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PRODUCTIVITY AND OIL QUALITY OF *THYMUS VULGARIS* L. UNDER GROWTH PROMOTERS AND SOILLESS CULTURE CONDITIONS

Fatma S.Aboud*, M. H. Mohammed, Nagwa M. ahmed and S. H. Ahmed
Closed Agricultural Methods Research Dept., Central Laboratory for Agriculture Climate, Agriculture Research Center (CLAC), Dokki, Giza, Egypt,
*Email: batta_fatma2@yahoo.com

ABSTRACT

Thyme (*Thymus vulgaris* L.) belongs to the lamiaceae family. It is characterized by its medicinal and economic values. Thymus is grown worldwide for cosmetic and medical use. Plant bio-stimulants are classified as substances which have positive effects on growth and productivity of plants. Bio-stimulants are obtained naturally from various economically and environmentally viable sources. The current experiment aimed to investigate the response of thymus physical and chemical properties to the different types and concentrations of bio-stimulators under soil less culture conditions for two successive seasons 2019 and 2020. Thymus seedlings were planted in pots filled with mixed media of perlite: sand (1:1v/v). Plants were sprayed with aqueous solution of ascorbic acid (1 and 2g/l), aspartic acids (500 and 1000 ppm) and dry yest (2 and 4g/l) and combination of them compared with control. The herp was harvested in mid- July (First cutting) and first week of October (second cut). Growth and yield characters were measured. The essential oil percentage was determined and analyzed by GC in both cuts from the fresh herb. Results showed that, spraying of mixtures of ascorbic acid (2g/l), aspartic acids (1000 ppm) and yeast (4g/l)) significantly increased yield, total carbohydrates, total phenols, total pigments, essential oil percentage and yield as well as its NPK content characters and components of essential oil high contents of thymol, α and β –Pinene during both cuts and for both seasons. Economic evaluation was carried out by calculating with the highest return in the mixture treatment. It is clear that all treatments are economically feasible however, one can find that the maximum return or profit comes from mixture treatments where the benefit to cost ratio were 3.99 and 3.9 for Mix2 and Mix1 respectively.

Keywords **:** *Thymus vulgaris* L., substrate Culture, vitamins, amino acids, active dry yeast, thymol, economic evaluation.

Introduction

Medicinal plants have an important value due to their natural antioxidant and antimicrobial activities, of them is thymus. Many authors confirmed the medicinal and economic importance of thymus (Satyal *et al*., 2016; Aouam *et al*., 2019; Golkar *et al.*, 2020) where it can be consumed locally or exported. Medicinal plants have an important socio-cultural, spiritual and medicinal value in rural and tribal lives (Shinwari, 2005) of them, Thymus plants which belongs to the Lamiaceae family and is characterized by its high nutritional and medicinal value. Thymus is rich in nutrients, thiamine, riboflavin and niacin. Thymus has been reported for its anti-oxidative, antimicrobial, antitussive expectorant, antispasmodic, and antibacterial effects (Sarikurkcu *et al*., 2015; Hashem 2018). Thymus essential oil contains thymol, carvacrol, linalool, α-terpineol, camphor, caryophyllene and γ-terpinene (Amiri, 2012; Fachini *et al*., 2012). Roby *et al.* (2013) reported that methanolic extracts of thyme are sources of phenolic acids, flavonols and flavanones. Natural foods which are rich in flavonoids helps protect from lung and cancers (Sharangi and Guha, 2013). Eqbal and Abdullah, (2017) confirmed the strong antimicrobial properties of thymol oil which found in thymus essential oil.

According to Ministry of Agriculture Statistics, 2018; the average cultivated area of thymus in the open field was 276fadden, with a yield quantity of 552 tons. Thymus plant height ranges between 15 to 30 cm and 40 cm wide (Fernanda *et al.*, 2012).

Ascorbic acid (vitamin C) is essential for plant growth, which is characterized by its antioxidant activity, enzymatic capabilities and promoting power on plant growth. Ascorbic acid is essential in plant photosynthetic activities, cell expansion and resistance to environmental stress, electron transport, plant flowering and senescence, cell death and fighting pathogens (Blokhina *et al*., 2003; Gomez and Lajolo, 2008). Many authors confirmed the basic role of ascorbic acid in timing of flowering and senescence (Ahlam and Mustaf, 2019; Carr, and Lykkesfeldt, 2020). The foliar application of mixtures (garlic extract, ascorbic acid and nicotinamide) significantly increased plant physical and chemical parameters as reported by Mohamed *et al*. (2020) on Faba bean.

Amino acids are the building blocks of proteins. Amino acids are of great importance in plant growth which attribute by a large extend in yield enhancement and plant overall development. Naturally extracted amino acids are an important source of nitrogen that can be directly absorbed by plants, as well as it can be considered as major transport forms of organic nitrogen in plants, which have been identified in different plant species (Guangzhe Yang *et al.*, 2020). Foliar application of amino acids was reported to significantly increase yield and plant growth attributes (Khattab *et al.*, 2012; Wassel *et al.*, 2015 and Kamal, 2017).

Active dry yeast as a biofertilizers is reported to has many promoting effects on plant growth. this is attributed to its richness of cytokinin which promotes cell division and synthesis of amino acids and vitamins in plant fruits (Ahmed *et al.*, 2011). In addition to its ability to release CO_2 into soil, which improves soil characteristics and increases plant photosynthetic activity and pigments formation. Many authors have reported the stimulant activities of yeast, of them are Aly *et al.* (2007) on coriander, Hemdan, (2008) on anise, Dahab *et al.* (2010) on marjoram and Kenawy, (2010) on Hibiscus sabdariffa, L plants and Matter and. El-Sayed (2015) on caraway plants. Active dry yeast foliar application enhanced thyme plants growth and essential oil yield quantity and quality (Heikal, 2005).

