World Journal of Pharmaceutical Sciences ISSN (Print): 2321-3310; ISSN (Online): 2321-3086 Published by Atom and Cell Publishers © All Rights Reserved Available online at: http://www.wjpsonline.org/ Original Article



# Growth and oil yield of *thymus vulgaris* plant as influenced by some amino acids and ascorbic acid

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### Received: 15-07-2015 / Revised: 25-08-2015 / Accepted: 24-09-2015

## ABSTRACT

Field experiment was carried out during two successive seasons (2012 and 2013) to evaluate the effect of some amino acids such as phenylalanine and tryptophan as well as ascorbic acid as vitamin c with different doses on yield and essential oil of *Thymus vulgaris L*. plants. Phenylalanine at 150 mg / 1 produced the most promising effect on yield and essential oil as percentage or yield (ml/plant). Thymol, p-cymene and linalool were the main constituents of essential oil of this plant. Based on the experimental results it is recommended to treat thyme plants with phenylalanine at 150 mg / 1 to produce high mass production and oil yield.

Key Words: Thymus vulgaris, amino acids, ascorbic acid, essential oil

## **INTRODUCTION**

Thyme is the common name of Thymus vulgaris L. It is belongs to family Lamiaceae (Labiateae) or mint family. It is native to Europe and the Mediterranean region. It has many uses; some of them were recorded as folklore such as culinary. Antiseptic, expectorant, carminative and antispasmodic activities are attributed to thyme oil. Such activities are associated with the content of thymol (2-isopropyl-5- methyl phenol) and its conformational isomer, carvacrol (5-isopropyl-2methylphenol), which have greater antibacterial and antifungal activities than phenol and are less toxic [1 and 2].

There has been a recent trend to use naturallyoccurring compounds such as amino acids to achieve the regulation of plant growth and biosynthesis of important economic chemical constituents. They are used as precursors or phytohormones activators of and growth substances. L. Methionine is a precursor of ethylene and growth factors such as espermine and espermidine [3]. Gamal El-Din et al [4] noticed that foliar application of ornithine and phenylalanine at 50 and 100 mgL<sup>-1</sup> on Cymbopgon citrates led to increase significantly in vegetative growth, number of leaves and tillers as well as fresh and dry weight of herb. Refaat and Naguib [5] found that spraying peppermint plant with alpha-alanine at 25 and 50 ppm increased fresh and dry weight of the plant and essential oil content. Talaat and Yousef [6] investigated the effect of lysine at (25, 50,100ppm) on growth and yield of Ocimum basilicum L. Moreover, they found that the maximum mean values of plant growth parameters (plant height, fresh and dry weight of herb as well as number of branches) were recorded by application of 25mg/L from lysine on Ocimum basilicum L. The highest rate of application (100ppm) was more effective than the low rate (50ppm). Youssef et.al [7] showed that, application of tyrosine at 50ppm on Ocimum basilicum resulted in maximum values of plant height, plant diameter, number of branches, plant fresh and dry weights, leaves fresh and dry weights as well as flowers fresh and dry weight. Mahgoub and Talaat [8] on Pelargonium graveolens plant and Nahed [9] on Salvia farinacea plants, found that amino acids significantly increased vegetative growth. Karima and Abd EL-Wahed [10] on chamomile plants (Chamomilla recutita L.), revealed that all treatments of ornithine, proline and phenylalanine led to significant increases in the plant height, number of branches, number of flowers per plant, fresh and dry weight of herb and flowers, the effect was more pronounced with 50 mgL-1 ornithine, 100 mgL-1 proline or phenylalanine. Moreover, the same authors found that, essential oil percent and yield increased by all treatments of the three amino acids at all cuttings. Wahba et. al. [11] reported that application of all amino acids forms significantly increased the determined parameters (plant height,

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number of branches, fresh and dry weight of herb, and yield of seeds) as well as chemical composition(total carbohydrate, total lipid content and total caffeic acid derivatives) in herb and seeds of plant as compared to untreated plants. The most effective treatment was application of tryptophan at 100ppm dose

