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Integrated management of fertilizer (NPK, chicken manure and yeast) to improve the growth, oil productivity and the volatile oil constituents of *Ocimum* basilicum, L.var Genoves plant

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ABSTRACT

Field Experiments were carried out at the Experimental Farm of Horticulture Dep, Fac. of Agric., Benha University, Egypt, in the two successive seasons of 2014-2015 and 2015-2016. The aim of this study was to investigate the effect of NPK chemical fertilization and combined organic chicken manure with yeast extract on growth, and oil productivity of basil (Ocimum basilicum, L.) cv. Genovese plant. In general, all applied treatments positively increased all growth parameters as plant height, number of branches, fresh and dry weights, photosynthetic pigments (i.e. chlorophyll a, b, and carotenoids), minerals (i.e. N, P, K), and total carbohydrates, especially the combined treatment between chicken manure 20 m³/ fed. with yeast extract 100 M/L in the two cuts and in the two seasons. Also, full dose of NPK ranked the next in this concern, additionally, the combined treatment between chicken manure 20 m³/ fed. with yeast extract 50 M/L produced the third results in two cuts in both seasons. Also, essential oil percentage in leaves gave the maximum values when plants treated with the combined treatment between chicken manure 20 m³/ fed. with yeast extract 100 M/L as compared with untreated ones, while the full dose of NPK ranked the second in this concern. The volatile oil composition of basil produced ten identified components, the main component was the linalool, that ranged from (53.91 to 62.66 %) followed by 1.8 cineol (11.02 to 12.88), β-caryophyllene (7.07 to 11.64%), camphor (3.61 to 5.66%) and geraniol (1.74 to 2.41%). The results proved the possibility to substitute chemical fertilizer with chicken manure 20 m³/ fed. with yeast extract 100 M/ L to decrease the world-wide dependence on hazardous chemical fertilizers which deteriorate the agro-ecosystems.

Key words: Ocimum basilicum, Genovese, chemical fertilization, chicken manure, yeast extract, growth, chemical composition and oil productivity and constituents.

Introduction

Basil (Ocimum basilicum L.) is a widely known member of Lamiaceae family and appreciated as an aromatic spice and medicinal plant (Omer et al., 2008 and Koba et al., 2009). The useful parts of the basil plant are their leaves and seeds. Where each of, the fresh and dry leaves are commonly used in food and spice industries. Furthermore, it is also considered as a source of aroma compounds and thus, possesses a range of biological properties such as insect repellent, nematocidal, antibacterial, antifungal agents and antioxidant properties (Juliani and Simon, 2002 and Lee et al., 2005). Mohamed et al., (2016) on Ocimum basilicum L. cv. Genovese, demonstrated that the volatile oil composition of basil included 10 compounds were identified, the main component was the linalool, followed by 1.8cineol, β-caryophyllene and camphor. Sarrou et al., (2016) on Ocimum basilicum L. found that the main constituents: linalool, 1.8 cineol, 4 terpineol, α-bergamotene and t-cadinene.

Recently, some efforts are paid to minimize the amounts of chemical fertilizers in which applied to medicinal and aromatic plants in order to reduce each of production cost and environmental pollution without reduction of yield (Simaei et al., 2012 and Mohamed, et al. 2015).

Chicken manure improves physical and mechanical characteristics and biological properties of soil; that is, increasing soil organic matter, cation exchange capacity, water holding capacity and availability of mineral nutrients. These results are in harmony with those obtained by other studies on marjoram [El-Desuki et al., (2001) and El-Ghadban et al., (2002)]. Organic manures and biofertilization are very important for medicinal and aromatic plants to produce the best product in both

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quantity and quality and it is safe for human, animal and the environment. Chicken manure is relatively resistant to microbial degradation. However, it is essential for establishing and maintaining optimum soil physical condition and important for plant growth. Chicken manure is also very cheap and effective as a good source of N for sustainable crop production, but its availability remains an important issue due to its bulky nature, while inorganic fertilizer is no longer within the reach of poor-resource farmers due to its high cost (Rahman, 2004). However, John *et al.*, (2004) had advocated for an integral use of organic manure and inorganic fertilizers for the supply of adequate quantities of plant nutrients required to sustain maximum crop productivity and profitability while minimizing environmental impact from nutrient use.

Many investigators reported the stimulating effect of chicken manure on different medicinal and aromatic plants; El-Naggar *et al.*, (2015) on *Ocimum basilicum*, El-Sayed *et al.*, (2015) on *Ocimum sp*, Zaghloul *et al.*, (2016) on *Thymus vulgaris* L plant Ahmed *et al.*, (2017) on *Coriandrum sativum* L Furthermore, Sakr (2017) showed that, the combination of ½ NPK + compost tea+ sheep manure tea was the best treatment examined for improving vegetative, and chemical composition as compared to the control (NPK treatment) in most cases of *Calendula officinalis* plant.

Yeast extract (biofertilizer) act as natural safety and rich source of phytohormones, sugar, vitamins, enzymes, amino acids and minerals. Also, yeast has stimulatory effects on cell division and enlargement synthesis of protein, nucleic acid and chlorophyll formation. The enhancement effect of yeast could be attributed to its stimulating effect on enzymes activity, phytohormones production, improving the uptake of nutrients which increased vegetative growth of plants. It also releases CO₂ which reflected in improving net photosynthesis Hanafy *et al.* (2012). Additionally, it has been demonstrated that yeast cell wall ingredients, such as polysaccharide fraction, can act as elicitors in plant signal transduction pathways as a plant defense response, so it is not only active yeast extract that may be used in agricultural applications. Because of their natural origin, these extracts are environment friendly (eco-friendly) products which can be used in agricultural applications for stimulating the plant defense mechanism and improving the nutraceutical quality of some plants (Złotek and Świeca, 2016).