Soilless culture has been found to has favorable effects on plant growth and yield quantity and quality (Putra and Yuliando, 2015), off-season crop production (Montagne *et al.*, 2015), introduce new crops which cannot grow on the normal soil (Al-Karaki and Othman, 2009). It improves plant soil interactions and effect on the nutrient availability in the plant root zone which may increase the yield of crops (Montagne *et al.*, 2015). Soilless culture was considered a key alternative in the case of soil borne diseases, which reduces the use of methyl bromide that is used in soil sterilization and led to more environmentally friendly agriculture system (D'Imperio *et al.*, 2018). The type of the used substrate media may significantly affect plant grown (Alsmairat *et al.*, 2018) where media with low bulk density and high-water holding capacity facilitate plants aeration and root penetration which enhance nutrient availability and absorption (Deepagoda *et al.*, 2013). It has been reported that the substrate physical and chemical properties effect the production quality and quantity such as yield, flower size and number, fruit sugar and phenolic compounds (Al-Ajmi *et al.*, 2009; Schwarz *et al.*, 2009; Al-Ajlouni *et al.*, 2017).

The objective of this study was to determine the growth, yield and chemical composition of Thymus vulgaris plant as influenced by the application of different sources of growth regulators in order to improve the yield of herb and essential oil content.

Materials and Methods

This experiment was conducted in the Central Laboratory for Agricultural Climate Research Centre (CLAC), Dokki, Giza, Egypt, under unheated greenhouse conditions for two successive seasons of the years 2019 and 2020.

Plant Material

At the first week of March, the seedlings were transplanted in plastic pots (5 liters in volume) filled with well mixed media of perlite: sand (1:1v/v). It was placed on terraces in the greenhouse with drainage capabilities and irrigated by balanced nutrient solution until they formed true leaves. Three weeks later after transplanting, the plants were sprayed with aqueous solution of different biostimulators (vitamin, amino acid (aspartic acids) and dry yeast). The plants were sprayed three times during each cut, the first spray application was occurred after new leaves, the second after one week from the first and the third one after one week from the second one. This sequence was implemented during the second cut and for the second seasons.

Experimental design

Experiment was carried out on a completely randomized design, with 36blocks arranged in a 1 x 9x 3 factorial schemes (one cultivar, nine concentrations and three replicated).

Experimental treatments

1. Control (Foliar application with water)
2. Ascorbic acid (1 g/L)
3. Ascorbic Acid (2 g/L)
4. Yeast (2 g/L)
5. Yeast (4 g/L)
6. Amino acid (0.5 g/L): (Aspartic acid 500 ppm/L)
7. Amino acid (1 g/L): (Aspartic acid 1000 ppm/L)
8. Mix 1: Foliar application with vitamin at 1 g/l, amino acid (aspartic acids) at 500 ppm and dry yeast at 2 g/l
9. Mix 2: Foliar application with vitamin at 2 g/l, amino acid (aspartic acids) at 1000 ppm and dry yeast at 4g/l

The used nutrient solution was adapted from Cooper solution (Cooper, 1979) depending on the analysis of the local water (El-Be hairy, 1994) as shown in table (a). The desired initial concentration of the nutrient solution was maintained by suitable dilution of the stock solutions with tap water. Electrical conductivity (EC) of the nutrient solution was maintained between (2–2.2) $m.mhos^{-1}$ and pH maintained between (6 - 6.5).

Table a : The chemical composition of nutrient solution used.

Nutrient solution	Macronutrients (ppm)					Micronutrients (ppm)					
	N	P	K	Ca	Mg	Fe	Mn	Zn	B	Cu	Mo
Chemical nutrient solution	200	45	300	180	60	3.0	0.8	0.4	0.5	0.25	0.02

Data sampling and analysis

Sampling: two cuts were harvested, mid- July (First cut)and first week of October (second cut). The plant herbage was harvested by cutting 5 cm above the soil surface and plant growth parameter for the two cuts were recorded as:

Vegetative growth characteristics

- Numbers of branches per plant.
- Plant height (cm).
- Fresh weight (g/herb).
- Dry weight (%): was determined according to A.O.A.C. (1990).

Determination of chemical composition of thymus herbage

Determination of moisture content

The moisture content was calculated according to A.O.A.C. (1990).

Pigments determination

Chlorophyll a, b and carotene were determined according to A.O.A.C. (1990) and Wettstein (1957).

Total carbohydrates

Total carbohydrates were determined according to A.O.A.C. (1990).

Phenolic compounds (mg /100g FW) as catichol

The colorimetric method of Folin-Denis as described by Shahidi and Naczk, (1995)

Essential oil (%):

Quantitative determination of essential oil obtained from different treatments was achieved by hydro-distillation. The distillation of 100 g fresh herb was continued for 2.5 to 3 hours after water boiling till no further increase in the oil was observed. The oil was permitted to stand undisturbed and the amount of oil obtained from plant materials was calculated:

Oil (%) = (observed volume of oil (ml)/weight of sample (g)) × 100.

Essential oil yield

Essential oil (ml/plant) was calculated by multiplying the average fresh weight of plant by the average oil percentage.

Oil yield/plant = plant fresh weigh (g) × oil %.

Mineral contents (N, P, & K) and Crude protein

Total nitrogen was determined according to kejldahl method as A.O.A.C. (1990). Total phosphorus was determined colorimetrically by ascorbic acid reductant method according to Murphy and Riley, (1962) as modified by Watanabe and Olsen, (1965)and total potassium was determined by using flame photometer according to A.O.A.C, (1990).Crude protein is calculated as mineral nitrogen multiplied by the protein factor, which is 6.25.

GLC analysis of the essential oil

The essential oil constituents were identified by Gas Liquid Chromatography (GLC) according to Radwan (1978)

Economic evaluation:

Total cost is the sum of total fixed cost and total variable cost (L.E/m^2), where total fixed cost is the sum of the cost of(substrate (sand+perlite) + plastic pots + nutrient solution +irrigation + power) and total variable cost is the sum of the cost of (seedlings + biostimulators + labours). Total revenue was calculated as the sum of the herb fresh weight for the two seasons multiplied by the number of plants in one square meter multiplied by the average market price (L.E./kg). The following table (b) illustrates the market price of the experiment constituents by Egyptian pound as recorded during the years of 2019 and 2020.

Table (b) : Price of the experiment constituents (L.E.)

Item	Cost (L.E.)	Item	Cost (L.E.)
Pot	2.5	Seedling	1
Sand (L)	0.30	Nutrient Solution (L)	12
Perlite (L)	1.80	Power (m^2)	10
Irrigation system (m^2)	54	Irrigation (m^2)	10
Tank	50	Ascorbic Acid (1 g)	1
N. plant/m^2	16	Yeast (1 g)	0.37
Thymus Market prices kg/FW	100	Amino acid (1g)	5

- Net return = Revenue (L.E/m^2)- Total cost (L.E/m^2)
- The revenue to total Cost (B/C) ratio was calculated to represent the profit percentage.

