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Effect of Foliar Application of Some Aromatic Waters on Productivity and Chemical Composition of Garlic under Sandy Soil Conditions

¹El-Saied R.M., ²Hala F. Mohammed and ²Reham S. Abd Elhamed

¹Soils, Water and Environment Research Institute, Agriculture Research Center, Giza, Egypt.

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ABSTRACT

Two field experiments were conducted during two successive seasons of 2017/2018 and 2018/2019 at the Horticulture Research Station Farm in El-Quassassin, Ismailia Governorate, Egypt, to assay the effect of various sources of aromatic waters of peppermint, geranium, spearmint, rosemary and sour orange (petitgrain) on growth parameters, yield, chemical constituents and volatile oil production of garlic plants (*Allium sativum* L.). Results showed that the foliar application with aromatic waters gave significant response in various aspects compared with plants treated with distilled water (control) during two seasons. Garlic plants that sprayed with petitgrain aromatic water significantly enhanced plant growth and yield. Moreover, the highest values of chemical contents and antioxidant activity were recorded for the same treatment. On the contrary, this treatment recorded the lowest nitrate content for both seasons. Results showed non-significantly affect among sulphur content values. It is remarkable that spraying with peppermint recorded significant volatile oil production compared with untreated plants during two seasons.

Keywords: Garlic, aromatic waters, peppermint, geranium, spearmint, rosemary, sour orange (Petitgrain).

Introduction

Garlic (Allium sativum L.) belongs to the Alliaceae family. Garlic is considered one of Egypt's oldest and most important vegetable crops, owing to its widespread consumption, exports, and medicinal benefits. It is the world's second most common Allium species, behind onion. Egypt is the fourth-largest producer of garlic in the world after China, India and Korea. The total cultivated garlic area in Egypt was 38483 Fedden produce about 280216 tons (Ministry of Agriculture and Land Reclamation, 2018). It is considered as one of the most important cultivated crops that used as a fresh vegetable or dried in the form of spices. It has anti-diabetic, antibacterial, antiviral, antiprotozoal and antibiotic properties due to the presence of selenium and sulphur active chemicals in the form of cysteine derivatives. In addition to a variety of other biological functions, this herb is a valuable source of therapeutic agents (Baliga et al., 2013). In garlic, the bioactive compounds derived from sulfur-containing compounds include allicin, diallyl sulfide, diallyl trisulphide, ajoene, diallyl disulfide and 2-vinyldithiins, which are the main antioxidant compounds that responsible for biological activity for garlic (Shang et al., 2019). Also, garlic is rich with other bioactive compounds such as bioactive peptides, dietary fibres, polyphenols, and micronutrients (especially Se) (Capasso, 2013). Besides containing bioactive organosulfur compounds, garlic also contains flavonoids and polyphenols, which are potential antioxidant agents (Shang et al., 2019).

Aromatic waters (hydrosol) are by-products produced by steam distillation of aromatic plants for extraction essential oil. Aromatic waters include intense herbal aroma and certain amounts of water-soluble components of the essential oils, phenols, flavonoids and other polar plant material components (D'Amato *et al.*, 2018). Aromatic waters offer benefits for agriculture as input or for human health as proposed by (Bosson, 2011). They exhibit antibacterial and antifungal agents (Boyraz and Özcan, 2005 & Boyraz and Özcan, 2006) also, they used in food industry as flavor and food preservation, in cosmetic field as in message and skin cleaning (Catty, 2000). Furthermore

Corresponding Author: Reham M. El-Saied., Soil Fertility and Plant Nutrition Department, Soils, Water and Environment Research Institute, Agriculture Research Center, Giza 12619, Egypt.

² Medicinal and Aromatic Plants Research Department, Horticulture Research Institute, Agriculture Research Center, Giza, Egypt.

aromatic waters have biological role in agriculture against mildew, insects and for fertilization of soils (Paolini *et al.*, 2008 and Rajeswara Rao, 2012). For repel insect pests and diseases causing organisms, aromatic waters are collected and sprayed on agricultural crops to (Rajeswara Rao, 2012).

In addition their biological role, they have very strong antioxidant activity (Belabbes *et al.*, 2017). Aromatic waters of different parts of medicinal and aromatic plants contain different kinds of components which include hydrocarbons (mainly terpenes and its derivatives), alcohols, esters, aldehydes, phenols, acids (Guenther, 1961 & Francezon and Stevanovic, 2017).