The aim of this work was to evaluate the benefits of supplementing some kind of fertilizers (i.e., chemical fertilizer and some combination treatments between chicken manure with yeast extract) on growth, seed yield, essential oil productivity and the chemical constituents of the essential oils of *Ocimum basilicum*, *L.* cv. Genovese plant.

Material and Methods

This trail was conducted to study the effects of mineral and organic fertilizer of chicken manure and spraying by yeast extract on vegetative growth, seed production, essential oil yield and chemical constituents of *Ocimum basillicum* L c.v. "Genovese". To achieve the mentioned investigation, field experiment was carried out during two successive seasons 2014/2015 and 2015/2016 in Experimental Farm of Faculty of Agriculture at Moshtohor, Benha university. EL Qalubia Governorate, Egypt.

Plant materials

The *Ocimum basillicum* c.v. Genovese seeds were obtained from the Research Center of Medicinal and Aromatic plants, Ministry of Agriculture, Egypt. Seeds were sown in the nursery on 13th of March in both seasons. The seedlings (15 cm in height with 6-8 leaves and 50 days old) were transplanted in the experimental plots in 20th of May in the two seasons. The area of each plot was 1m² containing 4 plants in two raws, and each row contained 2 plants. The mechanical and chemical analysis of the Experimental Field soil are presented in Table, 1.

Fertilizers

A- Chemical fertilizers

Recommended dose of (NPK) inorganic fertilizer according to the Egyptian Ministry of Agriculture and Land Reclamation (Reclaim) are ammonium sulphate (20.5/N) at 300 kg/fed, potassium sulphate (48.5% K_2O) at 250 kg/fed. The mixture of (NK) was divided into two equal doses added as

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basal dressing. The first does was applied one month after transplanting and the second one was applied after the first cut. while the calcium super phosphate (15% P₂O₂) at 150kg/fed was applied as one dose during the soil preparation.

Table 1: Mechanical properties and chemical analysis of the experimental soil

Parameters	Unit	Values	Parameters	Unit	Values
A. Mechanical proj	perties		B. Chemical analysis		
Coarse sand	(%)	8.01	Organic matter	(%)	1.44
Fine sand	(%)	9.63	CaCO ₃	(%)	0.55
Silt	(%)	30.22	Total nitrogen	(%)	0.28
Clay	(%)	52.14	Total phosphorus	(%)	0.16
Textural class	(%)	Clayey loam	Total potassium	(%)	0.25
			pН		7.66
			EC (dS/m)		0.66

B- The chicken manure:

The chicken manure organic fertilizer was obtained from chicken Farm at Animal experimental station Faculty of Agric., Moshtohor, EL Qualubia Governorate, and added when the soil was prepared at the rates of 0,10,20 m3/fed. The analytical data of chicken manure fertilizer are presented in Table ,2.

Table 2: Analytical data of the chicken manure fertilizer:

Parameters	Unit			
Humidity	% 27.9			
Density	g/cm ³	38.1		
E.C	ds/m	6.1		
pH		7.88		
Organic matter	%	38.55		
Total nitrogen	%	2.77		
Total phosphorus	%	0.71		
Total potassium	%	2.22		

C-The active dry yeast

The active dry yeast extract at 50 and 100 ml/L were sprayed four times, the first was applied after 30 days from transplanting and the second was applied after 15 days from the first one, the third spray took place one month after the first cut and the fourth was applied after on month from the third spray.

Yeast extract preparation:

Using a technique allowed yeast cells (commercial soft yeast) to be grown and multiplied efficiently during conducive aerobic and nutritional conditions. To produce denovo beneficial bioconstituents (i.e., carbohydrates, sugars, proteins, amino acids, fatty acids, hormones andetc), hence allowed such constituents to release out of yeast cells in readily form by using two followed cycles of thawing and freezing.

Such technique for yeast preparation based on:

- Nutritional medium glucose and casein as two favorite sources of C, N and other essential elements (P, K, Ca, Mg, Fe. Mn, Zn, Cu, B, Mo as well as Na and Cl) in suitable balance.
- Air pumping and adjusting incubation temperature.
- Freezing for disruption of yeast cells and releasing their content.

Procedure modified after Abd EI-Rahim et al., (1988). Analysis of prepared yeast stock solution was: total protein (5.3%), total carbohydrates (4.7%), N (1.2%), P (0.13%), K (0.3%), Mg (0.013%), Ca

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(0.02%), Na (0.01%); micro-elements (ppm), Fe (0.13), Mn (0.07), Zn (0.04), Cu (0.04), 3 (0.016), Mo (0.0003), IAA $(0.5 \mu g/ml)$ and GA $(0.3 \mu g/ml)$.

All common agricultural practices (irrigation, fertilization, manual weed control, ... etc.) were carried out when needed.