Statistical analysis

The statistical significance of observed differences among treatment means was evaluated by analysis of variance (ANOVA). Statistical analysis was made using the “agricolae” package in R software program according to (Duncan, 1955, R, 2017).

Results

The present study has put stress on some biochemical constituents to obtain good idea about the suitable condition for cultivating thymus plant. The bio-stimulatory influence of amino acids (AA), vitamins (Vits), yeast and mixtures combination of them (Mix1 and Mix2) on the different parameters of thymus grown under soilless culture conditions have been investigated herein after:

- Morphological characteristics: plant height (cm), No. of branches /plant, Fresh weight (g/plant), essential oil % and essential oil (g/herp).
- Chemical characteristics: dry weight%, moisture %, chlorophyll a, b and carotenes (mg/g FW), total pigment, carbohydrate %, total phenols (mg/100 g FW), N, P and K (%) and crud protein.

Effect of different growth promoters on vegetative growth

All treatments have exerted a significant positive influence on plant height, No. of branches /plant , plant fresh and dry weight and moisture content in comparison with the control treatment, which showed the lowest values as illustrated in table (1) and figure (1). Most of these differences did not reach level of significancy among each other. The highest values of plant height, No. of branches/plant and fresh weight have been attained in case of spraying with the highest concentrations of Vit. C, amino acids and yeast mixtures (Mix2). While, mixture of low concentration (Mix1) came at the second order in enhancing the vegetative growth. Plant height reached approximately about 1.42 and 1.32 times the corresponding control in the first and second seasons respectively. While, No. of branches /plant were approximately (1.56 and 1.59) times the control treatment in the first and the second seasons respectively. Foliar application of Mix2 led to an increase in plant fresh weight by 1.63 and 1.53 g/herb for the two seasons respectively. Figure (1) represents the plant productivity response to the different treatments where the mixture treatments were the highest during the two studied seasons.

Table 1 : Effect of different growth promoters on vegetative growth on Thymus Under Soilless Culture Conditions

Harvest	**height (cm)**		**N.of branches /plant**		**fresh weight (g/herb)**		**% Dry weight**		**% moisture**	
	1st	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**
Treatment	First season									
Control	57.67 ^{G}	61.67 ^{G}	5.33 ^{G}	15.00 ^{D}	88.04^{H}	215.06 H	20.99 ^{E}	21.29 F	76.13 E	77.57 F
Ascorbic acid (1 g/L)	69.61 ^{DE}	74.33 ^{DE}	7.00 ^{DE}	17.00 ^{BC}	108.00 ^{EF}	246.2 EF	23.42 ^{BCDE}	24.66 CDE	78.31 BCD	80.43 D
Ascorbic Acid (2 g/L)	74.95 ^{BC}	78.78 ^{BC}	8.00^{BC}	18.00 ^{AB}	126.17^{C}	274.75 BC	25.66^{B}	27.41 ABC	79.66 B	82.29 BC
Yeast (2 g/L)	63.39 ^{F}	67.78 ^{F}	6.00 ^{FG}	16.00 ^{CD}	95.17G ^{H}	225.5 GH	21.88 ^{DE}	23.16 EF	77.00 DE	78.04 EF
Yeast (4 g/L)	73.89 ^{BC}	75.78 ^{CD}	8.00 ^{BC}	18.00 ^{AB}	120.41 ^{CD}	265.77CD	24.63 ^{BC}	26.19 BCD	79.54 B	82.03 BC
Amino acid (0.5 g/L)	67.33 ^{EF}	71.33 ^{EF}	6.33 ^{EF}	16.22 ^{C}	100.25 ^{FG}	233.92 FG	22.41 ^{CDE}	23.78 DEF	77.64 CD	79.12 E
Amino acid (1 g/L)	71.56 ^{CD}	74.78 ^{CDE}	7.33 ^{CD}	17.00 ^{BC}	112.70 ^{DE}	258.17 DE	24.21 ^{BCD}	25.60 BCDE	78.52 BC	81.47 CD
Mix 1	76.97 ^{B}	81.94 ^{B}	8.89 ^{B}	18.00 ^{AB}	139.83 ^{B}	282.08 B	26.05 ^{B}	27.77 AB	81.36 A	82.86 AB
Mix 2	83.08 ^{A}	86.90 ^{A}	10.00 ^{A}	18.67 ^{A}	162.75 ^{A}	302.5A	30.19 ^{A}	30.12 A	81.48 A	83.51 A
Treatment	Second season									
Control	66.60 F	66.33 G	5.00 E	14.33 D	96.26H	226.8 G	21.92 E	20.89 F	77.21 F	77.37 D
Ascorbic acid (1 g/L)	71.96 E	75.47 EF	6.00 DE	16.00 BC	122.18 EF	247.41 DE	24.26 CDE	23.47 DE	80.13 CDE	79.22 BC
Ascorbic Acid (2 g/L)	80.22 BC	83.58 BC	7.67 BC	17.33 A	150.98 BC	273.69 B	26.04 BC	26.23 BC	82.75 A	81.17 A
Yeast (2 g/L)	69.45 EF	71.62 F	5.33 E	15.00 CD	104.72 GH	235.56 FG	22.57 E	22.07 EF	78.43 EF	78.17 CD
Yeast (4 g/L)	78.45 CD	81.20 CD	7.00 CD	17.00 AB	138.09 CD	262.02 C	25.41 BCD	25.00 CD	81.57 ABC	80.36 AB
Amino acid (0.5 g/L)	70.84 E	73.94 EF	6.00 DE	16.00 BC	113.50 FG	240 EF	23.33 DE	22.90 DEF	79.44 DE	78.30 CD
Amino acid (1 g/L)	75.37 D	77.57 DE	7.00 CD	16.89 AB	127.98 DE	252.87 CD	24.94 BCD	24.42 CD	80.51 BCD	79.38 BC
Mix 1	81.86 B	86.69 AB	8.22 B	18.00 A	159.31 AB	283.16 AB	26.79 B	27.38 AB	83.18 A	81.58 A
Mix 2	86.08 A	88.87 A	9.67 A	18.00 A	171.43 A	292.91 A	30.82 A	29.33 A	82.02 AB	81.51 A

