ENVIRONMENTAL STUDIES ON DIFFERENT NITROGEN FERTILIZERS ON GROWTH YIELD AND VOLATILE OIL OF *THYME* PLANT

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ABSTRACT

To investigate the effect of different types of nitrogen fertilization the growth and productivity of thyme within reducing the harm effects of the excess of nitrogen fertilizers as an environmental and healthy problems, two successive experiments were carried out in pots during 2016, 2017 season's at the experimental green house Faculty of Agriculture, Ain Shams University Cairo , Egypt. Three types of nitrogen fertilizers amonium nitrate, ammonium sulphate and Urea, with three concentrations (40%, 60%, 80%) were used. Data showed an increase in number of lateral branches, plant height and amino acids with 40 units of ammonium nitrate, while the best effect to reduce NO_3, NO_2 as a lowest value was 40 units of ammonium sulphate. The highest value of herb fresh weight was recorded by 40 units of urea, (42:48 g / plant) while herb dry weight (18:54 g/ plant) by 40 units of ammonium sulphate, volatile oil (0.92 % v/ w) with 60 units of ammonium nitrate , amino acid (3.71 g / 100 g fw) with 80 units of urea.

Key words: thyme plants, nitrogen fertilizers, growth parameters, oil quality.

INTRODUCTION

Thyme plant is an aromatic and medicinal plant indigenous to Europe and Asia. It is a member of the Lamiaceae family. *T. serpyllum*, known as wild *thyme* is native to Mediterranean Europe and North Africa, mainly at the higher altitudes. The plant is aromatic, antiseptic, diaphoretic, analgesic, carminative and diuretic; also it acts as carminative and stimulant, also being used in mouth washes, gargles, cough and colds. Its essential oil contains various compounds that are very powerful, proven disinfectants enhancing the immune system and fighting infection. The oil relieves rheumatism and is also used in hear loss-treatment (Aziz *et al.*, 2008) *Thymus* species are commonly used as herbal tea, flavoring agents (condiment and spice) and medicinal plants. The best time for harvesting of this plant is the beginning of flowering period (Yadegari, 2015). The major components are phenols (mainly thymol and carvacrol), monoterpen hydrocarbon and alcohol that have insecticide activity (Mendes *et al.*, 2014; Deletre *et al.*, 2015). Essential oil % of *thyme* has been reported from 0.32% (ÖZGÜVEN and Tansi, 1998). to 4.9% (Carlen *et al.*, 2009). Thymol and carvacrol, which are the major components of *thyme* oil (Atti-Santos *et al.*, 2004). have antioxidant activity (Kulisic *et al.*, 2005), antimicrobial activity (Prabuseenivasan *et al.*, 2006), antifungal activity (Šegvić Klarić *et al.*, 2007). In other publication major volatile constituents obtained from the aerial parts of the plant are geranial, linalool, carvacrol, thymol and trans-thujan-4-ol/terpinen-4-ol (Omidbaigi and Arjmandi, 2001; Ozcan and Chalchat, 2004). In *thyme* plants were collected during the flowering period in eastern Morocco (Taforalt) in May, essential oil yield was 1.0% and camphor (38.54%), camphene (17.19%), αpinene (9.35%), 1,8-cineole (5.44%), borneol (4.91%) and β-pinene (3.90%) were the major oil components. However, characteristic compounds of *T.vulgaris* essential oil are thymol (44.4–58.1 %), *p*-cymene (9.1-28.5%), γ-terpinene (6.9–18.9%) and carvacrol (2.4-4.2%) (Eissa *et al.*, 2005; Aziz *et al.*, 2008; El-Din *et al.*, 2009).

Nutrition plays a key role in the growth and development of all crop plants. In the case of medicinal plants that synthesize essential oils, nutrients can effectively increase oil yield and quality (Aziz *et al.*, 2010; Zheljazkov *et al.*, 2010; Jabbari *et al.*, 2011; Sharafzadeh *et al.*, 2011a; Sharafzadeh *et al.*, 2011b; Zheljazkov *et al.*, 2011). Nitrogen is one of essential minerals, is used by plants to syntheses many organic compounds: amino acids, proteins, enzymes, and nucleic acids. Amino acids and enzymes are very important in the biosynthesis of numerous compounds which are essential oil constituents (Koeduka *et al.*, 2006). Plants mainly use nitrate and ammonium nitrogen, and the nitrate ion moves from the soil solution to the roots by passive and active uptake. To undergo further transformations, the nitrate anion must be reduced to ammonia, which occurs in the cells of the roots and in the aerial parts ($NO_3 \rightarrow NO_2$) as well as in chloroplasts ($NO_2 \rightarrow NH_3$). Organic nitrogen compounds are formed by biosynthesis from nitrogen taken up in the cation form (NH_4^+) and plant species-specific amino acids are the main product of this synthesis (Nowacki, 1980; Kopcewicz, 2002). Nitrogen uptake and use by plants are dependent, among others, on the type of fertilizer (nitrogen form) and its amount (nitrogen rate). The response of plants to different nitrogen forms depends on the concentration of NH_4^+ and NO_3^- ions in the nutritional environment, whereas the excess uptake of NH_4^+ ions causes disturbances in plant metabolism leading even to toxicities (Barker and Maynard, 1972; Chaillou *et al.*, 1986; Lamb *et al.*, 1993).