The chemical composition of aromatic waters was analyzed by many researchers and they found that, the major components of aromatic waters are generally the same present in oxygenated fraction of the parents and corresponding essential oils (Price and Price, 2004 & Edris, 2008). 1,8-cineole, camphor, terpinen-4-ol, verbenone, bornyl acetate, α -terpineol, borneol and 3-octanone were dominant in both of aromatic water and essential oil of rosemary (Hay *et al.*, 2015 & Hay *et al.*, 2018). Edris (2008) analyzed the essential oil from some aromatic waters of plants which were donated from Horticultural Research Institute, Distillation laboratory of Medicinal and Aromatic Plant, El-Kanater El-Khayria, Egypt. The analysis appears linalool, α -terpinol and linayl acetate in sour orange (petitgrain), while 1, 8-cineole and carvone in spearmint. Also, peppermint aromatic water contains 1, 8-cineol, menthon and menthol. The main constituents of geranium essential oil are citronellol, geraniol, caryophyllene, menthone, linalool, β -bourbonene, iso-menthone and geranyl formate (Sharopov *et al.*, 2014).

The main ingredients of sour orange (petitgrain) oil were linalool, (was the main alcohol components), linally acetate, (which considered the major component of ester fraction), (limonene, β -ocimene, myrcene and β -pinene were the highest monoterpene hydrocarbons), α -terpineol, geranyl acetate, geraniol and geranial (Abd El-Rashid, 2005 & Gniewosz *et al.*, 2017).

Our objective for this study to determine the effect of different aromatic waters (hydrosols) on growth, productivity, chemical constituents and essential oil of garlic plants under sandy soil conditions.

Materials and Methods

The experimental site:

The Field experiment was conducted during the two winter seasons of 2017/2018 and 2018/2019 at the Experimental Farm of EL-Quassassin, Horticulture Research Station, Ismailia Governorate, Egypt (30° 34' N Latitude, 31° 56' E Longitude) to study the effect of foliar application of five aromatic waters as natural products on growth, yield and chemical composition of garlic plant. The aromatic waters of different fresh plant parts as herb of peppermint (*Mentha piperita*), herb of geranium (*Pelargonium graveolens*), herb of spearmint (*Mentha spicata* L.), leaves of rosemary (*Rosmarinus officinalis*) and petitgrain of sour orange (*Citrus aurantium* L.) were obtained from Horticultural Research Institute, Distillation laboratory of Medicinal and Aromatic Plant, El-Kanater El-Khayria, Egypt.

Garlic (Balady) grown in sandy soil conditions under drip irrigation system. The physical and chemical analyses of the soil are presented in Table (1).

The experimental design was set up in complete randomized blocks with six treatments; each treatment was replicated three times.

The experimental field was ploughed and pulverized, $20~\text{m}^3$ farmyard manure/fed., ammonium nitrate (33.5 % N) at the rate of 300 kg/fed., super phosphate (15.5 % P_2O_5) at the rate of 300 kg/fed., potassium sulphate (48 % K_2O) at the rate of 100 kg/fed. and sulphur at the rate of 100 kg/fed., were added. Other agricultural practices for garlic production were carried out as recommended by the Egyptian Ministry of Agriculture. The analysis of FYM fertilizers (average of two seasons) was 1.08 % total N, 0.36% total P, 0.64% total K. In addition, its organic matter = 23.84%, pH (1:10) = 6.35 and EC = 4.13 dSm⁻¹.

The garlic bulbs (Balady) were obtained from Horticultural Research Institute, Agriculture research center, Egypt. The largest size, uniform and healthy cloves were chosen then soaked them in water overnight. Cloves were sown on October 22nd and 26th in 1st and 2nd seasons, respectively, on both side of row at 10 cm distance between plants and 60 cm between rows.

Spraying treatments with different aromatic waters were started one month after planting and sprayed three times by 15 days.

Table 1: The physical and chemical analyses of the experimental soil.