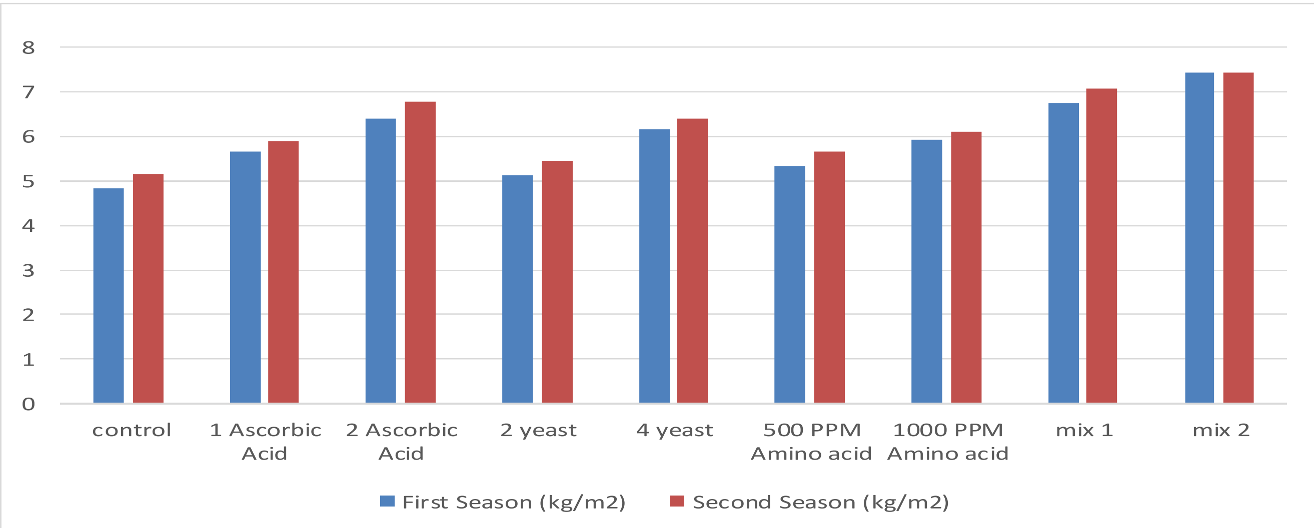

Fig. 1 : Productivity (kg/m2) of thymus herb for the two seasons

Concerning dry weight (%) and moisture content (%) of thymus plants, data illustrated in table (1) reveals that, the highest values recorded were due to the application of Mix treatments with a significant difference compared to all other treatments including the control one. This was followed by the application of ascorbic acid which was significantly different from the control treatment but, did not reach the level of significance compared to the other treatments.

Effect of different growth promoters on Essential oil %, essential oil (g/herp), total carbohydrates and total phenol:

It is well known that the economic value of thymus plant is attributed to its essential oil content therefore we have put stress on the positive variations that may be resulted due to cultivating thymus plant in perlite: sand (1:1v/v) soilless culture, and the regulatory effect of growth promoters (AA, vit. C, yeast and their mixtures). Thus, the evaluation and compression among all treatments were based on essential oil percentage together with its yield (ml oil/plant fresh weight). Data obtained are listed in table (2) for the first and second cut during the two studied seasons. Data reveals that thymus plants responded positively to the application of all biostimulators however this increase in the essential oil content did not reach the level of significancy compared with the control treatment. The highest significant increase in the oil content was a result of the mix application followed by the ascorbic acid treatment. This holds true for the two cuts during the two seasons.

Table 2 : Effect of different growth promoters on Essential oil, Total carbohydrates and Total phenolon Thymus Under Soilless Culture Conditions

Harvest	Essential oil %		Oil (g/herp)		%Total Carbohydrates		Total Phenols	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Treatment	Frist season							
Control	0.19 F	0.26 F	0.17 E	0.36 E	5.50 G	5.56 F	2.37 E	1.91 E
Ascorbic acid (1 g/L)	0.24 DEF	0.37 CD	0.26 CDE	0.65 CDE	6.07 EF	6.24 E	2.76 DE	2.25 DE
Ascorbic Acid (2 g/L)	0.33 BC	0.42 BC	0.41 BC	1.13 BC	7.03 C	7.49 BC	3.75 BC	3.13 BC
Yeast (2 g/L)	0.21 EF	0.3E F	0.20 E	0.45 DE	5.66 FG	5.90 EF	2.49 DE	1.99 E
Yeast (4 g/L)	0.31 BCD	0.4 C	0.37 BCD	0.98 BCD	6.75 CD	7.03 CD	3.27 CD	2.71 CD
Amino acid (0.5 g/L)	0.23 DEF	0.32 DE	0.23 DE	0.54 DE	5.76 EFG	5.99 EF	2.59 DE	2.08 DE
Amino acid (1 g/L)	0.28 CDE	0.38 CD	0.32 CDE	0.82 CDE	6.32 DE	6.44 DE	2.95 DE	2.51 CDE
Mix 1	0.37 B	0.47 B	0.51 B	1.45 B	7.70 B	7.91 AB	4.20 B	3.55 B
Mix 2	0.5 A	0.58 A	0.82 A	2.5 A	8.37 A	8.40 A	5.60 A	4.80 A
Second season								
Control	0.21 F	0.25 F	0.21 F	0.47F	4.35 F	5.56 F	2.84 F	2.28 F
Ascorbic acid (1 g/L)	0.27 CDEF	0.31 CDE	0.33 DEF	0.81 DEF	5.06 EF	6.24 E	3.75 E	3.10 DE
Ascorbic Acid (2 g/L)	0.34 BC	0.37 BC	0.51 BC	1.4 BC	6.81 BC	7.49 BC	4.78 BC	3.75 BC
Yeast (2 g/L)	0.23 EF	0.27 EF	0.24 EF	0.58 EF	4.58 F	5.90 EF	3.35 EF	2.74 EF
Yeast (4 g/L)	0.32 BCD	0.35 BCD	0.45 CD	1.17 CD	6.18 CD	7.03 CD	4.37 CD	3.54 BCD
Amino acid (0.5 g/L)	0.25 DEF	0.30 DEF	0.29 DEF	0.69 DEF	4.81 F	5.98 EF	3.58 E	2.97 DE
Amino acid (1 g/L)	0.30 CDE	0.34 CD	0.39 CDE	0.98 CDE	5.79 DE	6.44 DE	3.92 DE	3.23 CDE
Mix 1	0.39 B	0.40 B	0.62 B	1.76 B	7.18 B	7.91 AB	5.07 B	4.15 B
Mix 2	0.50 A	0.58 A	0.87 A	2.54 A	8.16 A	8.40 A	5.91 A	4.84 A