Considerably more work must be performed on the effect of different forms of chemical nitrogen on thyme plant, therefore the present work takes into consideration a study to cover the following points:

- Effect of different forms of nitrogen on thyme herbage yield by measuring fresh and dry weight.
- Effect of different forms of nitrogen on some chemical constituents of thyme like amino acids, nitrite and nitrate.
- Effect of different forms of nitrogen on thyme essential oil yield

MATERIALS AND METHODS

1. Location and duration:

Experiments were carried out during the two successive seasons of 2016 and 2017. Trials were undertaken and observed in the experimental farm of Fac. of Agric., Ain shams Univ. Cairo, Egypt.

2. Plant materials:

At the first week of February of both seasons seeds of *thyme* were kindly provided by "Sekem" company of medicinal and aromatic plants were sown in nursery. After 45 days, seedling were transplanted in 30 cm diameter plastic pots filled with 10 kg of clean and washed sand in combining with 1/2 kg compost from El-khalil company , 10 g of calcium superphate ,15.5 % (representing as 200 Kg per fed.) and 15 g potassium sulphate , 48 % representing as 150 Kg per fed.

Chemical and physical properties of used compost (El Khalil Compost):

Weight of m^3 (kg)	580.5
Moisture %	35
PH	7.5
EC (mmhos)	4.5
Total N (%)	1.69
Organic matter (%)	1.5
Total carbon (%)	28.6
Ash %	29.5
C/N ratio	20.5
Total P (%)	0.8
Total K (%)	1.0

All horticultural practices including irrigation, mineral fertilization except nitrogen source, micro nutrient sp eying and pest and diseases control were done as used recommendation in this respect.

3. Expermental treatments:

- Three types of nitrogen fertilizers were used; Ammonium Nitrate 33.5%, Ammonium Sulphate 20.6% and Urea 46%, also three doses of each type were used. So, the treatments were designed as Follows:

T_1 : 80% unit of Ammonium Nitrate T_2 : 60% unit of Ammonium Nitrate

T_3 : 40% unit of Ammonium Nitrate T_4 : 80% unit of Ammonium Sulphate

T_5 : 60% unit of Ammonium Sulphate T_6 : 40% unit of Ammonium Sulphate

T_7 : 80% unit of Urea T_8 : 60% unit of Urea

T_9 : 40% unit of Urea

-regarding to the time of application, the dose of each pot were divided into three doses: mid of may, mid of June and mid of July in the two seasons.

4. Experimental design and statistical analysis:

The Experimental of both seasons were arranged in complete Randomized Block design with three replicates. Every replicate contained three plants. The results of each season were statistically analyzed separately.

A Two – way analysis of variance with interaction was carried out, as described by SAS, (2009)

5. Methods:

Analytical methods were carried out on herbage and essential oil as follows:

On herbage:

Fresh herbage yield. dry weight ., free amino acids%, nitrate%, nitrite % and essential oil percent.

5.1. Determination of Free Amino Acids:

Free amino acids were determined calorimetrically by using ninhydrin solution according to (Chandrasekhar *et al.*, 1981). Ninhydrin solution was prepared by dissolving 2.0g ninhydrin in 25ml of acetone followed by addition of 25ml 0.2M acetate buffer (pH 5.5). Then the solution was stored in a brown bottle to protect its from light.

A standard solution of lysine was prepared by dissolving 20.0mg of lysine in 100ml of 10% aqueous isopropanol solution.

Aliquots of 0.05ml to 0.650ml of the standard lysine solution containing 10 to 130µg lysine or 1ml of sample extract were pipetted out into a series of test tubes, then total volume was made up to 4ml with distilled water. One ml of the ninhydrin reagent was added to each tube, mixed well, and the tubes were kept in a boiling water bath for 15 minutes. The tubes were then cooled and volume was made up to 10ml in a measuring flask with 50% ethanol.