Properties	Valu	ues
	2017-2018	2018-2019
Physical properties		
Sand (%)	77.00	78.25
Silt (%)	17.50	16.60
Clay (%)	5.50	5.15
Soil texture	Sandy loam	Sandy loam
Chemical parameters		
N (mg kg ⁻¹)	49.56	51.34
$P (mg kg^{-1})$	2.31	2.17
K (mg kg ⁻¹)	41.50	46.40
Organic matter(%)	0.47	0.39
E.C dSm ⁻¹ (1:5)	2.07	2.16
pH soil suspension(1:2.5)	7.84	7.90

The experiment comprised of six treatments

- 1- Control (spraying with distilled water).
- 2- Foliar application with aromatic water of peppermint.
- 3- Foliar application with aromatic water of geranium.
- 4- Foliar application with aromatic water of spearmint.
- 5- Foliar application with aromatic water of rosemary.
- 6- Foliar application with aromatic water of sour orange (petitgrain).

Total phenols, total flavonoids content and antioxidant activity were determined in different aromatic waters according to Amin *et al.* (2006), Zhuang *et al.* (1992) and Chen *et al.* (2007) respectively in Central lab of Horticulture Research Institute, Agriculture Research Center. The data displayed in Table (2).

Table 2: The average values of phenols, flavonoids, and antioxidant activity in aromatic waters for both seasons

Aromatic waters	Total phenols (mg/l)	Total flavonoids(mg/l)	Antioxidant activity (%)
Peppermint	256.18	229.59	27.17
Geranium	114.75	97.35	13.18
Spearmint	235.61	203.50	23.25
Rosemary	271.75	189.87	16.71
Sour orang (Petitgrain)	269.5	237.50	35.31

Data recorded

Plant growth parameters

After 135 days from planting in both seasons, a random sample of garlic plants was picked from each plot to measure plant height (cm), fresh weight of leaves (g)/plant and number of leaves/plant.

Yield parameters

Garlic plants were harvested when 70% of the plant leaves became yellow, after about 190 days from planting, then moved to a shaded location in the same day for curing (10 days) and the studied traits: bulb fresh weight (g)/plant, number of cloves/bulb/plant, neck diameter (cm)/plant, bulb diameter (cm)/plant.

Bulb in ratio which calculated as follow: $\frac{\text{Neck diameter(cm)/plant}}{\text{Bulb diameter (cm)/plant}} X 100$ [as described by (Mann, 1952)].

Clove weight (g)/bulb/plant which calculated in the formula: $\frac{\text{Bulb fresh weight(g)/plant}}{\text{Number of cloves/bulb/plant}} \times 100$

Also, fresh total yield of plants (ton fed⁻¹) and bulbs' yield (ton fed⁻¹) calculated after separating leaves from bulb for each plant.

Chemical composition

Fresh samples of 100 g of bulbs were oven dried at 65 °C for 48 h. The dry matter was finely ground and wet digested with sulphuric-perchloric acid mixture (1:1). Total nitrogen content by using the modified Micro-kjeldahl apparatus as described by Jones *et al.* (1991). Total phosphorus was determined spectrophotomitrically by Peters *et al.* (2003). Total potassium was estimated flame photometerically by Peters *et al.* (2003). While total protein was calculated by multiplying total nitrogen x 6.25 (AOAC, 1994). Total carbohydrate was estimated according to anthrone method (Sadasivam and Manickam, 1996). Nitrate content in the dried garlic bulb were determined according to the methods described by Cafado (1975). Sulphur percentage was estimated by turbidometric method (Tabatabai and Bremner, 1970).

- Antioxidant activity of garlic plant By DPPH assay: was evaluated in garlic plant cloves by 1, 1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging method according to the procedure of Chen *et al.*, (2007).
- Volatile oil from fresh bulb was isolated by steam distillation for 2 h. by a Clevenger Apparatus using the method of Guenther (1961). Volatile oil percentage (v/w %) was estimated according to British Pharmacopoeia (1963).

Statistical analysis

The treatments of each season were arranged in a complete randomized block design with three replicates. Data were analyzed with statistical analysis software; CoStat (2005). All multiple comparisons were first subjected to analysis of variance (ANOVA). Comparisons among means were made using Duncan's multiple range test at a P level of 0.05.

Results

Vegetative growth parameters

Data in Table (3) revealed that, the foliar spray with peppermint, geranium, spearmint, rosemary and petitgrain aromatic waters caused a significant increase in plant vegetative growth parameters. Results showed that foliar spraying the aromatic waters showed different responses in growth garlic plants comparing with control plants.

Table 3: Vegetative growth parameters of garlic plants as affected by foliar spray of different aromatic water during seasons of 2017/2018 and 2018/2019.