The higher concentration mixture of the applied biostimulators (Mix2) gave the highest values during the two cuts and the two successive seasons (0.5 and 0.58 %). While the other mixture (Mix1) came in the second order (0.37 and 0.47 %). The difference between Mix2 and Mix1 in essential oil was significantly different with each other and with the control treatment. The lowest essential oil percentage and essential oil (ml/herb fresh weight) was recorded in case of the control treatment (0.19 – 0.26%). This holds true for the two successive cuts during the two seasons. The marvelous results have been achieved by foliage spraying with Mix2, which produced highest significant essential oil % and essential oil (g/herp), which reached 2.63 – 2.2 essential oil% and 4.8 – 4.4 essential oil (g/herp) times than the corresponding control respectively, in the first and second season.

Data presented in table (2) shows the responses of total carbohydrates (%) and total phenols (mg/100g FW) responses due to the foliar spray of the investigated bio-stimulators for the two cuts during both seasons. Generally, all treatments increased the total carbohydrates and phenols in thymus plants. However, this increase did not reach the level of significance in some treatments such as yeast and amino acids treatments regardless the concentration compared to the control treatment. The Mix treatments have excreted a significant increase of both carbohydrates and phenols of thymus plants compared to all other treatments including the control one. But the differences between the two treatments was not almost significantly from cut to another or season to season with the highest value obtained in the case of Mix2 treatment. This holds true for the two cuts during the two studied seasons.

Effect of different growth promoters on GC profile of thymus essential oil:

The GC profile of thymus essential oil reveals the presence of eleven identified compounds of them, ten were existed in relatively minute proportions (minor components). While thymol oil was the major found oil as illustrated in tables (3a and 3b) for the two seasons respectively.

The main features that may be characterized the essential oil investigated in the present study which extracted from thymus plants grown in perlite: sand (1:1v/v) soilless culture, are:

- The superiority of thymol, which varied from one cut to another. The highest value was attained by Mix2 (53.42 – 53.22 %) compared with all other treatments. Whilst the lowest value (44.49 – 46.102%) recorded in the control in the first and second cuts respectively.
- ρ -cymene was the second major constituent in all treatments, and showed variable fluctuations due to growth protomers application. The control recorded (9.85 – 9.84 %) in first and second cut respectively.
- carvacrol low values compared with γ-terpinene and α-Pinene recorded.

Table (3a): Effect of different growth promoters on GC profile of Thymus Essential oil First Season

Treatment	components %	α-Pinene	Sabinene	myrcene	ρ -cymene	α -terpinnene	Borneol	γ-terpinene	thymol	carvacrol	und ecanol	α-Hunulene
Control	1st	1.47	8.16	3.036	9.86	3.39	5.486	7.75	44.49	6.76	3.702	4.49
	2nd	1.16	8.3	5.61	9.84	6.06	5.25	7.17	46.102	6.702	1.341	2.25
Ascorbic acid (1 g/L)	1st	0.86	6.48	2.24	10.82	0.65	4.19	6.02	47.91	8.83	3.73	4.14
	2nd	1.47	6.16	3.03	9.86	1.39	5.48	7.75	48.97	7.76	5.49	3.7
Ascorbic Acid (2 g/L)	1st	0.89	6.56	2.07	9.82	2.55	4.63	7.87	48.22	8.4	4.04	3.47
	2nd	1.39	6.85	2.51	9.87	1.809	5.47	7.98	49.66	6.09	2.051	3.67
Yeast (2 g/L)	1st	0.59	6.26	5.34	8.83	5.57	4.68	7.38	49.42	6.89	2.25	0.745
	2nd	0.811	6.67	2.49	8.18	5.41	6.35	8.09	48.018	5.83	2.11	2.02
Yeast (4 g/L)	1st	0.76	5.88	3.32	8.29	2.32	3.41	8.9	52.04	7.24	0.63	1.35
	2nd	1.4	8.9	3.06	6.29	2.37	6.63	7.3	53.29	6.29	1.82	0.99
Amino acid (0.5 g/L)	1st	0.53	1.912	8.01	9.92	4.78	0.007	7.63	50.3	8.79	0.68	1.24
	2nd	0.48	1.66	10.042	10.29	3.322	0.021	8.23	50.21	6.15	0.63	0.26
Amino acid (1 g/L)	1st	0.74	5.74	1.77	9.4	4.68	6.67	7.38	50.91	6.22	0.61	1.45
	2nd	1.61	8.45	2.7	8.14	7.29	4.59	7.23	51.19	5.27	1.132	1.15
Mix 1	1st	0.86	6.48	2.24	10.82	0.65	4.19	6.024	48.91	8.83	3.73	4.17
	2nd	0.85	5.1	4.47	9.44	5.68	6.04	9.36	50.08	4.89	1.36	1.36
Mix 2	1st	0.75	5.3	6.9	9.3	6.7	4.9	7.14	53.41	5.35	0.14	1.21
	2nd	0.67	5.98	6.47	8.86	6.54	5.07	8.61	53.22	5.39	1.42	1.42

Table (3b): Effect of different growth promoters on GC profile of Thymus Essential oil Second Season