Little information is available about the role of vitamins in regulating the biosynthesis of essential oil in plants. Robinson [12] reported that vitamin B complex and vitamin C act as co-enzymes in the enzymatic reactions by which carbohydrates, fats and proteins are metabolized and involved in photosynthesis and respiration. Ascorbic acid (vitamin C) is known as a growth regulating factor which influences many biological processes. Price [13] reported that ascorbic acid increased nucleic acid content, especially RNA. Moreover, it also influenced the synthesis of enzymes, nucleic acids and protein, as well as it acts as coenzyme in metabolic changes. On tomato plants, Abd El-Halim [14] reported that foliar application of ascorbic acid significantly increased growth and yield parameters such as stem length, number of branches, leaves, flowers and fruit set as well as dry weight of shoot per plant comparing with control plants. On fennel plants, Hendawy and Azza [15] found that ascorbic acid at75 mgL<sup>-1</sup> recorded the best value of plant height, number of branches, number of flower heads and seed weight per plant. No significant difference was shown between aspartic acid and ascorbic acid. Phenylalanine at 300 ppm resulted the highest essential oil percent (2.78%) compared with control (2.00%).

This investigation was, therefore, undertaken to examine the influence of foliar applied Phenylalanine, tryptophan and ascorbic acid on yield and essential oil of *Thymus vulgaris L*.

## MATERIALS AND METHODS

Seedlings of *Thymus vulgaris* L family Lamiaceae were obtained from Sekem Co. The experiment was conducted under field conditions during 2012 and 2013, at the Experimental farm of Applied Research Center for Medicinal Plants, National Organization for Drug Control and Research, Egypt .Seedlings were transplanted in 15 Mars. 2012, 2013 at the field plots of loamy sand soil. The seedlings were fertilized the following fertilizers (given per feddan): 250 kg ammonium nitrate (33.5% N), 200 kg calcium super-phosphate (16% P2O5), 50 kg potassium sulphate (48% K2O). The fertilizers were added in three doses; the first dose 15 days before the first cutting of plants, the second and third doses were added after 15 days of the first

and second cuttings, respectively. The recommended cultural practices for Thyme production were applied according to the Egyptian Ministry of Agriculture.

Foliar application of Amino acids and vitamin C (50,100 and 150mg/L for each), were sprayed twice with freshly prepared solutions ( in water) at 1May and 1 August in each season, as well as untreated plants (control; distilled water). The spraying was done manually using a spraying bottle. Untreated plants sprayed with distilled H<sub>2</sub> O which serves as control. The experimental treatments were laid out in a randomized complete block design (RCBD) with 3 replicates. Mechanical and chemical analysis of the soil were carried out before sowing and presented in Table (1). Two cuts were taken during the growing seasons (the first one on 30 Jun and the second on 30 Sep 2012 and 2013, respectively). The following parameters were recorded at each cut: plant height (cm), fresh and dry weight (g/plant), N. of Branches/ plant, essential oil percentage and the chemical composition of the volatile oil. The percentage of the volatile oil of air dried herb for each treatment was determined by hydro-distillation of vegetative parts of plants (100 g) of each treatment for 3h according to British Pharmacopoeia [16] using 6890 series GC-system. The oils were dried over anhydrous sodium sulphate and kept at -4°C until it was analyzed. The volatile oils compositions were performed with GC equipped model 6890 Hewlett- Packard (Pittsburgh, PA, USA) with FID detector on a split injector. HP5 capillary column  $(25 \text{ m} \times 0.12 \text{ }\mu\text{m})$  was used with the injector and detector temperature maintained at 225 C and 275 C, respectively. The oven temperature was programmed from 60 C to 300 C at 20 C/min. The carrier gas was nitrogen at a flow rate of 1 ml/min. 2 µL of sample was injected by hand and in the split mode (1:20). Volatile oil contents were identified by comparison of their retention times with those of the reference standards.

#### **RESULTS AND DISCUSSION**

**Analysis of variance:** Analysis of variance for sex quantitative characters related to growth parameters, essential oil content (%) and yield are shown in Tables (2&3). In both seasons differences between treatments were highly significant at 5% with some exceptions such as essential oil percentage.

