalool, α -pinene, limonine and myercene (12).

RESULTS AND DISCUSSION

1- Camphor

It is noticed from Table (2) the superiority of red basil cultivar plants in increasing the active compound content of camphor, as it achieved a concentration of 6.43 mg.gm^{-1} compared to the plants of the class of lemon basil and sweet basil cultivar which reached 5.42 and 4.71 mg.gm^{-1} respectively.

The treatment of fertilization with bio-fertilizer and spraying with liquid organic nitrogen achieved significant increase in the concentration of the active compound camphor amounted to 7.82 and 6.91 mg .gm^{-1}, While the interaction between the plant variety and the type of nitrogen source showed the superiority of the red basil plants treated with the bio- fertilizer *Azotobacter chroococccun* (V2N3) significantly, it reached 8.96 mg.gm^{-1} compared with the intervention treatment of the lemon basil cultivar without spraying with nitrogen V3N0 as it reached 2.13 mg.gm $^{-1}$.

2- Linalool

show Table (2) the superiority of the plants of the red and lemon basil variety in increasing the content of the active compound Linalool, with an increase of 64.96 and 63.90 mg.gm^{-1}, which differed significantly About sweet basil cultivar which 58.03 mg.gm^{-1}.The treatment with the Bio-fertilizer was significant increasing the oil content of the active substance by 77.92 mg.gm $^{-1}$ and with a significant compared with control treatment of 37.66 mg.gm $^{-1}$,While the interaction between the plant variety and the type of nitrogen source showed the superiority of the lemon basil plants treated with the Bio-fertilizer *Azotobacter chroococccun* V3N3 significantly, it reached 88.45 mg.gm^{-1} compared with the treatment of the sweet basil cultivar without spraying with nitrogen V1N0 as it reached 34.08 mg.gm $^{-1}$

3- α – pinene

From data in the same table, we note the superiority the red basil cultivar in increasing the concentration of the active compound α-pinene 1.09 mg. gm^{-1} camper by sweet and lemon basil reach 0. 98 and 0.93 mg.gm^{-1},respectively. The treatment of fertilization with the bio-fertilizer *Azotobacter chroococccun* and spraying with liquid organic nitrogen achieved significant increase in the concentration of the active compound α-pinene amounted to 1.30 and 1.20 mg.gm $^{-1}$,While the interaction between the plant variety and the type of nitrogen source showed the superiority of the red basil plants treated with the bio-fertilizer Azotobacter chroococccun V2N3 and the interference of V2N2 significantly, it reached 1.39 and 1.33 mg.gm $^{-1}$

4- Limonine

The results of the same table indicate significant difference for variety in Limonine content of red basil reached 18.05 mg.gm $^{-1}$, while the content of the plants of the class of lemon basil and sweet basil cultivar was 17.60 and 16.56 mg.gm $^{-1}$, respectively.The treatment of spraying with liquid organic nitrogen(N2) and Bio-fertilizer achieved significant superiority in increasing the concentration of the active compound by 16.39 and 18.45 mg.gm $^{-1}$, and with a significant difference from the urea spray treatment and control treatment 8.63 and 6.83 mg.gm $^{-1}$.Interference between the plant variety and the type of nitrogen source showed the superiority of the red basil plants treated with the bio-fertilizer *Azotobacter chroococccun* V2N3 and sprayed with liquid organic nitrogen(optmas) significantly, reaching 23.58 and 23.09 mg.gm^{-1} compared with the sweet basil cultivar without spraying with nitrogen V1N0 which arrived to 9.59 mg.gm $^{-1}$

5- myercene

The results of the table indicate the superiority of the plants of the red basil cultivar in increasing the oil content of compound myercene, as it reached 29.87 mg.gm^{-1}.