Treatment	components%	α-Pinene	Sabinene	myrcene	ρ -cymene	α -terpinnene	Borneol	γ-terpinene	thymol	carvacrol	und ecanol	α-Hunulene
Control	1st	1.55	2.93	8.03	9.94	4.80	7.20	7.65	44.32	8.81	0.70	1.26
	2nd	1.52	2.90	8.00	9.91	4.77	7.17	7.62	42.40	8.78	0.67	1.23
Ascorbic acid (1 g/L)	1st	1.17	6.58	2.50	9.82	2.68	5.10	7.03	52.31	7.45	3.36	3.05
	2nd	1.17	6.41	2.59	10.17	1.57	4.92	6.93	50.20	8.01	4.19	3.63
Ascorbic Acid (2 g/L)	1st	1.15	6.61	2.39	9.95	1.98	5.01	7.59	48.87	7.50	3.43	3.59
	2nd	1.14	6.67	2.32	9.88	2.11	5.04	7.81	51.23	7.33	3.17	3.58
Yeast (2 g/L)	1st	0.53	1.91	8.01	9.92	4.78	6.76	7.63	50.02	8.79	0.68	1.24
	2nd	0.81	2.67	8.20	8.18	5.41	6.35	7.03	51.34	5.83	2.11	2.02
Yeast (4 g/L)	1st	1.29	1.15	8.77	9.02	5.54	7.52	8.39	50.78	6.55	1.44	2.00
	2nd	1.99	1.87	6.56	9.13	4.60	4.34	8.49	52.82	8.56	1.27	2.14
Amino acid (0.5 g/L)	1st	0.75	3.66	6.77	9.01	4.18	1.66	7.55	51.05	8.17	0.97	1.18
	2nd	0.61	2.66	8.41	9.65	3.75	0.84	7.89	50.63	7.16	0.80	0.72
Amino acid (1 g/L)	1st	0.68	4.20	5.09	9.53	4.21	3.76	7.63	52.03	6.69	0.70	1.08
	2nd	1.18	7.10	2.24	8.77	5.99	5.63	7.31	52.05	5.75	0.87	1.30
Mix 1	1st	0.95	6.08	3.01	10.05	2.81	5.09	7.73	50.23	7.02	2.30	2.58
	2nd	0.89	5.89	3.24	10.10	3.05	5.11	7.71	51.04	6.91	2.01	2.25
Mix 2	1st	0.47	3.76	4.46	6.05	4.41	3.32	5.25	53.45	6.58	3.63	3.33
	2nd	0.63	5.01	5.94	8.07	5.88	4.43	6.25	52.87	7.73	2.89	0.54

Effect of different growth promoters on N, P and K Contents (% of dry matter) and crud protein

Data illustrated in tables (4) show that the effect of different treatments on thymus contents of N, curd protein, P and K %. Data reveals that the Mix and ascorbic acid treatments excreted a significant increase on the herb contents of N, P, K, and crud protein in the first and second cuts during the two studied seasons. The other treatments increased the NPK and crud protein compared with the control treatment however this increase did not reach the level of significance during the two seasons.

The NPK and crud protein content of thymus responded differently to the concentration of the added bioregulators where Mix2 led to a significant increase compared with Mix1. Also, the higher concentration of yeast (4 g/L) led to a significant increase compared with the lower one (2 g/L). this holds true for NPK and crud protein contents for both cuts during both seasons.

Effect of different growth promoters on Chlorophyll a, chlorophyll b, carotene and total pigments content:

All photosynthetic pigments increased due to the application of growth promoters compared with the control treatment however this increase did not reach the level of significance. Data in tables (5) illustrate the response of Chl. a, b, carotene and total pigments to the application of different treatments. Data reveals that the Mix and ascorbic acid treatments excreted a significant increase on the herb contents of all photosynthetic pigments in the first and second cuts during the two studied seasons. the other treatments increased the Chlorophyll a, chlorophyll b, carotene and total Pigments content compared with the control treatment however this increase did not reach the level of significance during the two seasons.

The Chlorophyll a, chlorophyll b, carotene and total Pigments content of thymus responded differently to the concentration of the added bioregulators where Mix2 led to a significant increase compared with Mix1. Also, the higher concentration of yeast (4 g/L) led to a significant increase compared with the lower one (2 g/L). this holds true for the Chlorophyll a, chlorophyll b, carotene and total Pigments content for both cuts during both seasons.

Table 4 : Effect of different growth promoters on Mineral Content on Thymus Under Soilless Culture Conditions

Harvest	% Nitrogen		% crud protein		% Phosphorus		% potassium	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Treatment	Frist season							
Control	1.44F	1.41F	9.02F	8.83F	0.41F	0.50F	1.13F	0.94G
Ascorbic acid (1 g/L)	1.72DE	1.83CD	10.73DE	11.42CD	0.6CDE	0.59DE	1.42CD	1.18DE
Ascorbic Acid (2 g/L)	1.94BC	2.04B	12.10BC	12.75B	0.69B	0.65BC	1.55BC	1.3C
Yeast (2 g/L)	1.49F	1.60EF	9.29F	10.02EF	0.52E	0.55E	1.22EF	1.07F
Yeast (4 g/L)	1.83CD	1.99BC	11.42CD	12.46BC	0.67BC	0.64BCD	1.51C	1.25CD
Amino acid (0.5 g/L)	1.65E	1.70DE	10.29E	10.60DE	0.57DE	0.56E	1.3DE	1.12EF
Amino acid (1 g/L)	1.77DE	1.94BC	11.08DE	12.10BC	0.65BCD	0.62CD	1.46C	1.22CDE
Mix 1	2.05B	2.11B	12.81B	13.19B	0.71B	0.68B	1.65AB	1.41B
Mix 2	2.30A	2.34A	14.35A	14.60A	0.81A	0.78A	1.78A	1.54A
Second season								
Control	1.29 F	1.16 G	8.09 F	7.24 G	0.35 F	0.47 F	1.12 F	1.06 F
Ascorbic acid (1 g/L)	1.51 DE	1.39 DEF	9.41 DE	8.67 DEF	0.44 CDE	0.56 DE	1.57 CD	1.28 DE
Ascorbic Acid (2 g/L)	1.79 C	1.56 BC	11.1 7C	9.76 BC	0.49 BC	0.60 BC	1.77 B	1.59 BC
Yeast (2 g/L)	1.42 EF	1.30 F	8.87 EF	8.11 F	0.38 EF	0.50 F	1.38 E	1.21 EF
Yeast (4 g/L)	1.72 C	1.50 CD	10.7 4C	9.38 CD	0.47 BCD	0.59 BCD	1.72 BC	1.52 BC
Amino acid (0.5 g/L)	1.47 EF	1.32 EF	9.18 EF	8.25 EF	0.41 DE	0.55 E	1.46 DE	1.23 EF
Amino acid (1 g/L)	1.66 CD	1.44 CDE	10.3 6CD	9.00 CDE	0.46 BCD	0.58 CDE	1.67 BC	1.42 CD
Mix 1	2.00 B	1.68 B	12.4 8B	10.5 0B	0.52 B	0.62 AB	1.82 AB	1.66 B
Mix 2	2.29 A	1.85 A	14.2 9A	11.5 4A	0.61 A	0.66 A	1.94 A	1.86 A