#### Vegetative growth

**Plant height:** *Thymus vulgaris L.* plant treated with phenylalanine, tryptophan or ascorbic acid at different concentrations caused a significantly increment of plant height (cm) compared with

untreated plants (Tables,4 and 5). Concentrations of phenylalanine were more effective than other treatments. The best treatment for plant height was resulted with phenylalanine at 150 mg / L. which recorded 29.57 and 33.21cm at  $1^{st}$  season as well as 34.44 and 37.71 cm at  $2^{nd}$  season for  $1^{st}$  and  $2^{nd}$  cuts, respectively.

Number of branches: It is obvious from data in Tables (5 and 6) that, all amino acids (Phenylalanine and tryptophan) as well as ascorbic acids significantly increased number of branches/plant at medium and high concentration (100 and 150ppm) in both seasons.. The amino acid sources and ascorbic acid and their concentrations showed that phenylalanine at 150ppm produced the most promising effect on branching of Thymus vulgaris L plants. There no significant differences between the effect of phenylalanine at 150 mg / L and tryptophan at the same concentration on number of branches / plant.

**Fresh and dry weights of herb:** Fresh weight of herb, significantly responded to phenylalanine tryptophan and ascorbic acid foliar application at different concentrations compared with untreated plants. Phenylalanine at 150 mg / L was superior to the other treatments in biomass production giving the highest mean values which recorded 65.33g and 72 g/ plant at 1<sup>st</sup> season as well as 77.33 and 80.33 g/ plant at 2<sup>nd t</sup> seasons for 1<sup>st</sup> and 2<sup>nd</sup> cuts, respectively (Tables 5 and 6). Results of herb dry weight g/plant gave the same trend as mentioned for fresh weight with exception for 1<sup>st</sup> season.

Foliar application of different concentrations of either ascorbic acid or amino acids stimulates a gradual increase in growth parameters. Similar results were obtained by Balbaa and Talaat [17] who concluded that phenylalanine treatments significantly promoted plant height, number of branches, fresh and dry weights of rosemary plants. Abd El-Aziz and Balbaa [18] indicated also that foliar application of tyrosine significantly increased plant height, number of leaves and branches, fresh and dry weights of branches and shoots and stem diameter in both cuttings of Salvia farinacea Plants. It was recorded that application of certain amino acids significantly increased the vegetative growth of Chrysanthemum [19], peppermint plant [5], datura plant [20] and *Pelargonium* graveolens [8]. Furthermore, salicylic acid caused significant increases in most growth parameters of different plant species.

**Essential oil percentage and yield (ml / plant):** Data shown in Table (6) indicate that all treatments significantly increased oil percentage during the 1<sup>st</sup> season where phenylalanine at 150 mg / 1 and ascorbic acid at 150 mg / 1 gave the maximum mean values of essential oil % which recorded 1.92 and 2.03 % in the  $1^{st}$  and  $2^{nd}$  cuts, respectively. In the 2<sup>nd</sup> cut of first season, there is no significant difference between ascorbic acid at 150 mg / 1 and phenylalanine at 150 mg / l. which recorded 2.03 and 2.02 %, respectively. On the otherhand, all treatments had no significant effect on essential oil % during 2<sup>nd</sup> season (Table 6). In the 2<sup>nd</sup> season, phenylalanine at 150 mg / 1 caused the highest mean values of essential oil % which recorded 2.11 and 2.32 % for 1st and 2nd cuts, respectively. Regarding the effect of amino acids and ascorbic acid treatments on essential oil vield (ml / plant), data tabulated in Table (6) indicated that there is a pronounced and significant effect on essential oil yield (ml / plant) during both seasons. The maximum value of this character was obtained as a result applying phenylalanine at 150 ppm for 1<sup>st</sup> cut (0.395 ml / plant) of first season as well as 1<sup>st</sup> cut (0.582 ml / plant) and 2<sup>nd</sup> cut (0.704 ml / plant) of  $2^{nd}$  season. On the other hand, the highest mean value of essential oil yield reached to its maximum value (0.520 ml / plant) as a result of applying tryptophan at 150 mg / 1 for  $2^{nd}$  cut of  $1^{st}$  season.