Optical density was measured at 570nm and free amino acids content were calculated as lysine mg/100g f.wt.

5.2. Determination of nitrate concentration:

Nitrate content was determined calorimetrically by using salicylic acid solution according to (Cataldo *et al.*, 1975). as follows:

0.1g of ground plant samples were suspended in 10mL distilled water, kept at 45˚C for 1h and then filtered through Whatman No.40 filter paper .Samples were extracted and analyzed immediately or within 24 h after extraction when stored at 4˚C .

Aliquots of 10µl to 100µl of the standard KNO_3 solution containing 1 to 10µgK NO_3 or 1ml of sample extract were mixed thoroughly with 0.4mL salicylic acid solution (5% w/v dissolved in concentrated sulfuric acid). After 20 min at room temperature, 9.5 mL 2N NaOH solution was slowly added .This method is based on the formation of nitro salicylic acid that shows, in highly basic solution, a maximum absorption at 412 nm. Nitrate–N in plant tissue expressed as mg NO_3/100g dry weight.

5.3. Determination of nitrite content:

Nitrite content was determined colourimetrically according to (Aydın *et al.*, 2005). as follows: 90.0 mL of sample in a 100 mL volumetric flask is acidified by 0.1N AcOH solution to get approximately pH of 5.0 and diluted to the mark with pH 5 buffer. An aliquot (maximum 60.0 mL) of that solution containing nitrite in the working range (1.7–300.0 g NO_2) is used in nitrite analysis. Following the addition of 30 mL 5 × 10−4 M barbituric acid solution, the mixture is left to stand for half an hour. 5.6 mL of 5N NaOH solution is added and the mixture is diluted to the volume with buffer of pH

5.50. Absorbance is measured at 310 nm against reagent blank. Nitrite–N in plant tissue expressed as mg NO_2/100g dry weight.

5.4. Extraction of essential oil:

The oil herbage was distilled and oil volume was measured according to (Banthorpe, 2013). The procedure can be summarized in the following:

The fresh plant material subjected to distillation immediately after cutting to avoid loss of oil using a micro distilling apparatus.

The plant material was weighted, immediately chipped, placed in 500ml capacity flask of the apparatus and about 200ml of water was added to the flask. The oil trap was filled with water. The oil trap and the condenser were attached to the flask where tap water was circulated around the condenser for condensation of the oil vapor. The flask was placed on heating mental. Steam Distillation was continued usually 2 hr after water boiling until a constant amount of oil was observed, after complete distillation, the oil was permitted to stand undisturbed to obtain clear separation.

Oil content was determined as ml of oil per 100g fresh weight of plant material.

RESULTS AND DISCUSSION

Herb fresh weight (g/plant):

Data presented in Table (1) show the effect of different types of nitrogenous fertilizers and doses on fresh weight of *Thymus vulgaris* during 2016 and 2017 seasons. It is clear that high value was obtained with ammonium sulphate (M2) followed by urea (M3). whereas ammonium nitrate came later. However, 40 units recorded the highest effect on Herb fresh weight (g/plant). Whereas 80 units exhibited the less effect on herb fresh weight. Interaction between the two studied factors was significant in most cases; the higher interaction value (42.48) was obtained with 40 units from urea. However, the least interaction value (9.83) was obtained with 80 units from ammonium nitrate, these results were found to be agree with the finding of **(Omidbaigi and Nejad, 2000; Tabrizi *et al.*, 2011)** they demonstrated that nitrogen fertilization had a significant effect on thyme productivity.The same trend results as also recorded in the second season study.

But, the high value was obtained with ammonium sulphate (27.61) and the least value was obtained with ammonium nitrate (19.42).

Table (1): Effect of ammonium nitrate (A N), ammonium sulphate (A S) and urea with three different doses of nitrogen (80, 60 and 40%) on the fresh weight of thyme during two successive seasons in 2016 and 2017**.**

Units of nitrogen (%)									
	80	60	40	Mean	80	60	40	Mean	
	1st season				2nd season				
Herb fresh weight (g)									
A N	9.83 b	23.88 ab	21.61 ab	18.45 A	10.81 b	25.28 ab	22.15 ab	19.42 A	M 1
A S	18.81 ab	27.63 ab	33.92 ab	26.79 A	19.49 ab	28.37 ab	34.96 ab	27.61 A	M 2
Urea	10.91 b	24.85 ab	42.48 a	26.09 A	11.38 b	25.30 ab	43.25 a	26.65 A	M 3
Mean	13.19 B	25.45 AB	32.68 A		13.89 B	26.32 AB	33.46 A		

Means followed by different letters are significantly different at P ≤0.05; LSD's multiple range tests.