The treatment was conducted as follows:

- 1- Control without any additions
- 2- Full dose of NPK: (ammonium sulphate (20.5N) at the rate of 300kg/fed, potassium sulphate (48.5%K₂O) at the rate of 250 kg/fed calcium super phosphate (15%P₂O₅) at the rate of 150kg/fed.
- 3- Chicken manure20 m³/ fed.+ yeast extract 100 M/ L
- 4- Chicken manure 20 m³/ fed.+ yeast extract 50 M/L
- 5- Chicken manure10 m³/ fed.+ yeast extract 100 M/ L
- 6- Chicken manure 10 m³/ fed.+ yeast extract 50 M/ L

Harvesting and seed yield.

The plants were harvested twice (second week of July and third week of September) for both seasons by cutting the herb at 10cm above the soil surface

The seeds were collected randomly from 12 plants for each tested treatment at harvesting time (16 weeks from sowing date for both seasons).

The layout of the experiment was a complete randomized block design included 6 treatments with three replicates. Each replicate contained 3 beds (each bed containing 4 plants).

Recorded data:

The vegetative growth parameters:

Plant height, branch number per plant, herbs fresh weight (g/plant), herbs dry weight (g/plant) seed yield (g/plant) and seed yield (kg/fed.)

Chemical composition:

Photosynthetic pigments: chlorophyll a, b and carotenoids were calorimetrically determined in leaves of basil according to the method described by Inskeep and Bloom (1985) and calculated as mg/g fresh weight

The percentage of nitrogen, phosphorus, potassium and total carbohydrates were determined in basil herbs at the flowering stage according the methods described by Horneck and Miller (1998), Sandell (1950), Horneck and Hanson (1998) and Herbert *et al.* (1971), respectively.

Determination of essential oil production.

- Essential oil percentage
- Essential oil yield /fed (kg)

The percentage of volatile oil were determined in the fresh herb using 100 g samples for each cut per plant. Distillation of volatile oil for 3hr in order to extract the essential oils according to the method described by British Pharmacopeia (1963).

Gas liquid chromatography analysis of essential oil (GLC).

The gas liquid chromatography analysis was carried out at the medicinal and Aromatic plant laboratory. Dokki.

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Essential oil samples were performed using Ds chrom 6200 gas chromatograph equipped with aflame ionization detector for separation of volatile oil constituents. The analysis conditions were as follows: The chromatograph apparatus was fitted with capillary column BPX-5.5% phenyl (eguiv.) polysillphenylene – siloxane 30m x 0.25mm ID x 0.25 μ m film. Temperature program ramp increase with rate of 10°C/min from 70° to 200°C. Flow rates of gases were nitrogen at 1ml/min, hydrogenat 30ml/min and 330ml/min for air. Detector and injector temperatures were 300°C and 250°C respectively. The obtained chromatogram and report of GC analysis for each sample were analyzed to calculate the percentage of main components of volatile oil.

Statistical analysis

The design of this experiment was CRD (Complete Randomized Block Design) (Snedecor and Cochran, 1989) as 6 treatments for each treatment replicated three times and each replicate continued 3 beds (each bed containing 4 plants). The differences between the mean values of various treatments were compared by Duncan's multiple range test (Duncan's, 1955).

Results and Discussion

I- Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* cv.genovese plants on vegetative growth during the two seasons (2014-2015 and 2015-2016).

I-1-Plant height (cm) and number of branches / plant

Data presented in Table (3) revealed that all different fertilizer treatments significantly increased the plant height and number of branches / plant of *Ocimum bacillicum* cv. Genovese when compared to control in both seasons. At the two cuts in the first and second seasons, the maximum plant height was observed in plants treated with full dose of NPK when compared to control, followed in descending order by using the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 M/L. Whereas, the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 M/L gave the highest number of branches per plant in the two cuts of both seasons. On the reverse, control (without any addition) gave the lowest values of two parameters at both cuts in the first and second seasons. The rest treatments came in-between the above-mentioned treatments in both seasons.

I-2-Fresh and dry weights of herbs/plant (g)

Data in Table (4) reveal that, fresh and dry weights of herb/plant of *Ocimum bacillicum* cv. Genovese were greatly affected by different investigation treatments in both cuts during two growing seasons. At the two both cuts in the two seasons, the heaviest fresh and dry weights of herbs / plant were observed in plants treated with the combined treatment between chicken manure20 m^3 / fed. with yeast extract 100 M/ L, while full dose of NPK produced the next value in this concern. On the other hand, chicken manure20 m^3 / fed. with yeast extract 50 M/ L produced the third value in this concern in both season. Regardless control the lowest value of these parameters were recorded by using the combination treatments between chicken manure10 m^3 / fed. with yeast extract 50 M/ L at both cuts in the first and second seasons.

In this respect, other studies strongly confirmed results of our study, the results of Chicken manure fertilizer with yeast extract are in parallel with those obtained by El-Naggar *et al.*, (2015) on *Ocimum basilicum*, El-Sayed *et al.*, (2015) on *Ocimum sp*, Zaghloul *et al.*, (2016) on *Thymus vulgaris* L plant, Ahmed *et al.*, (2017) on *Coriandrum sativum* L, Mahmoud *et al.*, (2017) on *Carum carvi* L, Also, Sakr (2017) showed that, the combination of ½ NPK + compost tea+ sheep manure tea was the best treatment examined for improving vegetative) as compared to the control (NPK treatment) in most cases of *Calendula officinalis* plant.

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Table 3: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on plant height (cm) and number of branches / plant during 2014/2015 d 2015/2016 seasons.