Amino acids accumulation in plants plays different roles such as osmolyte. Regulation of iron transport, modulating stomata opining and detoxification of heavy metals. Moreover, amino acids affect synthesis and activity of some enzymes, gene expression and redox-homeostasis [25]. Many investigators such as Gamal El-Din et. al.[4], Karima and Abd EL-Wahed [10], Talaat and Youssef [26], Salamon and Honcariv [27] and Omer et.al.[28], mentioned that, foliar application of amino acids significantly increased essential oil percentage and yield. Also, Taraf et.al.[29] on lemongrass, Youssef and Talaat [30] on rosemary, Eid et.al. [31] on Jasminum grandiflorm and Safaa et.al. [32] on geranium reported that foliar application of ascorbic acid caused pronounced increment in the percentage and yield of essential oil

Correlation coefficient between growth parameters and essential oil: The correlation coefficients among the some growth characters as well as essential oil (percentage and yield) are given in Tables (7 and 8). Results were almost the same for the two seasons with slight differences. A character by character examination showed that different characters were differentially associated with each other. All the coefficients of correlation were positive and significant. All growth parameters had a positive and significant correlation with essential oil (percentage and yield) especially fresh and dry weight of herb [33].

Chemical composition of essential oil: The essential oil composition varies according to cuts and / or different treatments and was characterized by a high percentage of oxygenated compounds ranged from 61.99 to 68.76 %. The components of the essential oil in herb for different treatments during 2 cuts during both seasons were shown in Tables (9 and 10). The identified components were 11 compounds representing about 92.04% - 98.55 % as a result of different treatments during both seasons. Thymol was identified as the major compound in the different treatments ranging from 48.98 % to 53.83%. p-Cymene, the second main component, ranged 19.98% - 22.50 % in the essential oil followed with Linalool which was identified as the third main constituent in the essential oil and its relative percentage accounted for3.98% - 5.54%. Data tabulated in Table (9) indicate that phenylalanine at 150 mg / 1 gave the

maximum relative percentage of Thymol (52.32 and 53.51% for 1<sup>st</sup> and 2<sup>nd</sup> cuts, respectively during 1<sup>st</sup> season). In the 2<sup>nd</sup> season, phenylalanine at 100 mg / l resulted in the highest relative percentage of thymol which recorded 51.23 and 51.44% in the 1st and 2<sup>nd</sup> cuts, respectively (Table, 10). Concerning the effect of these treatments on p-cymene, data in Table (9) indicated that p-cymene reached to its maximum values (22.15 and 22.50 % for 1st and 2nd cuts, respectively) as a result of phenylalanine at 150 mg / 1 during the  $1^{st}$  season. On the other hand, tryptophan at 150 mg /l and phenylalanine at 50 mg /l gave the highest relative percentage of p-cymene which recorded 20.72 and 22.26 % in the 1st and 2nd cuts of 2<sup>nd</sup> season, respectively. The effect of different treatments on essential oil and its constituents may be due to its effect on enzyme activity and metabolism of essential oil production.

Table (1) Mechanical and Chemical Analysis of the Experimental Soil

Characteristi	cs											
Mechanical		Chemic	al analy	/sis								
analysis		Soluble		Soluble anion		Macro	Macro		Micro		EC	CaCO3
			cations		(m.equ/L)		(ppm)	elements			m.mohs/cm	
	(m.equ/	L)	_		· · · · ·		(ppm)					
Coarse	16.9	Ca+	1.35	HCO	0.74	Total N	9.5	Fe	3.3		0.57	2.97
sand%												
Fine	63.5	Mg++ 0.79		Cl	1.62	$P_2O_5$	4.8	Cu	035	8.3		
sand%		Ũ										
Silt %	3.7	Na+	2.1	SO	3.25	K <sub>2</sub> O	4.33	Za	094			
Clay %	9.01	1.4	1.54					Mn	4.7			
Soil	Loam	y sand										
texture												

 Table (2) Analysis of Variance for Amino Acids and Ascorbic Acids on Growth Characters and Essential

 Oil of *Thymus vulgaris* (1<sup>st</sup> Season)