Herb dry weight (g/plant):

Data in Table (2) show that herb dry weight (g/plant) of *Thymus vulgaris* plants greatly affected with three different types of nitrogenous fertilizers and doses during 2016 and 2017 seasons.

Regarding the type of nitrogenous fertilizer, it is clear that high value was obtained with ammonium sulphate (M2). Whereas ammonium nitrate (M1) came later. However, 40 units recorded the highest effect on herb dry weight. Whereas 80 units exhibited the less effect on herb dry weigh. Interaction between the two studied factors was significant in most cases; the higher interaction value (18.54) was obtained with 40 units from ammonium sulphate. However, the least interaction value (11.18) was obtained with 80 units from ammonium nitrate (Habibi *et al.*, 2004). Explained the effect of nitrogen doses on dry matter production of garden thyme. The second season data showed that no significant differences between the type on the dose of nitrogenous fertilizers were obtained. But, the high value was obtained with ammonium sulphate (16.53) and the least value was obtained with ammonium nitrate (14.32).

Table (2): Effect of ammonium nitrate (A N), ammonium sulphate (A S) and urea with three different doses of nitrogen (80, 60 and 40%) on the dry weight of thyme during two successive seasons in 2016 and 2017.

Units of nitrogen (%)									
	80	60	40	Mean	80	60	40	Mean	
	1st season				2nd season				
Herb dry weight (g)									
A N	11.18 a	16.31 a	14.39 a	13.96 A	11.53 a	16.64 a	14.77 a	14.32 A	M 1
A S	15.14 a	15.04 a	18.54 a	16.24 A	15.46 a	15.26 a	18.87 a	16.53 A	M 2
Urea	11.35 a	15.91 a	15.41 a	14.23 A	11.63 a	16.27 a	15.61 a	14.51 A	M 3
Mean	12.56 A	15.75 A	16.12 A		12.88 A	16.06 A	16.42 A		

Means followed by different letters are significantly different at P ≤0.05; LSD's multiple range tests.

Amino acid concentration (g/100g fw):

Data illustrated in Table (3) show that three different types of nitrogenous fertilizers and doses greatly affected Amino acid (g/100gfw) in *Thymus vulgaris* during 2016 and 2017 seasons.

It is clear that high value was obtained with ammonium nitrate (M1). Whereas urea came later. However, 80 units recorded the highest effect on the concentration of Amino acid. On the other hand, 40 units exhibited the less effect on amino acid. Interaction between the two studied factors was significant in most cases, the higher interaction value (3.71) was obtained with 80 units from urea. Moreover, the least interaction value (2.27) was recorded with 40 units from ammonium sulphate.

Data of second season showed that higher value was obtained with ammonium nitrate (M1). Whereas ammonium sulphate (M2) recorded the lower values.

Significant differences were only obtained between M1 and M2. Moreover, 80 units recorded the highest effect on amino acid. Whereas 40 units recorded the less effect on amino acid. Interaction between the two studied factors was significant in most cases, the higher interaction value (5.11) was obtained with 80 units from urea. On the other hand, interaction lowest value (3.33) were obtained with 40 units from ammonium sulphate and 60 units from urea without significant differences between them , these results are harmony with (Chung *et al.*, 2010). Reported that nitrogen starvation decreased plant growth and amino acids content in Yarrow (*Achillea collina*) .

Table (3): Effect of ammonium nitrate (A N), ammonium sulphate (A S) and urea with three different doses of nitrogen (80, 60 and 40%) on concentration of the amino acids of thyme plant during two successive seasons in 2016 and 2017.

Units of nitrogen (%)									
	80	**60**	**40**	**Mean**	**80**	**60**	**40**	**Mean**	
	1st season				**2nd season**				
Amino acid (g/100gfw)									
A N	3.25 ab	3.46 ab	2.62 c	3.11 A	4.26 b	4.49 b	3.48 cde	4.08 A	M 1
A S	3.16 b	3.17 b	2.27 c	2.87 A	3.95 bcd	3.99 bc	3.33 e	3.76 A	M 2
Urea	3.71 a	2.53 c	2.35 c	2.86 A	5.11 a	3.33 e	3.42 de	3.95 A	M 3
Mean	3.37 A	3.05 B	2.41 C		4.44 A	3.94 B	3.41 C		

Means followed by different letters are significantly different at P $\leq$0.05; LSD's multiple range tests.