		Plant height (cm)				Number of branches/ plant			
Parameters	1 st se	ason	2 nd se	eason	1 st s€	eason	2 nd se	eason	
Treatments	1 st cut	2 nd cut	1 st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut	
Control	55.20 ^f	58.82 ^f	60.56e	61.46e	29.33 ^f	33.67 ^f	33.66e	35.00e	
Full dose of NPK	99.74ª	99.22a	98.35a	99.58a	80.00 ^b	81.67 ^b	82.33 ^b	80.33 ^b	
Chicken manure20 m³/ fed.+ yeast extract 100 M/ L	97.23 ^b	96.47 ^b	97.96ª	99.30ª	87.33ª	88.67ª	88.00ª	89.33ª	
Chicken manure20 m ³ / fed.+ yeast extract 50 M/ L	90.81°	92.51°	87.61 ^b	89.92 ^b	72.33 ^d	74.67 ^d	77.00°	80.00 ^b	
Chicken manure10 m³/ fed.+ yeast extract 100 M/ L	83.91 ^d	88.07 ^d	83.14 ^c	85.50°	76.33°	79.00°	77.00°	76.33°	
Chicken manure10 m ³ / fed.+ yeast extract 50 M/ L	76.84 ^e	80.96e	74.77 ^d	79.98 ^d	65.67e	64.33 ^e	60.33 ^d	64.67 ^d	

Table 4: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on fresh and dry weight of herbs/plant (g) during 2014/2015 d 2015/2016 seasons.

Parameters		Fresh weig	ht of herbs	(g/plant)		Dry weight of herbs (g/pl		
		1st season	2	2 nd season		1st season		eason
Treatments	1st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut
Control	371.20 ^f	389.18 ^f	415.29f	410.42e	65.81 ^f	69.38 ^f	72.81 ^f	71.52 ^f
Full dose of NPK	1011.48 ^b	1021.87 ^b	1065.30 ^b	1071.10 ^a	174.90 ^b	178.80 ^b	185.40 ^b	187.6 ^b
Chicken manure20 m³/ fed.+ yeast extract 100 M/ L	1030.66ª	1050.28ª	1079.95ª	1071.89ª	180.00ª	184.60ª	191.30 ^a	189.31ª
Chicken manure20 m³/ fed.+ yeast extract 50 M/ L	901.08°	905.61°	889.66°	901.16 ^b	146.60°	148.70°	141.11°	143.70°
Chicken manure10 m³/ fed.+ yeast extract 100 M/ L	810.76 ^d	828.74 ^d	870.58 ^d	867.70°	118.40 ^d	123.00 ^d	128.33 ^d	126.0 ^d
Chicken manure10 m³/ fed.+ yeast extract 50 M/ L	671.24°	656.83°	631.76°	642.80 ^d	91.04°	88.43°	82.23 ^e	80.74°

The aforementioned results of vegetative growth measurements as affected by NPK fertilizer are in parallel with those of Nurzyńska-Wierdak *et al.*, (2011) on *Ocimum basilicum*, Nurzyńska-Wierdak and Bartáomiej (2011) on *Ocimum basilicum*, Also, Nurzyńska-Wierdak *et al.*, (2012) on *Ocimum basilicum L.*, Mohamed and Ghatas (2016) on *Viola odorata* L plant., Zaghloul *et al.*, (2016) found that, the treatment of full dose of NPK with or without Saccharomyces extract gave highly significant values of growth characteristics i.e. plant height, branches number/plant, fresh and dry weights were higher of *Thymus vulgaris* L plant in two cuts and in both seasons. Rival *et al.*, (2017) showed that for plant growth of *Anchomanes difformis*, amixed medium of soil/cocopeat (1:1) was the best of the three-media evaluated, and the application of the NPK fertilizer at a dosage equivalent to 100 kg/ha was the optimum fertilizer level.

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I-3-Seeds yield (g) / plant:

Data in Table (5) realized that all different fertilizer treatments with or without yeast extract recorded highly significant increase of yield of seeds (g) / plant of *Ocimum bacillicum* cv. Genovese when compared to control in the two seasons. However, the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 M/ L gave highest values as compared to control in both seasons, followed in descending order by full dose of NPK. Moreover, the combined treatment between chicken manure20 m³/ fed. with yeast extract 50 M/ L gave the third values in this concern. On the other side, the control gave the lowest values of this parameter in the first and second seasons.

Table 5: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on seed yield (g) / plant during 2014/2015 d 2015/2016 seasons.

Parameters	Yield of see	ds (g) / plant
Treatments	1st season	2 nd season
Control	9.293 ^f	10.540 ^e
Full dose of NPK	25.513 ^b	26.550 ^b
Chicken manure20 m ³ / fed.+ yeast extract 100 M/ L	26.927a	29.290a
Chicken manure20 m ³ / fed.+ yeast extract 50 M/ L	22.640°	24.797°
Chicken manure10 m ³ / fed.+ yeast extract 100 M/ L	18.987 ^d	21.953 ^d
Chicken manure10 m ³ / fed.+ yeast extract 50 M/ L	17.367e	21.403 ^d

I-4- Seeds yield / fed.(kg):

Data in Table (6) clear that, yield of seeds /fed.(kg) of *Ocimum bacillicum* cv. Genovese was more affected by using different treatments as compared to control in both seasons, especially the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 M/ L was the most effective one for increasing yield of seeds /fed.(kg), followed by full dose of NPK at the first and second seasons. Additionally, the combined treatments between chicken manure20 m³/ fed. with yeast extract 50 M/ L gave the third values in this concern. On the other reverse, the control gave the lowest values of this parameter in the first and second seasons.