Characte		height m)		of leaves	Fresh weight (g) of leaves/plant		
	1 st	2 nd	1 st	1 st 2 nd		2 nd	
Treatment	season	season season s		season	season	season	
Control (distilled water)	61.75e	63.00e	8.00c	9.00c	62.00e	64.13e	
Aromatic water of peppermint	69.76c	71.20c	9.15bc	9.50bc	72.50b	73.67b	
Aromatic water of geranium	65.35d	64.88d	11.00 a	11.00a	61.45e	63.50e	
Aromatic water of spearmint	73.60b	75.52b	10.50ab	10.00abc	67.00d	69.37d	
Aromatic water of rosemary	67.00d	69.75c	9.50abc	9.50bc	70.50c	71.58c	
Aromatic water of sour orange (petitgrain	76.37a	77.75a	10.00ab	10.75ab	75.42a	76.48a	

Different letters within each column indicate a significant difference using Duncan's Multiple Range Test at P-value of ≤ 0.05 .

Data also showed that the foliar application of petitgrain aromatic water gave increasing in plant height (cm) comparing with control by (23.68 and 23.41%) in 1st and 2nd seasons, respectively. Also, it significantly increased fresh weight of leaves (g) by (21.65 and 19.26%). Foliar with geranium gave highest number of leaves by (37.50, 22.22 %) in the two seasons comparing with untreated plants, while this application had no stimulation comparing with control for fresh weight

Bulb characteristics and garlic yield

Regarding to (Table 4) represented application of different types of aromatic waters on cured bulb characters and garlic yield during the two seasons. Results showed that foliar spraying the aromatic waters increased the yield responses of garlic plants through both seasons. It worth to be mentioned that, highest yield parameters were recorded with foliar application by petitgrain aromatic water. The increments over the control were (23, 17%) for neck diameter, (22.35, 19.56%) for bulb diameter, (28.37, 22.29%) for bulb weight, (34.44, 37.81%) for cloves weight through 1st and 2nd seasons, respectively. While spraying with peppermint and rosemary increased number of cloves comparing with others. While, spraying with rosemary has no effect in bulb diameter comparing with control plants through in the 1st season. Yield of plant (ton fed -1) increased by (25.45, 20.76%) and yield of bulb (ton fed -1) by (28.46, 22.35%) in case spraying with petitgrain aromatic water during the two seasons comparing to control.

Chemical constituents of bulb

Data showed in Table (5) clear that, all foliar spraying with different aromatic waters affected on chemical components of garlic positively except nitrate decreased with spraying. There are no significant differences between values of nitrogen and protein content among spraying with spearmint and petitgrain aromatic waters through 1st seasons. Spraying with petitgrain aromatic water increased protein content by (16.51and 21.85%) for 1st and 2nd seasons, respectively, compared with control. Concerning with phosphorous content there are no significant difference when spraying with peppermint and petitgrain aromatic waters through both seasons. Where, spraying with peppermint aromatic water gave increasing over control by (35.60, 38.40%) through both seasons, respectively. Also, potassium content has same behavior through 2nd season. While spray with petitgrain aromatic water recorded increasing by (16.57, 18.21%) for carbohydrate through 1st and 2nd seasons, respectively. On other hand, nitrate content decreased with spraying with petitgrain aromatic water by (2.71, 3.81%) and peppermint aromatic waters by (2.59, 2.69%) through 1st and 2nd seasons, respectively, comparing with control. It is noticeable that, there were non-significant differences among values of sulphur content.

Volatile oil and antioxidant activity

This study showed that volatile oil and antioxidant activity of garlic plants were significantly improved with foliar application of aromatic waters during both seasons. Data recorded in Table (6) revealed that, there were no significant effects on volatile oil percentage between plants sprayed with peppermint and spearmint aromatic waters through 1st season. Where, spraying plants with peppermint aromatic waters gave maximum values through both seasons comparing with control by (54.26, 42.34%) and (88.63, 64.91%) for volatile oil percentage and volatile oil yield per plant, respectively. While spraying with petitgrain aromatic water, increased over control by (50.73, 37.94%) for antioxidant activity through 1st and 2nd seasons. There were non-significant differences for antioxidant activities among spraying with geranium, spearmint aromatic waters and distilled water (control) during the two seasons

Table 4: Garlic yield parameters as affected by foliar spray of different aromatic water during seasons of 2017/2018 and 2018/2019.