Nitrate concentration (g/100gdw):

Data tabulated in table (4) show that NO_3 (g/100gdw) of *Thymus vulgaris* plants greatly affected with both different types of nitrogenous fertilizers and doses during 2016 and 2017 seasons.

Regarding the type of nitrogenous fertilizer, it is clear that the best value was obtained with ammonium sulphate (M2) whereas ammonium nitrate was higher. However, 40 units recorded the best effect on NO_3. Also, 80 units recorded the highest effect on NO_3 Interaction between the two studied factors was significant in most cases; the best interaction value (0.52) was obtained with 40 units from ammonium sulphate. Moreover, the higher interaction value (8.15) was obtained with 80 units from ammonium nitrate. Data of second season showed that the best value was recorded with ammonium sulphate (M2). Whereas ammonium nitrate was higher.

Significant differences were only obtained between M2 and M1. However, 40 units recorded the best effect on NO_3. Also, 80 units recorded the highest effect on NO_3. Interaction between the two studied factors was significant in most cases, the best interaction value (0.31) was recorded with 40 units from urea. On the other hand, the higher interaction value (9.24) was obtained with 60 units from ammonium nitrate. The response of plants to different nitrogen forms depends on the concentration of NH_4^+ and NO_3^- ions in the nutritional environment, whereas the excess uptake of NH_4^+ ions causes disturbances in plant metabolism leading even to poisoning (Chaillou *et al.*, 1986; Lamb *et al.*, 1993; Barker, 1999).

Table (4): Effect of ammonium nitrate (A N), ammonium sulphate (A S) and urea with three different doses of nitrogen (80, 60 and 40%) on the NO_3 of thyme during two successive seasons in 2016 and 2017.

	Units of nitrogen (%)								
	80	**60**	**40**	**Mean**	**80**	**60**	**40**	**Mean**	
	1st season				**2nd season**				
	NO_3 (g/100gdw)								
A N	8.15 a	4.64 b	1.87 c	**4.89 A**	9.08 a	9.24 a	2.85 c	**7.06 A**	**M 1**
A S	1.8 d	0.94 fg	0.52 g	**1.10 C**	2.04 d	1.68 de	1.35 e	**1.69 B**	**M 2**
Urea	2.68 c	1.35 ef	1.48 de	**1.84 B**	3.42 b	1.82 d	0.31 f	**1.86 B**	**M 3**
Mean	**4.23 A**	**2.31 B**	**1.29 C**		**4.85 A**	**4.25 B**	**1.51 C**		

Means followed by different letters are significantly different at P ≤0.05; LSD's multiple range test.

Nitrite concentration (g/100gdw):

Data illustrated in Table (5) show that different types of nitrogenous fertilizers and doses greatly affected NO_2 concentration (g/100gdw) in *Thymus vulgaris* during 2016 and 2017 seasons

Regarding the type of nitrogenous fertilizer, it is clear that the best value was obtained with ammonium sulphate (M2). Whereas urea was higher. Significant differences were only obtained between M2 and M3. Moreover, 40 units recorded the best effect on NO_2. Whereas 80 units recorded the highest effect on NO_2.

Interaction between the two studied factors was significant in most cases, the best interaction value (0.44) was obtained with 40 units from ammonium sulphate. However, the higher interaction value (1.76) was obtained with 80 units from ammonium nitrate. Data of second season showed that the best value was recorded with ammonium sulphate (M2). Whereas ammonium nitrate was higher. However, 40 units recorded the best effect on NO_2. Also, 80 units recorded the highest effect on NO_2,this results are in agree with the results of (Baranauskienė *et al.*, 2003), they determined that nitrogen fertilizers influence the amount of nitrites in thyme plants.

Interaction between the two studied factors was significant in most cases; the best interaction value (0.92) was obtained with 40 units from ammonium nitrate. On the other hand, the higher interaction value (2.59) was recorded with 80 units from ammonium nitrate.

Table (5): Effect of ammonium nitrate (A N), ammonium sulphate (A S) and urea with three different doses of nitrogen (80, 60 and 40%) on the NO_2 concentration of thyme during two successive seasons in 2016 and 2017.

	Units of nitrogen (%)								
	80	**60**	**40**	**Mean**	**80**	**60**	**40**	**Mean**	
	1st season				**2nd season**				
	NO_2 (g/100gdw)								
A N	1.76 a	0.99 cd	0.73 e	**1.16 A**	2.59 a	1.21 d	0.92 f	**1.57 A**	**M 1**
A S	1.24 b	0.78 de	0.44 f	**0.82 B**	1.49 b	0.98 ef	0.93 f	**1.13 C**	**M 2**
Urea	1.19 bc	1.11 bc	0.88 de	**1.86 A**	1.48 bc	1.28 cd	1.17 de	**1.31 B**	**M 3**
Mean	**1.39 A**	**0.96 B**	**0.68 C**		**1.86 A**	**1.16 B**	**1.01 C**		

Means followed by different letters are significantly different at P $\leq$0.05; LSD's multiple range test.