Table 6. Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on seed yield /fed.(kg) during 2014/2015 d 2015/2016 seasons.

Parameters	Yield of seeds	s / fed.(kg)
Treatments	1st season	2 nd season
Control	156.122 ^f	177.072 ^e
Full dose of NPK	428.618 ^b	446.040 ^b
Chicken manure20 m³/ fed.+ yeast extract 100 M/ L	452.374a	492.072a
Chicken manure20 m³/ fed.+ yeast extract 50 M/ L	380.352°	416.590°
Chicken manure10 m ³ / fed.+ yeast extract 100 M/ L	318.980 ^d	368.810 ^d
Chicken manure10 m ³ / fed.+ yeast extract 50 M/ L	291.766 ^e	359.570 ^d

This result is in agreement with those recorded by Bishr *et al.*, (2006) on *Silybum maricmum* L plant. El-Gendy *et al.*, (2012) on roselle demons

trated that all levels of cattle manure and in occulated bio-fertilizer and their interaction enhanced the seeds yield of roselle plants, as well as their quality. Also, Nassar *et al.*, (2015) revealed that foliar application with yeast extract (YE) at (2,4 and 8 g YE/L.) increased significantly yield of seeds of *Ocimum basilicum* L plant.

In this context, Organic fertilization is very important for providing plants with their nutritional requirements without having an undesirable impact on the environment El-Sayed *et al.*, (2002). The increase in *Ocimum* plant height due to application of chicken manure might be attributed to the effect of organic fertilizer in improving physical, chemical and biological properties of soil; that is, increasing soil organic matter, cation exchange capacity, water holding capacity and availability of

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mineral nutrients. These results are in harmony with those obtained by other studies on marjoram (El-Desuki *et al.*, (2001) and El-Ghadban *et al.*, (2002).

In addition, Hanafy *et al.*, (2012) stated that the use of yeast extract as a natural, safety and rich source of phytohormones in order to improve plant growth and productivity has acquired a great attention nowadays. Also, Złotek and Świeca (2016) reported that yeast has stimulatory effects on cell division and enlargement. This enhancement effect of yeast could be attributed to its improving the uptake of nutrients which increased vegetative growth of plants.

Furthermore, mineral fertilizer of *Ocimum* plants, nitrogen is a major component of some important substances which occur in plants, protein, chlorophylls, hormones, amino acids, alkaloids and enzymes are compounds of nitrogen. Plant highly, number of main and secondary branches, number of leaves and herb fresh and dry weights were significantly influenced by application of adequate nitrogen dose. This may be attributed to more proliferation of root biomass, resulting in the higher absorption of nutrients and water from the soil leading to the production of higher vegetative biomass (Hamblin, 1985). Need to constitute amino acids which from protein that participates in cell enlargement and cell division consequently producing more branches bearing more leaves (Cox,1992).it is a well-known fact that nitrogen is a major constituents of several most important substances, which occur in plants, such as nucleoproteins, amino acids and poly peptides in many miscellaneous compound occurring largely in proto plasma and nuclei of the cell plants (Ram *et.al.*1989).Another important effect, (Nguen and Niemeyer, 2008) reported that the first increment of N added to the soil are almost always effective in increasing dry matter yield and secondary product accumulation in herbs, also when nitrogen levels are adequate, the plant growth will be favored with production of photosynthetic pigments and secondary metabolites.

II. Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* cv.genovese plants on chemical composition during the two seasons (2014-2015 and 2015-2016).

II.1. - Chlorophyll "a, b" and carotenoids (mg/g.fw) of leaves:

Data in Table (7) illustrate that, the Chlorophyll "a, b" and carotenoids content in the fresh leaves of *Ocimum bacillicum* L cv. Genovese were affected by all tested treatments when compared to control at the both cuts in the two seasons, especially full dose of NPK showed to be the most effective treatment for inducing the highest values in these parameters, followed in descending order by using the combined treatment between chicken manure20 m3/ fed. with yeast extract 100 M/L. Also, the combined treatment between chicken manure20 m3/ fed. with yeast extract 50 M/L gave the third values in this concern. On the other hand, control (without any addition) gave the lowest values of these parameters at both cuts in the first and second seasons.

II -2- Nitrogen and Phosphorus percentage

Data presented in Table (8) show that all the fertilizer treatments succeeded in increasing the values of N% and P% in leaves of *Ocimum bacillicum* cv. Genovese plants. However, the best results of these parameters were obtained in the treatment of full dose of NPK as compared to control in both cuts in the two seasons. Moreover, either treatments of the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 m/ l or yeast extract 50 m/ l resulted in highly increments of this parameter as compared to control in the two cuts in the two seasons. On the contrary, the lowest values of N% and P% were recorded by control in both seasons in the first and second cuts.

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Table 7: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on Chlorophyll "a, b" and carotenoids (mg/g.fw) of leaves

during 2014/2015 d 2015/2016 seasons.