Source of	df	Mean squares														
variation		Plant	Branches	Fresh	Dry	Oil %	Oil yield									
		height	No	weight	weight		-									
		1 <sup>st</sup> Cut	1 <sup>st</sup> Cut													
Replication	2	0.70	0.30	0.70	0.70	4.3e-4 8e-7										
Treatments	9	34.25***	28.51***	359.6***	47.11***	0.183***	0.027***									
Error	18	1.03	0.97	1.03	1.03	1.3e-4	3.06e-6									
CV%		4.08	6.31	2.13	5.69	0.74	0.62									
		2 <sup>nd</sup> Cut														
Replication	2	1.20	1.60	1.60	1.60	2.5e-4	3.03e-6									
Treatments	9	38.66***	59.61***	335.1***	59.6***	0.166ns	0.042***									
Error	18	0.98	0.71	0.82	0.71	0.001	7e-7									
CV%	CV%		4.10	1.62	4.10	1.81	0.23									

df= degree of freedom CV= standard deviation

 Table (3) Analysis of Variance for Amino Acids and Ascorbic Acids on Growth Characters and Essential

 Oil of *Thymus vulgaris* (2<sup>nd</sup> Season)

Source of	df	Mean squares									
variation		Plant	Branches	Fresh	Dry	Oil %	Oil yield				
		height	No	weight	weight						
		1 <sup>st</sup> Cut									
Replication	2	0.70	1.66	0.10	0.27	5.7e-4					
Treatments	9	54.83***	51.35***	429.56***	58.09***	0.14ns	0.05***				
Error	18	1.03	0.94	0.99	1.03	0.20	0.004				
CV%		3.60	5.23	1.85	4.66	25.52	15.49				
		2 <sup>nd</sup> Cut									
Replication	2	2.10	3.10*	0.40	0.30	0.01	0.001				
Treatments	9	61.06***	50.23***	360.68***	67.91***	0.195ns	$0.07^{***}$				
Error	18	0.77	0.77	1.07	1.08	0.224	0.005				
CV%		2.84	3.89	1.67	4.28	24.36	14.14				

df= degree of freedom CV= standard deviation

Table (4): Effect of Amino acids & Ascorbic acid on growth and yield of *Thymus yulgaris* in the First Season

reatments	Plant	height	Numbe	er of	Herb	fresh	Herb d	ry wt.					
	(c	m)	Bran	ches	wt	Ξ.	(g/plant)						
					(g/p]	Lant)							
	1 <sup>st</sup> cut	2 <sup>nd</sup> Cut											
Control	20.22	23.55	11.61	13.55	30.51	38.33	10.33	12.55					
Phenylalanine	23.43	25.77	14.33	16.58	44.25	50.55	14.22	17.83					
50 mg/L													
100 mg/L	27.44	29.33	16.87	18.33	48.52	55.87	17.88	20.44					
150 mg/L	29.57	33.21	20.66	22.45	65.33	72.33	20.55	24.33					
Tryptophan	22.42	24.44	12.11	14.55	37.55	46.88	15.33	16.34					
50 mg/L													
100 mg/L	24.88	28.33	15.33	18.77	43.21	58.4	19.55	21.33					
150 mg/L	28.33	31.52	19.44	24.21	58.34	68.44	21.33	26.42					
Ascorbic acid	20.34	22.81	12.32	16.35	42.77	48.34	15.54	17.57					
50 mg/L													
100 mg/L	24.36	28.14	15.75	20.48	44.36	53.77	20.55	23.35					
150 mg/L	28.28	31.23	17.33	23.85	61.42	65.48	23.36	25.41					
LSD at 0.05	1.74	1.70	1.69	1.77	1.74	1.55	1.74	1.45					
CV%	4.08	3.55	6.31	5.46	2.13	1.62	5.69	4.10					