Volatile oil % (v/w): Data in Table (6) show that Volatile oil % (v/w) of *Thymus vulgaris* plants greatly affected with different types of nitrogenous fertilizers and doses during 2016 and 2017 seasons. It is clear that high value was obtained with ammonium nitrate (M1) followed by ammonium sulphate (M2) whereas urea ranked in last order. Significant differences were only obtained between M1 and M3. However, 80 units recorded the highest effect value on volatile oil. whereas 60 units recorded the less effect value on volatile oil , these results agree with (Ehsanipour *et al.*, 2012). they proved an increase in essential oil concentration in fennel fruit under the influence of a higer rate of nitrogen , and in the same trend was found by (Sharafzadeh, 2011). on garden thyme.

Interaction between the two studied factors was significant in most cases; the highest interaction value (0.92) was obtained with 60 units from ammonium nitrate. However, the least interaction value (0.48) was obtained with 80 units from urea. The same trend results as also recorded in the second season study. But, the high value was obtained with ammonium nitrate (0.90) and the least value was obtained with urea (0.55).

Table (6): Effect of ammonium nitrate (A N), ammonium sulphate (A S) and urea with three different doses of nitrogen (80, 60 and 40%) on the percentage of volatile oil in thyme during two successive seasons in 2016 and 2017.

Units of nitrogen (%)									
	80	**60**	**40**	**Mean**	**80**	**60**	**40**	**Mean**	
	1st season				**2nd season**				
Volatile oil % (v/w)									
A N	0.84 a	0.92 a	0.79 a	**0.89 A**	0.89 a	0.97 a	0.83 a	**0.90 A**	**M 1**
A S	0.85 a	0.56 a	0.84 a	**0.75 A**	0.87 a	0.59 a	0.92 a	**0.80 A**	**M 2**
Urea	0.48 a	0.54 a	0.49 a	**0.51 A**	0.53 a	0.58 a	0.54 a	**0.55 A**	**M 3**
Mean	**0.73 A**	**0.67 A**	**0.71 A**		**0.77 A**	**0.72 A**	**0.77 A**		

Means followed by different letters are significantly different at P ≤0.05; LSD's multiple range tests.

REFFRENCES

Atti-Santos, A.; M. Pansera; N. Paroul; L. Atti-Serafini and P. Moyna. (2004). Seasonal variation of essential oil yield and composition of Thymus vulgaris L.(Lamiaceae) from South Brazil. Journal of Essential Oil Research, 16 (4): 294-295.

Aydın, A.; Ö .Ercan and S. Taşcıoğlu. (2005). A novel method for the spectrophotometric determination of nitrite in water. Talanta, 66 (5): 1181-1186.

Aziz, E.; S. Hendawy; E. El- Din Azza and E. Omer. (2008). Effect of soil type and irrigation intervals on plant growth, essential oil yield and constituents of Thymus vulgaris plant, Vol 4

Aziz, E. E.; M. El-Danasoury and L. Craker. (2010). Impact of sulfur and ammonium sulfate on dragonhead plants grown in newly reclaimed soil. Journal of herbs, spices & medicinal plants, 16 (2): 126-135.

Banthorpe, D. (2013). XIII Mentha Species (Mints). In Vitro Culture and Production of Lower Terpenoids and Pigments. Medicinal and Aromatic Plants IX, 37: 202.

Baranauskienė, R.; P. R. Venskutonis; P. Viškelis and E. Dambrauskienė. (2٠٠٣). Influence of nitrogen fertilizers on the yield and composition of thyme (Thymus vulgaris). Journal of Agricultural and Food chemistry, 51 (26): 7751-7758.

Barker, A. and D. Maynard. (1972). Cation and nitrate accumulation in pea and cucumber plants as influenced by nitrogen nutrition. Amer Soc Hort Sci J ،

Barker, A. V. (1999). Foliar ammonium accumulation as an index of stress in plants. Communications in soil science and plant analysis, 30 (1-2): 167-174.

Carlen, C.; M. Schaller; C. Carron; J. Vouillamoz and C. Baroffio. (2009). The new Thymus vulgaris L. hybrid cultivar'Varico 3'compared to five established cultivars from Germany, France and Switzerland. *In* IV International Symposium on Breeding Research on Medicinal and Aromatic Plants-ISBMAP20 ،٨٦٠ ٠٩pp 161-166.