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Table (2):The effect of cultivar and nitrogen source on leaves content of the active compound in essential oils

Variety	Nitrogen Source	Characteristics				
		Camphor	Linalool	α-pinene	Limonine	Myercene
Sweet basil (V1)	N0	2.59	34.08	0.51	9.59	13.62
	N1	4.85	53.60	0.97	15.94	26.15
	N2	6.52	70.43	1.16	19.75	31.73
	N3	7.73	74.02	1.29	20.98	35.65
Red basil (V2)	N0	3.12	40.95	0.64	10.93	16.45
	N1	5.37	67.92	1.03	13.10	28.02
	N2	8.30	79.66	1.33	23.09	36.38
	N3	8.96	71.31	1.39	23.58	38.66
Lemon basil (V3)	N0	2.13	39.95	0.52	8.95	12.92
	N1	4.11	61.73	0.88	12.85	23.63
	N2	5.93	65.49	1.10	20.62	29.34
	N3	6.68	88.45	1.22	22.02	33.29
L.S.D.0.05		0.32	1.12	1.06	3.06	1.43
Mean of Variety	**Sweet basil (V1)**	5.42	58.03	0.98	16.56	26.78
	Red basil (V2)	6.43	64.96	1.09	18.05	29.87
	Lemon basil (V3)	4.71	63.90	0.93	17.60	24.79
L.S.D.0.05		0.78	1.22	0.03	2.67	1.90
Mean of Nitrogen Source	N0	2.61	38.32	0.55	6.83	14.33
	N1	4.77	61.08	0.96	8.63	25.93
	N2	6.91	71.86	1.20	16.39	32.48
	N3	7.82	77.92	1.30	18.45	35.86
L.S.D.0.05		1.98	2.43	0.43	1.43	1.23

The fertilization treatment with the Bio-fertilizer *Azotobacter* succeeded by increasing the concentration of the myercene compound significantly by 35.86 $mg.gm^{-1}$, with a significant difference from the comparison treatment of 14.33 $mg.gm^{-1}$,While the interaction between the variety and source of nitrogen source showed the superiority of the red basil plants treated with the Bio-fertilizer *Azotobacter chroococcccun* (V2N3) significantly, it reached 38.66 $mg.gm^{-1}$ compared with the intervention treatment the lemon basil cultivar without spraying with nitrogen V3N0 as it reached 12.92 $mg.gm^{-1}$.

The effect of the genetic factors related to the cultivars may have had a major role in the variability of the volatile oil and its active compounds (13),or the increasing concentration of active compounds Camphor, Linalool, α –pinenem, Limonine and Myercene in *O. basilicum* var. purple (red basil),perhaps caused growth natural of red basil which character in Profuse growth(6 and 11),the reason for the increase in the active compounds of the volatile oil by the effect of the treatment of *Azotobacter chroococcccun* is the increase in the amount of nitrogen installed by the bacteria that contribute in formation of the chlorophyll molecule and thus increase the efficiency of the photosynthesis process,

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which led to an increase in total soluble carbohydrates (2). The increase in the production of carbohydrates as a result of the efficiency of the process of photosynthesis leads to an increase in the production of Terpenes compounds through the entry of carbohydrates in the process of glycolysis and the production of pyruvic acid and then mefalonic subsequently, which is the main key for the production of isoprene units C5H8 which is the structural unit for the formation of turbine volatile oils (17),These results are in agreement with Salman (16) on the red basile and Abid Ameen (3) on the *Magnesium Fertilization*.

Also Perhaps the reason is due to the structural structure of organic fertilizers represented by the abundance of nutrients such as carbon and some micronutrients and their transporters of organic acids led to stimulating physiological processes, especially photosynthesis, which caused the increase and accumulation of nutrients manufactured in the plant, especially the total dissolved carbohydrates (1) which indicated that nitrogen fertilization increases the production of secondary metabolism compounds, which are an important source of formation of active compounds in the plant, these results are in agreement with Mahmood and Slman (10),Salama and Yousef (15).

CONCLUSION

Under the experimental treatment of the present study it was revealed that the dominant constituent for linalool in three variety.While the component concentrated in *Ocimum basilicum* var. purple (red basil) whereas,At the same time, the chemical composition of O. *basilicum* var. *citriodorum*. (lemon basil) hybrid plant differed substantially from the two basil varieties: it had meddle value of compound content,represented by Camphor , linalool and limonine

with another prevalent constituent

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