Table (5): Effect of Amino acids & Ascorbic acid on growth and yield of <i>Thymus vulgaris</i> in the second season												
Treatments	Plant	height	Numbe	er of	Herb	fresh	Herb dry wt.					
	(c	m)	Bran	ches	wt	Ξ.	(g/plant)					
					(g/p]	Lant)						
	1 <sup>st</sup> cut	2 <sup>nd</sup> Cut										
Control	23.62	25.67	13.66	16.52	39.41	44.37	14.35	17.52				
Phenylalanine	26.48	28.38	17.37	19.78	49.27	58.45	18.28	21.63				
50 mg/L												
100 mg/L	30.48	32.45	20.57	23.35	58.72	68.27	24.28	27.48				
Tryptophan	34.44	37.71	24.61	28.41	77.33	80.33	27.57	30.35				
50 mg/L												
100 mg/L	24.47	26.64	14.15	18.52	42.51	49.86	18.31	19.34				
150 mg/L	27.38	31.38	19.37	24.71	53.21	66.54	22.34	24.37				
Ascorbic acid	34.37	36.72	25.41	27.27	60.34	69.46	25.37	28.62				
50 mg/L												
100 mg/L	22.38	24.85	14.37	18.33	44.73	51.32	17.55	18.55				
150 mg/L	27.34	30.11	16.77	22.43	45.38	59.72	23.57	25.38				
LSD at 0.05	1.74	1.50	1.67	1.50	1.71	1.77	1.74	1.78				
CV %	3.60	2.84	5.23	3.89	1.85	1.67	4.66	4.28				

Tabl	e (6) Effect o	of Amino aci	al Oil % and	Yield of Thy	mus vulgaris				
		1 <sup>st</sup>	Season			2 <sup>nd</sup>	Season		
Treatments	Essentia	al Oil %	Essent	ial Oil	Essenti	al Oil %	Essential Oil		
			Yi	eld			Yield		
			(ml/	plant			(ml/p	lant	
	1 <sup>st</sup> cut	2 <sup>nd</sup> Cut							
Control	1.12	1.34	0.116	0.168	1.42	1.64	0.204	0.287	
Phenylalanine	1.43	1.65	0.203	0.294	1.63	1.72	0.298	0.372	
50 mg/L									
100 mg/L	1.76	1.88	0.315	0.384	1.79	1.93	0.435	0.530	
150 mg/L	1.92 2.02		0.395	0.491	2.11	2.32	0.582	0.704	
Tryptophan	1.34	1.53	0.205	0.250	1.56	1.73	0.279	0.338	
50 mg/L									
100 mg/L	1.53	1.87	0.299	0.399	1.68	1.93	0.375	0.470	
150 mg/L	1.83	1.97	0.390	0.520	1.95	2.21	0.495	0.632	
Ascorbic acid	1.34	1.55	0.208	0.286	1.51	1.72	0.266	0.320	
50 mg/L									
100 mg/L	1.53	1.78	0.314	0.396	1.77	1.94	0.418	0.492	
150 mg/L	1.59	2.03	0.358	0.508	1.93	2.31	0.507	0.681	
LSD at 0.05	0.02	0.05	0.003	0.001	ns	Ns	0.103	0.117	
CV %	0.74	1.81	0.619	0.226	25.52	24.36	15.49	14.14	

Table (7):Correlation coefficients among different agromorphological and essential oil traits in Thymus														
vulgarisl plant for first sea	son (upper va	lues in each ce	ll for 1st cut a	and lower valu	es in each cel	1 for 2 <sup>nd</sup> cut)								
Traits	X1	<u>K1 X2 X3 X4 X5 X6</u>												
Plant height X1														
Number of branches X2	0.96***													
	0.91***													
Herb fresh weight X3	$0.90^{***}$	0.92***												
	$0.94^{***}$	0.93***												
Herb Dry weight X4	0.83**	$0.80^{**}$	$0.82^{**}$											
	$0.89^{***}$	$0.98^{***}$	0.93***											
Oil % X5	0.93***	0.96***	$0.88^{***}$	0.77**										
	$0.94^{***}$	$0.92^{***}$	0.96***	$0.95^{***}$										
Oil yield X6	0.94***	0.93***	0.91***	0.94***	0.94***									
-	0.93***	$0.98^{***}$	$0.97^{***}$	0.99***	$0.98^{***}$									