Ch	aracter	Neck diameter (cm)/plant			diameter a)/plant	0	t of bulb plant	No. of cloves /plant		
Treatment	_	1st season	2 nd season	1 st season	2 nd season	1st season	2 nd season	1 st season	2 nd season	
Control (distilled water)		1.13d	1.23d	4.25d	4.50d	47.19e	51.00d	17.5c	18.00ab	
Aromatic water of peppermint		1.28b	1.38b	4.92b	4.97b	57.04bc	59.07b	18.20a	18.00ab	
Aromatic water of geranium		1.26b	1.36bc	4.72c	4.81c	51.87d	53.94c	15.75e	16.00c	
Aromatic water of spearmint		1.36a	1.41ab	4.80bc	4.90bc	57.7b	59.25b	17.75bc	17.50b	
Aromatic water of rosemary		1.17d	1.31c	4.30d	4.50d	55.16c	54.50c	18.00ab	18.25a	
Aromatic water of sour orange (petit	grain)	1.39a	1.44a	5.20a	5.38a	60.58a	62.37a	16.7d	16.00c	

Different letters within each column indicate a significant difference using Duncan's Multiple Range Test at P-value of ≤ 0.05 .

Table 4: Cont.

Ch	naracter	Weight of cloves (g)/plant		Bulbing (%			vhole plant / fed.)	Yield of bulb (ton/ fed.)	
Treatment	_	1st season	2 nd season	1st season	2 nd season	1st season	2 nd season	1st season	2 nd season
Control (distilled water)		2.70e	2.83d	0.27b	0.27c	9.47e	9.78f	6.29e	6.80d
Aromatic water of peppermint		3.13cd	3.28b	0.26c	0.28b	11.38b	11.27b	7.61bc	7.88b
Aromatic water of geranium		3.29b	3.37b	0.27b	0.28b	9.93d	10.09e	6.92d	7.19c
Aromatic water of spearmint		3.25bc	3.38b	0.28a	0.29a	11.16bc	10.87c	7.69b	7.90b
Aromatic water of rosemary		3.06d	2.99c	0.27b	0.29a	10.91c	10.64d	7.35c	7.27c
Aromatic water of sour orange (petit	tgrain)	3.63a	3.90a	0.27b	0.27c	11.88a	11.81a	8.08a	8.32a

Different letters within each column indicate a significant difference using Duncan's Multiple Range Test at P-value of ≤ 0.05 .

Table 5: Bulb chemical constituents as affected by foliar spray of different aromatic water during two seasons of 2017/2018 and 2018/2019.

Charac		trogen (%)		horous %)		ssium %)		rate %)		ohur 6)		nydrate %)	Pro (%	tein %)
Treatment	1st Season	2 nd Season	1 st Season	2 nd season										
Control (distilled water)	1.88d	1.83f	0.251e	0.263d	1.32e	1.47d	221.75a	223.00a	0.392a	0.381a	47.61e	48.37e	11.75d	11.44f
Aromatic water of peppermi	nt 2.03b	2.07c	0.339a	0.364a	1.60b	1.69a	216.00d	217.00c	0.410a	0.392a	51.50c	53.71c	12.69b	12.94c
Aromatic water of geranium	1.98b	1.97d	0.294c	0.307b	1.49c	1.54c	220.50b	222.35a	0.400a	0.380a	49.07d	51.10d	12.38bc	12.31d
Aromatic water of spearming	2.16a	2.11b	0.307b	0.319b	1.56b	1.61b	218.35c	219.23b	0.385a	0.390a	54.03b	55.50b	13.50a	13.19b
Aromatic water of rosemary	1.94c	1.89e	0.274d	0.290c	1.44d	1.52cd	217.65c	217.75c	0.416a	0.401a	50.00d	50.12d	12.13c	11.81e
Aromatic water of sour oran (petitgrain)	ge 2.19a	2.23a	0.327a	0.351a	1.68a	1.73a	215.75d	214.50d	0.403a	0.388a	55.50a	57.18a	13.69a	13.94a

Different letters within each column indicate a significant difference using Duncan's Multiple Range Test at P-value of $\leq .0.05$.

Table 6: Garlic volatile oil and antioxidant activity as affected by foliar spray of different aromatic water during two seasons of 2017/2018 and 2018/2019.