Table 5 : Effect of different growth promoters on pigment content on Thymus Under Soilless Culture Conditions

Harvest	Ch. A (mg/100 FW)		Ch.B (mg/100 FW)		Carotene (mg/100 FW)		Total pigment (mg/100 FW)	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Treatment	Frist season							
Control	0.11F	0.13F	0.07F	0.05E	0.05D	0.04F	0.23G	0.23E
Ascorbic acid (1 g/L)	0.18CD	0.16DE	0.08DE	0.08CD	0.06D	0.07CD	0.32DE	0.31D
Ascorbic Acid (2 g/L)	0.20BC	0.21B	0.10BC	0.11B	0.08BC	0.09B	0.39BC	0.41B
Yeast (2 g/L)	0.14EF	0.13F	0.08EF	0.07D	0.05D	0.05EF	0.27FG	0.25E
Yeast (4 g/L)	0.20BC	0.19BC	0.10CD	0.09C	0.08BC	0.09B	0.37C	0.37BC
Amino acid (0.5 g/L)	0.16DE	0.15EF	0.08EF	0.08CD	0.06D	0.06DE	0.3EF	0.29D
Amino acid (1 g/L)	0.19CD	0.18CD	0.09CDE	0.09C	0.07C	0.08BC	0.35CD	0.35C
Mix 1	0.22B	0.23A	0.12AB	0.12AB	0.09AB	0.11A	0.43B	0.46A
Mix 2	0.28A	0.24A	0.12A	0.13A	0.09A	0.12A	0.49A	0.5A
Second season								
Control	0.13 F	0.12 G	0.08 G	0.05 D	0.05 G	0.06 F	0.25 G	0.23 F
Ascorbic acid (1 g/L)	0.16 DE	0.17 DE	0.09 EF	0.11 D	0.06 EF	0.08 DE	0.31 EF	0.36 E
Ascorbic Acid (2 g/L)	0.19 BC	0.21 BC	0.11 BC	0.56 AB	0.08 BC	0.10 B	0.38 BC	0.86 BC
Yeast (2 g/L)	0.14 EF	0.14 FG	0.08 FG	0.07 D	0.05 G	0.07 EF	0.27 G	0.27 EF
Yeast (4 g/L)	0.18 C	0.19 CD	0.10 CD	0.50 B	0.07 CD	0.09 BC	0.35 CD	0.79 C
Amino acid (0.5 g/L)	0.15 EF	0.15 EF	0.08 EFG	0.08 D	0.05 FG	0.07 EF	0.29 FG	0.30 EF
Amino acid (1 g/L)	0.17 CD	0.19 CD	0.09 DE	0.38 C	0.06 DE	0.08 CD	0.33 DE	0.65 D
Mix 1	0.20 B	0.24 B	0.12 AB	0.58 AB	0.08 B	0.11 A	0.41 B	0.93 AB
Mix 2	0.23 A	0.28 A	0.13 A	0.62 A	0.09 A	0.12 A	0.46 A	1.02 A

Economic Evaluation:

The economic feasibility of this experiment was carried out and the net return and benefit cost ratio (B/C) of the field experiment for thymus fresh leaves are shown in table (6) and figures (2). The results showed that the ascorbic acid (2 g/l) came in the first order in terms of (B/C) where it scored 1.8 B/C, Yeast (4 g/L) came at second where it scored 1.73 in terms of B/C ratio.

Table 6 : Economic evaluation of field experiment (leaves of Thymus crop)Under soilless culture conditions (L.E./m^2).

Treatment	Input			Output			
				Economic criterion			
	Fixed Cost (L.E/m^2)	Variable Cost (L.E/m^2)	Total cost (L.E/m^2)	Revenue (L.E/m^2)	Net Revenue (L.E/m^2)	B/C ratio	Treatment Order
Control	254	178	432	1001.86	569.86	1.32	7
Ascorbic acid (1 g/L)	254	198	452	1158.06	706.06	1.56	4
Ascorbic Acid (2 g/L)	254	218	472	1320.94	848.94	1.80	1
Yeast (2 g/L)	254	192.8	446.8	1057.52	610.72	1.37	6
Yeast (4 g/L)	254	207.6	461.6	1258.06	796.46	1.73	2
Amino acid (0.5 g/L)	254	228	482	1100.27	618.27	1.28	8
Amino acid (1 g/L)	254	278	532	1202.75	670.75	1.26	9
Mix1	254	262.8	516.8	1383.01	866.21	1.68	3
Mix2	254	347.6	601.6	1487.34	885.74	1.47	5