Cataldo, D.; M. Maroon; L. Schrader and V. Youngs. (1975). Rapid colorimetric determination of nitrate in plant tissue by nitration of salicylic acid. Communications in soil science and plant analysis, 6 (1): 71-80.

Chaillou, S.; J. Morot-Gaudry; L. Salsac; C. Lesaint and E. Jolivet. (1986). Compared effects of NO3-and NH4+ on growth and metabolism of French bean. Physiol. vég, 24: 679-687.

Chandrasekhar, J.; J. G. Andrade and P. v. R. Schleyer. (1981). Efficient and accurate calculation of anion proton affinities. Journal of the American Chemical Society, 103 (18): 5609-5612.

Chung, R.-S.; C.-C. Chen and L.-T. Ng. (2010). Nitrogen fertilization affects the growth performance, betaine and polysaccharide concentrations of Lycium barbarum. Industrial crops and products, 32 (3): 650-655.

Deletre, E.; F. Chandre; L. Williams; C. Duménil; C. Menut and T. Martin. (2015). Electrophysiological and behavioral characterization of bioactive compounds of the Thymus vulgaris, Cymbopogon winterianus ،Cuminum cyminum and Cinnamomum zeylanicum essential oils against Anopheles gambiae and prospects for their use as bednet treatments. Parasites & vectors, 8 (1): 316.

Ehsanipour, A.; J. Razmjoo and H. Zeinali. (2012). Effect of nitrogen rates on yield and quality of fennel (Foeniculum vulgare Mill.) accessions. Industrial crops and products, 35 (1): 121-125.

Eissa, A.; A. Abou-Haddid and E. Omer. (2005). Effect of saline water on the dry matter production, nutritional status and essential oil content of thyme (Thymus vulgaris) grown in sandy soil. Egypt. J. of Hort, 31: 181-193.

El-Din, A. E.; E. E. Aziz; S. Hendawy and E. Omer. (2009). Response of Thymus vulgaris L. to salt stress and alar (B9) in newly reclaimed soil. Journal of Applied Sciences Res, 5.٢١٧٠-٢١٦٥ :

Habibi, R.; A. Farahani and H. Habibi. (2004). Message Research-Scientific Calendar-Applied Research Center, Agriculture and Natural Resources of Tehran. *In* ،

Jabbari, R.; M. A. Dehaghi; A. M. M. Sanavi and K. Agahi. (2011). Nitrogen and iron fertilization methods affecting essential oil and chemical composition of thyme (Thymus vulgaris L.) medical plant. Advances in Environmental Biology: 433-439.

Koeduka, T.; E. Fridman; D. R. Gang; D. G. Vassão; B. L. Jackson; C. M. Kish; I. Orlova; S. M .Spassova; N. G. Lewis and J. P. Noel. (2006). Eugenol and isoeugenol, characteristic aromatic constituents of spices, are biosynthesized via reduction of a coniferyl alcohol ester. Proceedings of the National Academy of Sciences, 103 (26): 10128-10133.

Kopcewicz, J. (2002). Lewak S. Fizjologia roślin. PWN, Warszawa ، Kulisic, T.; A. Radonic and M. Milos. (2005). Antioxidant properties of thyme (Thymus vulgaris L.) and wild thyme (Thymus serpyllum L.) essential oils. Italian journal of food science, 17.٣١٥ :(٣)

Lamb, M.; G. Clough and D. Hemphill. (1993). Pretransplant watermelon nutrition with various nitrate: ammonium ratios and supplemental calcium. HortScience, 28 (2): 101-103.

Mendes, M. D.; A. C. Figueiredo; M. M. Oliveira and H. Trindade. (2014 .(Influence of culture media and fungal extracts on essential oils composition and on terpene synthase gene expression in Thymus caespititius. Plant Cell, Tissue and Organ Culture (PCTOC), 118 (3): 457-469.

Nowacki, E. (1980). Gospodarka azotowa roślin uprawnych. PWRiL, Warszawa ،

Omidbaigi, R. and A. Arjmandi. (2001). Effects of NP supply on growth, development, yield and active substances of garden thyme (Thymus vulgaris L.). *In* International Conference on Medicinal and Aromatic Plants. Possibilities and Limitations of Medicinal and Aromatic Plant 576, pp 263-265.