Parameters	Chloroph	nyll a (mg/	/g.fw)		Chlorop	hyll b (mg	g/g.fw)		Caroteno	oids(mg/g.:	fw)	
Farameters	1st se	ason	2 nd se	eason	1st se	ason	2 nd se	eason	1st se	ason	2 nd se	eason
Treatments	1st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut	1 st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut
Control	0.770e	0.803e	0.733 ^d	0.801 ^d	0.607d	0.593 ^d	0.653 ^d	0.637e	0.340e	0.327e	0.427bc	0.443 ^b
Full dose of NPK	1.257ª	1.340ª	1.313 ^a	1.340 ^a	0.897ª	0.873ª	0.913ª	0.897ª	0.557ª	0.607ª	0.547ª	0.550a
Chicken manure20 m3/ fed.+ yeast extract 100 M/ L	1.207 ^{ab}	1.267 ^b	1.307ª	1.293ª	0.873ª	0.897ª	0.857 ^b	0.847 ^b	0.507 ^b	0.543 ^b	0.543ª	0.507ª
Chicken manure20 m3/ fed.+ yeast extract 50 M/ L	1.173 ^b	1.160°	1.147 ^b	1.157 ^b	0.797 ^b	0.777 ^{bc}	0.803°	0.797°	0.467 ^{bc}	0.487°	0.397°	0.383°
Chicken manure10 m³/ fed.+ yeast extract 100 M/ L	0.950°	0.993 ^d	1.110 ^b	1.070°	0.727°	0.743°	0.697 ^d	0.717 ^d	0.407 ^d	0.433 ^d	0.433 ^{bc}	0.413 ^{bc}
Chicken manure10 m³/ fed.+ yeast extract 50 M/ L	0.890 ^d	0.957 ^d	0.863°	0.810 ^d	0.783 ^b	0.807 ^b	0.811 ^{bc}	0.827 ^{bc}	0.450°	0.467 ^{cd}	0.450 ^b	0.427 ^{bc}

Table 8: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on nitrogen and phosphorus percentage of leaves during 2014/2015 d 2015/2016 seasons.

Parameters		Nitrogen percentage				Phosphorus percentage			
	1st se	ason	2 nd so	eason	1st se	ason	2 nd se	eason	
Treatments	1st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut	
Control	1.55e	1.61e	1.69e	1.71 ^f	0.229°	0.250°	0.228°	0.225 ^d	
Full dose of NPK	3.21ª	3.20a	3.23a	3.20a	0.335a	0.339a	0.340a	0.347a	
Chicken manure20 m ³ / fed.+ yeast extract 100 M/ L	2.98 ^b	3.10 ^b	3.11 ^b	3.14 ^b	0.311 ^{ab}	0.330 ^{ab}	0.228ab	0.310 ^b	
Chicken manure20 m ³ / fed.+ yeast extract 50 M/ L	2.64°	2.71°	2.76°	2.81°	0.291 ^{ab}	0.295 ^{abc}	0.296 ^{ab}	0.301 ^b	
Chicken manure 10 m ³ / fed.+ yeast extract 100 M/ L	2.42 ^d	2.50 ^d	2.49 ^d	2.59 ^d	0.281 ^b	0.285bc	0.289 ^{ab}	0.280°	
Chicken manure10 m ³ / fed.+ yeast extract 50 M/ L	2.61°	2.53 ^d	2.42 ^d	2.51e	0.277 ^b	0.296°	0.268bc	0.269°	

II <u>-</u>3- Potassium and total carbohydrates percentage

Data presented in Table (9) illustrate that, all the fertilizers treatments statistically affected leaf potassium and total carbohydrates percentage in both seasons in the two cuts of *Ocimum bacillicum* cv. Genovese plants. However, in both season using the application of full dose of NPK showed to be most affected for increasing potassium and total carbohydrates percentage at two cuts, followed in descending order by using the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 m/l. Additionally, the untreated plants had a lowest percentage of these parameters at both cuts in the two seasons.

In this concern, results are coincided with those obtained by Nurzyńska-Wierdak *et al.*, (2011) on *Ocimum basilicum*, Mohamed and Ghatas (2016) on *Viola odorata* L plant. Zaghloul *et al.*, (2016) demonstrated that, the treatment of full dose of NPK with or without Saccharomyces extract gave the highest significant values of N, P, K percentages and total carbohydrates % in leaves of *Thymus vulgaris* L plant as compared to control in two cuts and in both seasons.

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Table 9: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on potassium and total carbohydrates percentage of leaves during 2014/2015 d 2015/2016 seasons.

Parameters		Potassium percentage			Total carbohydrates percentage			
	1st se	ason	2 nd se	eason	1st se	ason	2 nd se	eason
Treatments	1st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut	1st cut	2 nd cut
Control	1.14 ^d	1.17 ^f	1.21 ^f	1.17 ^f	8.40 ^d	9.27 ^d	9.01 ^e	8.79e
Full dose of NPK	2.96a	2.91a	2.89a	2.93a	13.27a	13.25a	14.04 ^a	14.10 ^a
Chicken manure 20 m ³ /								
fed.+ yeast extract 100	2.88^{a}	2.85 ^b	2.82^{b}	2.90^{b}	13.10 ^a	13.20a	14.07a	13.47 ^b
M/L								
Chicken manure 20 m ³ /								
fed.+ yeast extract 50	2.55^{b}	2.64 ^c	2.65 ^c	2.70^{c}	12.79 ^b	12.88 ^b	13.19 ^b	13.29 ^b
M/L								
Chicken manure 10 m ³ /								
fed.+ yeast extract 100	2.40^{c}	2.51 ^d	2.51^{d}	2.60^{d}	10.89 ^c	11.56 ^c	12.60 ^c	12.14 ^c
M/L								
Chicken manure 10 m ³ /								
fed.+ yeast extract 50	2.52^{b}	2.31e	2.30^{e}	2.29 ^e	10.71 ^c	9.37^{d}	11.09 ^d	10.99 ^d
M/L								

III. Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* cv. genovese plants on essential oil yield and composition during the two seasons (2014-2015 and 2015-2016).