Ch	aracter	Volatile oil (%)		Volat ml/p	ile oil lant	Antioxidant activity (%)		
		1st 2nd 1st 2nd				1 st	2 nd	
Treatment		season	season	season	season season		season	
Control (distilled water)		0.094c	0.111d	0.044d	0.057d	45.32d	43.60d	
Aromatic water of peppermint		0.145a	0.158a	0.083a	0.094a	51.00c	48.75c	
Aromatic water of geranium		0.101b	0.121cd	0.052cd	0.065c	46.00d	44.34d	
Aromatic water of spearmint		0.139a	0.143b	0.080a	0.085b	46.17d	45.00d	
Aromatic water of rosemary		0.104b	0.119cd	0.058bc	0.065c	57.00b	55.23b	
Aromatic water of sour orange (petitgr	ain)	0.108b	0.128c	0.065b	0.080b	68.31a	60.14a	

Different letters within each column indicate a significant difference using Duncan's Multiple Range Test at P-value of \leq .0.05.

Discussion

Because of the growing demand for natural products, it's critical to look for available and low cost of natural substances as aromatic waters (hydrosols). These natural products are promising, new and important sources that could be used as a raw material in a variety of products (Wajs-Bonikowska et al., 2015), used in food, cosmetics, human health as well as in organic agriculture and improvement growth, yield and chemical composition of plants. The above mentioned results regarding the positive effects of foliar application of aromatic waters on growth garlic parameters were in harmony with those obtained by Haridy and Soliman (2008) who found that aromatic waters showed different effects on growth, yield and essential oil of rose plant. Also, Suradkar et al. (2016) found that aromatic water of *Ipomea Carnea* flowers can be suited as fertilizer for maize plants in limited doses. In addition, aromatic waters inducing increases of plants yield through plant health improvement through their biological properties (antifungal, antibacterial and antioxidant) (Paolini et al., 2008, Ulusoy et al., 2009 and Aazza et al., 2011). The different responses of garlic plants may be attributed to the composition of aromatic waters used; phenols (Guenther, 1961) which consider as natural growth bioregulators (Vyvyan, 2002) where, phenolic compounds are an essential of plant secondary metabolites which play physiological roles through the plant life cycle. Phenolic compounds are playing key role in developmental processes like cell division, photosynthetic activity, nutrient mineralization and reproduction (Sharma et al., 2019), beside its antioxidant activity (Shen et al., 2017). Furthermore, the capacity of flavonoids that acts as antioxidants (Saxena et al., 2012). Foliar spraying with antioxidant compounds is very beneficial to growth and yield of pea plants as well as mineral and protein contents (Gad El-Hak et al., 2012), in the same harmony, (El-Hawary and Nashed, 2019) on maize plants. Linalool was a stronger antioxidant (Aazza et al., 2011), it can be degraded to terpineol, geraniol and citral (Bedoukian, 1985). Geraniol formation from linalool was β myrcene to (S)-linalool and its isomerization to geraniol (Marmulla et al., 2016) which consider the precursor of gibberellins, the main effective compound in stimulate plants growth improvement (Haridy et al., 2008 and Hamouda et al., 2009). In this concern, the formation of geraniol and geranial was observed in culture composed of linalool as sole electron and carbon source in nitrate limited (Fob and Harder, 1997). Furthermore, the increase in yield and its components could be attributable to the effect of antioxidants on promoting protein synthesis and delaying senescence (El-Bassiouny et al., 2016). Moreover, aromatic waters may be used as a tool for increasing essential oil content or essential oil yields in the crops (Zheljazkov et al., 2010). The enhancement of garlic plants in response to peptigrain aromatic water may be due to that linalool enhance GABA receptors (Milanos, 2017), this component has the ability to enhance growth and productivity of garlic through enhancing mineral composition and organic compounds (Eltohamy et al., 2019). On contrast, foliar spray of sour orange (petitgrain) aromatic water showed contrast results with Haridy and Soliman (2008) on rose plant.

Conclusion

Aromatic waters (hydrosols) are by-products of the steam distillation of the plant parts. They contain biologically active volatile compounds. Foliar application with aromatic water of sour orange

petitgrain (*Citrus aurantium* L.) exhibited increases in growth, yield and chemical components of garlic plants. While spraying with peppermint aromatic water, increased volatile oil production of garlic plants compared with those treated with water.

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