Table (8):Correlation coefficients among different agromorphological and essential oil traits in Thymus													
vulgarisl plant for second	season (upper	values in each	cell for 1st cut	and lower valu	ues in each cell	for 2 <sup>nd</sup> cut)							
Traits	X1	X2	X3	X4	X5	X6							
Plant height X1													
Number of branches X2	$0.97^{***}$												
	$0.97^{***}$												
Herb fresh weight X3	$0.87^{***}$	0.85**											
	$0.94^{***}$	0.96***											
Herb Dry weight X4	$0.90^{***}$	0.86**	0.86**										
	$0.96^{***}$	0.96***	0.96***										
Oil % X5	0.92***	0.85**	0.93***	0.95***									
	$0.86^{**}$	$0.90^{***}$	0.91***	$0.86^{**}$									
Oil yield X6	0.89***	0.83**	0.91***	0.97***	0.99***								
	$0.90^{***}$	$0.92^{***}$	$0.90^{***}$	$0.92^{***}$	$0.98^{***}$								

Table (9)	Table (9) Effect of Effect of Amino acids on The chemical composition of Volatile oil of Thymus vulgaris Plant in the First season																			
Compounds	Cont.		Phen1		Phen	2	Phe	n3	Tryp1		Tryp	2	Tryp3	3	Asc 1	·	Asc2		Asc3	
									• •		• -		• •							
	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	$2^{nd}$	1 <sup>st</sup>	$2^{nd}$	1 <sup>st</sup> cut	$2^{nd}$	$1^{st}$	$2^{nd}$	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup> cut	$2^{nd}$	1 <sup>st</sup>	2 <sup>nd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup>
	cut	cut	cut	cut	cut	cut	cut	cut		cut	cut	cut	cut	cut		cut	cut			cut
α - Pinene	0.80	1.00	0.71	0.82	1.02	1.11	1.18	1.21	0.73	0.83	0.73	0.97	1.17	1.20	1.09	1.21	1.08	1.31	1.11	1.41
Myrecen	2.81	3.01	2.01	2.13	2.42	2.63	2.97	3.35	2.92	3.02	2.52	2.75	2.98	3.14	2.49	2.60	2.51	2.92	3.10	3.01
P -Cymene	20.91	21.03	20.85	21.95	20.72	21.63	22.1	22.50	20.51	20.87	20.4	21.06	21.48	21.82	20.98	21.26	20.95	21.94	20.55	22.0
							5				3									
γ- Terpinene	1.87	2.1	1.92	2.11	1.87	2.01	2.20	2.54	1.98	2.03	2.58	2.28	1.96	2.32	1.98	2.15	1.98	2.31	2.08	1.97
Linalool	5.25	5.40	4.69	4.88	4.72	4.93	5.23	5.54	4.78	4.92	4.49	4.89	5.05	5.50	3.98	4.13	4.33	5.13	4.34	4.95
Camphor	0.74	0.91	0.73	0.99	0.43	0.61	0.82	1.03	0.78	0.99	1.22	1.56	0.73	0.93	0.78	0.95	1.10	0.83	0.89	1.04
Boroneol	2.35	2.45	2.43	2.87	2.54	2.77	2.97	3.01	2.57	2.75	2.91	3.10	2.77	2.81	2.89	2.94	2.43	3.12	2.59	2.85
Terpineol	1.52	1.84	1.41	1.68	1.45	1.53	1.78	1.85	1.31	1.65	1.93	2.20	1.65	1.73	1.59	1.56	1.44	2.01	1.57	2.24
Thymol	48.98	49.15	49.87	49.95	50.83	51.14	52.32	53.51	50.21	49.88	49.48	49.74	49.82	50.35	48.98	49.53	50.73	51.44	50.34	51.31
Carvacrol	3.22	3.43	2.86	3.11	2.92	3.28	3.54	3.82	2.81	3.12	3.43	2.98	3.28	3.62	2.98	3.15	3.36	3.58	3.11	3.12
Caryophyllene	3.86	3.92	3.74	3.93	3.25	3.65	4.02	4.31	3.41	3.69	3.87	4.12	3.88	4.11	3.39	3.43	3.14	4.06	3.38	3.32
Non-	30.25	30.42	29.23	30.58	29.28	31.00	28.95	29.19	29.58	30.44	30.13	31.18	31.47	32.59	29.93	30.65	29.66	28.46	30.22	31.71
Oxygenated																				
compounds																				
Oxygenated compounds	62.06	63.18	61.99	63.48	62.89	64.26	66.66	68.76	62.43	63.31	63.46	64.47	63.30	64.94	61.20	62.26	63.33	66.11	62.84	65.51
Total compounds	92.31	93.60	91.22	94.42	92.17	95.29	95.61	97.95	92.01	93.75	93.59	95.65	94.77	97.53	91.13	92.91	92.99	94.57	93.06	97.22