III.-1- Essential oil percentage

Data presented in Table (10) indicate that, essential oil percentage of *Ocimum bacillicum* cv. Genovese was more affected by using all different fertilizers treatments as compared to control in both cuts and both growing seasons. However, the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 m/ l showed the highest significant increase of essential oil yield percentage/plant, followed by full dose of NPK at the first and second cuts in both seasons. Furthermore, the combined treatment between chicken manure 20 m³/ fed. with yeast extract 50 m/ l gave the third value at both cuts, in the first and second seasons. The lowest value of oil percentage per plant was produced by unfertilized plants(control) at both cuts and two seasons.

III. -2- Essential oil yield /fed (kg)

Data presented in Table (11) reveal that, plant of *Ocimum bacillicum* cv. Genovese which received the combined treatment between chicken manure20 m³/ fed. with yeast extract 100 m/ l showed the highest significant increase of essential oil yield / fed (kg), followed by full dose of NPK at the first and second cuts in both seasons On the other side, the other hand, chicken manure20 m³/ fed. with yeast extract 50 m/ l gave the third value at both cuts, in the first and second seasons. Furthermore, the lowest value of oil percentage / fed (kg) was produced by control plants at both cuts and two seasons.

The aforementioned results with those obtained by Sharaf EL-Din *et al.*, (2013) on *Rosmarinus officinalis* L., El-Naggar *et al.*, (2015) on *Ocimum basilicum L.*, El-Sayed *et al.*, (2015) on *Ocimum sp.* Bajeli *et al.*, (2016) on *Mentha arvensis* L. highest yield attributes and oil yield were recorded in combined application of FYM, vermicompost and chicken manure as compared with other treatments. Zaghloul *et al.*, (2016) demonstrated that, using half dose of zeolite combined with PGPR and Saccharomyces extract gave the highest significant values of essential oil yield of *Thymus vulgaris* L plant as compared to control in two cuts and in the two growing seasons.

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Table 10: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extra,.ct of *Ocimum basillicum* L cv. genovese plants on essential oil percentage during 2014/2015 d 2015/2016 seasons.

Daramatara	Essential oil %				
Treatments	Parameters 1 st season			2 nd season	
Treatments	1st cut	2 nd cut	1 st cut	2 nd cut	
Control	0.760 ^f	0.803 ^d	0.813 ^f	0.837e	
Full dose of NPK	1.310 ^b	1.353a	1.250 ^b	1.313a	
Chicken manure20 m ³ / fed.+ yeast extract 100 M/ L	1.387a	1.387a	1.427a	1.340a	
Chicken manure20 m ³ / fed.+ yeast extract 50 M/ L	1.207 ^c	1.167 ^b	1.163°	1.193 ^b	
Chicken manure10 m ³ / fed.+ yeast extract 100 M/ L	1.097 ^d	1.053°	0.953 ^e	0.917 ^d	
Chicken manure 10 m ³ / fed.+ yeast extract 50 M/ L	0.953e	1.060°	1.037 ^d	0.967°	

Table 11: Effect of mineral fertilizer and the combination treatments between chicken manure with yeast extract of *Ocimum basillicum* L cv. genovese plants on essential oil yield / fed (kg), during 2014/2015 d 2015/2016 seasons.

Parameters	Essential oil / fed.(kg)				
	1 st se	ason	2 nd season		
Treatments	1 st cut	2 nd cut	1 st cut	2 nd cut	
Control	12.768 ^f	13.490 ^d	13.658 ^f	14.062e	
Full dose of NPK	22.008 ^b	22.730a	21.00 ^b	22.058a	
Chicken manure20 m ³ / fed.+ yeast extract 100 M/ L	23.302a	23.302a	23.974a	22.512a	
Chicken manure20 m ³ / fed.+ yeast extract 50 M/ L	20.278°	19.606 ^b	19.538c	20.042 ^b	
Chicken manure10 m ³ / fed.+ yeast extract 100 M/ L	18.430 ^d	17.690°	16.010e	15.406 ^d	
Chicken manure10 m ³ / fed.+ yeast extract 50 M/ L	16.010 ^e	17.808°	17.422 ^d	16.246 ^c	

III. -2- Essential oil constituents:

Table (12) and Figs. (1,2,3 and 4) show the effect of the assigned treatments of full dose of NPK, the combined treatment between chicken manure 20 m³/ fed. with yeast extract 100 M/ L, the combined treatment between chicken manure 20 m³/ fed. with yeast extract 50 M/ L and the control on the constituents of essential oil distilled from *Ocimum bacilicum* cv. Genovese. The volatile oil constituents of basil included 10 compounds were identified, i.e. linalool, 1.8cineol, β-caryophyllene, camphor, Geraniol, myrcene, α-Pinene, , B- pinene, , methyl charicole, and Eugenol. The main component was the linalool, that ranged from (53.91 to 62.66 %) followed by 1.8 cineol (11.02 to 12.88), β-caryophyllene (7.07 to 11.64%), camphor (3.61 to 5.66%) and geraniol (1.74 to 2.41%). In addition, unknown component with vales from (5.63 to 8.03%). However, the combined treatment between chicken manure 20 m³/ fed. with yeast extract 100 M/ L gave the maximum values of linalool (62.66 %) followed by full dose of NPK (62.04%)when compared to the values of (53.91%) as control. On the other hand, different treatments caused decreases in the percentage of β-caryophyllene from 11.64 in control to 11.02, 8.92 and 7.07%.