Omidbaigi, R. and A. R. Nejad. (2000). The influence of nitrogen-fertilizer and harvest time on the productivity of Thymus vulgaris L. International Journal of Horticultural Science, 6 (4): 43.٤٦-

Ozcan, M. and J.-C. Chalchat. (2004). Aroma profile of Thymus vulgaris L. growing wild in Turkey. Bulgarian Journal of Plant Physiology, 30 (3-4): 68-73.

ÖZGÜVEN, M. and S. Tansi. (1998). Drug yield and essential oil of Thymus vulgaris L. as in influenced by ecological and ontogenetical variation. Turkish Journal of Agriculture and Forestry, 22 (6): 537-542.

Prabuseenivasan, S.; M. Jayakumar and S. Ignacimuthu. (2006). In vitro antibacterial activity of some plant essential oils. BMC complementary and alternative medicine, 6 (1): 39.

SAS. 2009. Statistical Analysis System, SAS user’s Guide: Statistics’. SAS institute Inc. Editors, Cary, NC

Šegvić Klarić, M.; I. Kosalec; J. Mastelić; E. Pieckova and S. Pepeljnak. (2007). Antifungal activity of thyme (Thymus vulgaris L.) essential oil and thymol against moulds from damp dwellings. Letters in applied microbiology, 44 (1): 36-42.

Sharafzadeh, S. (2011). Effect of nitrogen, phosphorous and potassium on growth, essential oil and total phenolic content of garden thyme (Thymus vulgaris L.). Advances in Environmental Biology: 699-704.

Sharafzadeh, S.; M. Esmaeili and A. H. Mohammadi. (2011a). Interaction effects of nitrogen, phosphorus and potassium on growth, essential oil and total phenolic content of sweet basil. Advances in Environmental Biology: 1285-1290.

Sharafzadeh, S.; M. Khosh-Khui and K. Javidnia. (2011b .(Effect of nutrients on essential oil components, pigments and total phenolic content of lemon balm (Melissa officinalis L.). Advances in Environmental Biology: 639-647.

Tabrizi, L.; A. Koocheki; P. R. Moghaddam; M. N. Mahallati and M. Bannayan. (2011 .(Effect of irrigation and organic manure on Khorasan thyme (Thymus transcaspicus Klokov). Archives of Agronomy and Soil science, 57 (3): 317-326.

Yadegari, M. (2015). Foliar application of micronutrients on essential oils of borago, thyme and marigold. Journal of soil science and plant nutrition, 15 (4): 949-964.

Zheljazkov, V. D.; C. L. Cantrell; T. Astatkie and J. B. Cannon. (2011). Lemongrass productivity, oil content, and composition as a function of nitrogen, sulfur, and harvest time. Agronomy Journal, 103 (3): 805-812.

Zheljazkov, V. D.; C. L. Cantrell; T. Astatkie and M. W. Ebelhar. (2010). Peppermint productivity and oil composition as a function of nitrogen, growth stage, and harvest time. Agronomy Journal, 102 (1): 124-128.

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دراسات بيئية لأسمدة نيتروجينية مختلفة على النمو والإنتاجية والزيت الطيار فى نبات الزعتر

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المستخلص

يهدف البحث إلى دراسة أهمية التسميد النيتروجيني على وزن العشب ونوعية نبات الزعتر في الحد من الآثار الضارة لزيادة الأسمدة النيتروجينية كمشاكل بيئية وصحية. تم إجراء تجربتان متتاليتان في الأصص خلال عام ٢٠١٦ – ٢٠١٧ في مزرعة كلية الزراعة، جامعة عين شمس، مصر. تم استخدام ثلاثة أنواع من الأسمدة النيتروجينية، نترات الأمونيوم، كبريتات الأمونيوم واليوريا ، بثلاثة تراكيز (٤٠٪ ، ٦٠٪ ، ٨٠٪) أظهرت البيانات أن أفضل تأثير لأقل كمية من NO_3 ، NO_2 وهو ٤٠ وحدة من كبريتات الأمونيوم وأعلى قيمة عشب وزن طازج (٤٢،٤٨ جم / نبات) بـ ٤٠ وحدة يوريا، بينما الوزن الجاف للأعشاب (١٨،٥٤ جم / نبات) كان مع ٤٠ وحدة من كبريتات الأمونيوم، وأعلى نسبة من الزيوت الطيارة (٠,٩٢٪ v/w) مع ٦٠ وحدة من نترات الأمونيوم أما أعلى قيمة للأحماض الأمينية (٣,٧١ جم / ١٠٠ جم) كانت مع ٨٠ وحدة من اليوريا.

الكلمات الدالة: نباتات الزعتر، الأسمدة النيتروجينية، عوامل النمو، جودة الزيت.