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Table (10	Table (10) Effect of Effect of Amino acids on The chemical composition of Volatile oil of Thymus vulgaris Plant in the Second																			
									Se	ason										
Compounds	Cont.		Phen	l	Phen	2	Phen	3	Tryp1	_	Tryp2	2	Tryp3		Asc 1		Asc2		Asc3	
					d at and												d at l and		d at and	
	1 <sup>st</sup>	$2^{nd}$	1 <sup>st</sup>	$2^{nd}$	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	$2^{nd}$	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	$2^{nd}$	1 <sup>st</sup>	$2^{nd}$	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	$2^{nd}$ cut
	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	cut	1.01
α - Pinene	1.00	1.10	1.13	1.03	1.16	1.31	1.19	1.21	0.96	1.10	0.92	1.12	1.23	1.41	1.11	0.99	1.12	1.32	0.89	1.21
Myrecen	2.95	3.12	2.02	2.41	2.71	2.92	3.11	3.51	3.11	3.11	2.82	2.93	3.15	3.30	2.28	2.75	2.62	2.64	2.78	3.13
P -Cymene	21.01	21.13	21.55	22.26	21.22	21.94	21.14	21.12	20.71	21.03	20.72	21.21	21.75	22.02	19.98	20.56	21.51	22.01	20.89	21.82
γ - Terpinene	2.03	2.30	2.05	2.21	2.14	2.31	2.22	2.43	2.18	2.28	2.31	2.52	2.11	2.41	1.99	2.21	1.87	2.02	1.94	2.38
Linalool	5.25	5.40	4.52	4.82	4.93	5.13	4.82	5.13	4.98	5.12	4.31	5.01	4.53	4.82	3.98	4.53	4.09	4.32	4.13	5.16
Comphor	0.79	0.94	0.94	1.12	0.73	0.83	1.02	1.23	1.02	1.18	1.32	1.71	1.07	1.14	1.44	0.94	1.28	1.24	1.18	1.34
Boroneol	2.45	2.70	2.64	3.12	2.86	3.12	2.98	3.11	3.86	3.02	3.11	3.42	3.13	3.16	2.77	3.02	2.98	3.37	2.65	2.98
Terpineol	1.80	2.20	1.65	1.82	1.75	2.01	1.88	2.04	1.66	1.84	2.21	2.43	2.00	2.04	2.18	2.31	1.86	2.12	2.08	2.34
Thymol	49.05	49.00	50.31	50.51	51.23	51.44	50.89	51.05	50.43	50.18	50.01	50.12	50.18	50.63	48.98	49.54	50.96	51.12	50.68	51.04
Carvacrol	3.32	3.21	3.07	3.31	3.16	3.58	3.38	3.71	3.34	3.42	3.32	3.21	3.32	3.13	3.55	2.67	2.43	2.87	3.38	3.23
Caryophyllene	4.12	4.32	3.88	3.61	3.52	4.06	3.66	4.01	3.66	3.79	4.11	4.33	4.00	4.31	3.78	3.55	3.99	3.37	3.41	2.98
N on -	31.11	31.97	29.50	26.70	30.35	32.50	31.32	32.28	29.76	31.31	30.85	30.99	32.24	28.96	29.14	30.06	31.11	31.59	29.91	31.53
compounds																				
Oxygenated compounds	62.66	63.45	63.13	68.31	64.66	66.12	64.97	66.27	65.29	64.76	64.28	67.02	64.23	69.23	62.90	63.01	63.60	65.04	64.10	66.09
Total compounds	93.77	95.42	92.63	95.01	95.85	98.65	96.29	98.55	95.05	96.07	95.16	98.01	96.47	98.19	92.04	93.07	94.71	96.63	94.01	97.62

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