Similar results had been obtained by Mohamed *et al.*, (2016) on *Ocimum basilicum L.* cv. Genovese, demonstrated that The volatile oil composition of basil included 10 compounds were identified, i.e. linalool, 1.8cineol, β -caryophyllene, camphor, α -Pinene, B- pinene, , α -Terpineneol, general acetate, methyl charicole and Sabinene. The main component was the linalool, that ranged from (57.41 to 62.72 %) followed by 1.8cineol (8.41 to 11.36), β -caryophyllene (8.19 to 9.23%) and camphor (4.44 to 5.45%). Also, Sarrou *et al.*, (2016) on *Ocimum basilicum L.* found that the essential oil content ranged between 1.06-1.27% and 0.88-1.36% under mulching and fertilizing treatments, respectively, with main constituents: linalool, 1.8 cineol, 4 terpineol, α -bergamotene and t-cadinene. Moreover, significant quantitative differences in basil volatile constituents were observed under the influence of

different types of fertilizers. The components 1, 8 cineol, α -bergamotene and t-cadinol were mostly affected by organic fertilization (O-L).

Table 12: Effect of different treatments on essential oil constituents.

				Area %	
Peak	Component		Full dose	Chicken manure20	Chicken manure20 m ³ /
No.	name	Control	of NPK	m ³ / fed.+ yeast extract	fed.+ yeast extract 50 M/
			OI INI K	100 M/ L	L
1	α-Pinene	0.78	0.56	0.67	0.68
2	Myrcene	1.18	1.48	1.74	1.75
3	B- pinene	0.89	0.59	0.49	0.88
4	1 ,8-Cineole	12.38	11.02	12.88	12.78
5	Linalool	53.91	62.04	62.66	56.44
6	camphor	3.61	5.48	5.66	4.01
7	Methyl charicole	4.44	1.99	1.13	2.29
8	Geraniol	2.41	1.74	1.74	1.96
9	Eugenol	0.73	0.38	0.33	0.55
10	β-caryophyllene	11.64	8.92	7.07	11.02
*	Unknown	8.03	5.80	5.63	7.64
Total		100.00	100.00	100.00	100.00

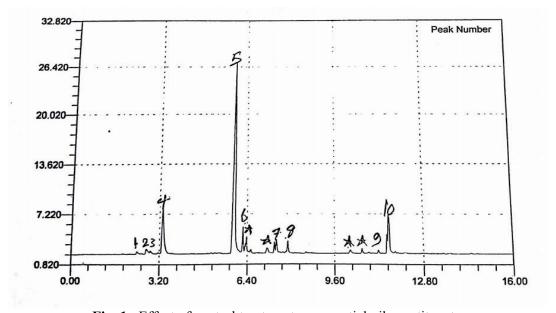


Fig. 1: Effect of control treatment on essential oil constituents.

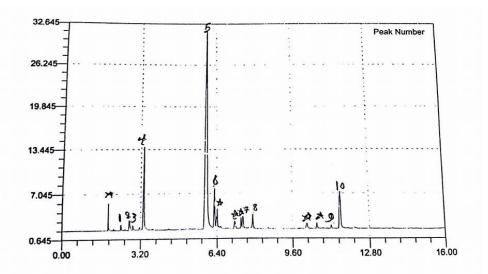


Fig. 2: Effect of full dose of NPK treatment on essential oil constituents.

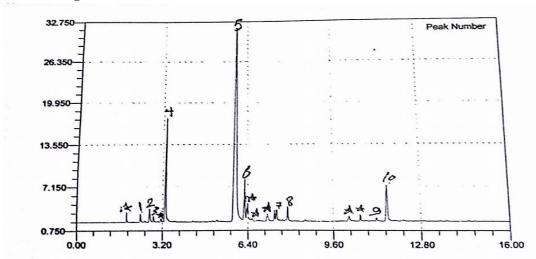


Fig 3: Effect of chicken manure 20 m³/ fed.+ yeast extract 100 M/ treatment on essential oil constituents

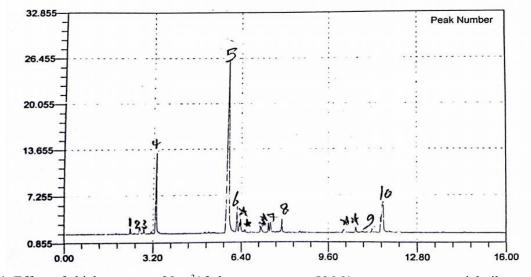


Fig 4: Effect of chicken manure 20 m³/ fed.+ yeast extract 50 M/ treatment on essential oil constituents.

Conclusion

Conclusively, it is preferable from the previous results that treating *Ocimum bacillicum* cv. Genovese plants with the combined treatment between chicken manure $20~\text{m}^3/\text{ fed.}$ with yeast extract 100~M/L or full dose of NPK the most effective treatments to improve growth, oil productivity and volatile oil constituents.

The results proved the possibility to substitute chemical fertilizer with chicken manure $20 \text{ m}^3/\text{ fed.}$ with yeast extract 100 M/L to decrease the world-wide dependence on hazardous chemical fertilizers which deteriorate the agro-ecosystems.

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