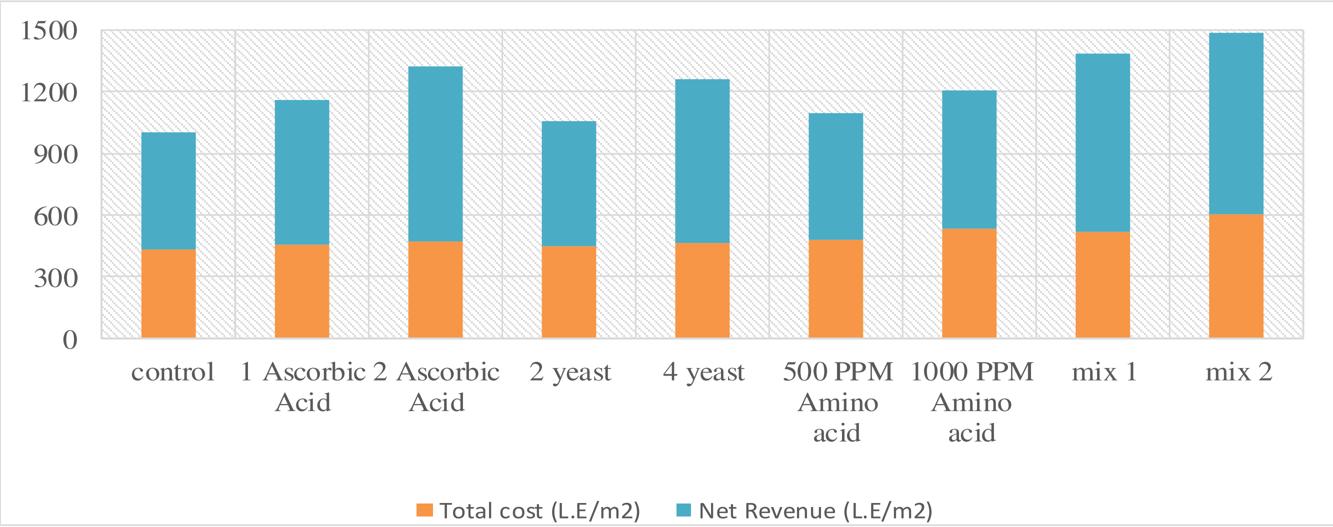

Fig. 2: Relationship between operating costs and net revenue of Thymus plants under soilless culture conditions.

Discussion

In this experiment, results showed the positive influence of yeast, amino acids, vitamins and their mixtures on thymus herbs quality and quantity. Many authors agree with these findings such as Abou EL-Yazied and Mady (2012) on bean plants and Mahmoud *et al.* (2016) on lupine (*Lupinus termis* L.) plants, where they confirmed the significant increase in plants vegetative growth when sprayed with yeast. The superiority of Mix2 treatment may be attributed due to its richness of amino acids which are considered as the building blocks in cell biosynthesis, improve various biochemical processes and facilitate nutrients availability (Dawood and Sadak 2007). Moreover, this mixture is rich in its content of macro and micro nutrients due to existence of high concentrations of yeast, vitamins and amino acids which may play an important role in improving plant vegetative growth and increases plant resistance to diseases (Bevilacqua *et al.*, 2008). The stimulant role of yeast as a growth promoter was attributed to the role it may plays in cell division, and photosynthetic activities such as pigments formation, protein and nucleic acid synthesis. Moreover, yeast has been found to contains protective agents such as amino acids, proteins and vitamins that catalysis plant growth (Wanas, 2006).

The application of Mix treatments were significantly increased plant vegetative parameters with the superiority of Mix2 that led to an average increase of 1.5 times the control treatment of plant fresh weight. Many authors found that foliar application of vitamins significantly increased plant height, No. of branches/plant, fresh, dry weight and essential oil percentage of plants, of them are Fatma *et al.* (2008 and 2014) on lavender and strawberries plants respectively, Nahed *et al.* (2010) on *Thuja orientalis* L., Hendawy *et al.* (2010;2015) on Thymus and Mentha piperitaplants respectively. Azza Ezz El-Din and Hendawy, (2010) confirmed the superiority of the higher concentration of ascorbic acid on *Borago officinalis* plants vegetative growth such as plant height, number of flowers, number of branches and seed weight. While Shafeek *et al.* (2015) and Ahmed and Farm (2015) reported the significant increase vegetative growth of garlic plants and Fawzy *et al.* (2012) on onion plant when sprayed with EM, yeast and amino acids respectively.

All treatments increased thymus essential oil with the superiority of Mix treatments, where Mix2 doubled thymes essential oil percentage compared to the control treatment. These results are similar with many studies were the positive effect of plant growth bio-stimulators on essential oil percentage and oil (ml/herb) were reported (Aly *et al.*, 2007; Hemdan, 2008; Dahab *et al.*, 2010; Kenawy, 2010). All treatments increased the carbohydrate contents of thymus plants in comparison with the control treatment. The application of Mix2 treatment led to an average increase of 1.5 times the control carbohydrates Total carbohydrate contents results are similar to those of El-Sherbeny *et al.* (2012) on *Brassica rapa*, Ali, *et al.* (2014) (2017) on turnip and Garlic plants, Hanafy *et al.* (2017) on Artemisia abrotanum and El-Khateeb *et al*. (2017), Hanafy *et al*. (2018) on *Majorana* plants where they reported the significant increase of total carbohydrates compared to the control treatment when plants were sprayed with some amino acids.

For GC essential oil profile our results confirmed the dominance of thymol among all other constituents which is in agreement with Aly *et al*. (2007), Hemdan (2008), Dahab *et al*. (2010), Kenawy (2010), Maher *et al.* (2011), Olga Kosakowska *et al.* (2020), and Shahad *et al*. (2020). According to the total phenols content; one can find that; mixture treatments increased the total phenols by an average of 2.25 and 1.8 for Mix2 and Mix1 respectively. Many authors agree with these results and confirm the importance of increasing total phenols which may be reflected as increasing in the antioxidant power (Prasanth Reddy *et al*., 2014; Kocira., 2019; Gema Nieto, 2020). The results of increasing pigments content with mixture treatments are in conformity with that recorded by Refaat and Balbaa (2001) on lemongrass, Hassanain *et al*. (2006) on Matricaria chamomilla and Naglaa, (2020) *Helianthus a*nnuus.

Results of this study, directly indicate economic feasibility of the application of plant growth bio-stimulators, which are extremely important to the farmers (Kocira *et al.*, 2020). In contrast to the physical and chemical response of thymus plant to the used bio-stimulators where the mixture treatments were the highest among all other treatments, ascorbic acid treatment (2g/l) and yeast (4 g/l) were the most profitable treatments. this may be attributed to the difference of the variable cost where the mixture treatments variable cost is high.

Conclusion

From the obtained results it can be concluded that, foliar spraying of mixtures of bio-stimulators enhanced the growth, oil percentage and oil yield of thymus. Based on the trial results, it is possible recommended that treating thymus plants with high concentrations mixture of AA+ Vit. C+ yeast significantly increased the yield, productivity, high oil yield. However, from the economic evaluation due to the high cost of this mixture it was not the highest in terms of net return. While the spray with yeast (4 g/l) came at the first order in the B/C ratio due to the low cost of yeast. Thymus cultivation are economically profitable including the control treatment. The bio-stimulants could result in environmentally safe plants to minimize the hazards of pollution caused by using mineral fertilizers.

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