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## Research Article

# Effect of Salinity Stress and Soil Types on Growth, Photosynthetic Pigments and Essential Oil of *Artemisia annua* L.

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## Abstract

**Background and Objective:** *Artemisia annua* L. (*A. annua*) has various biological activities such as anti-malarial, anti-microbial, anti-inflammatory, anti-tumor and anti-allergenic. Salt stress and soil types (ST) are the most important conditions, which have a very high impact on yield and active principals of medicinal plants. The aim of this study was to investigate the effects of saline irrigation water (SIW), ST and their interactions on growth, yield, photosynthetic pigments (PP) and essential oil (EO) composition of *A. annua*. **Materials and Methods:** *Artemisia annua* L. plants were subjected to different levels (0.0, 1.6, 3.2, 4.7, 5.3 and 7.9 dS m<sup>-1</sup>) of SIW under two ST [clay (CLS) and sandy loam (SALS)]. Vegetative growth characters (VGC) [Plant height, PH (cm/plant), branch number, BN (per plant), fresh mass (FM) (g/plant) and dry mass (DM) (g/plant)], PP [Chlorophyll (Chl) a, Chl b and total carotenoids (TC)] and EO composition were evaluated. Randomized Complete block design (RCBD) was used and the averages of data were statistically analyzed using two-way ANOVA. **Results:** The highest values (112.75, 90.00; 32.42, 36.17; 83.15, 106.86; 45.43, 64.85) of VGC for various ST were recorded with control treatments. CLS recorded the greatest values of BN (31.13/plant), FM (79.95 g/plant) and DM (46.77 g/plant) while the highest value of PH (95.69 cm) was recorded under SALS. SALS produced higher values of Chla or Chlb (3.21 and 1.58) than CLS (3.09 and 1.47) while CLS recoded higher accumulation of TC (3.01) than SALS soil (3.00). The highest values (1.41 and 0.87%) of EO were produced under 1.6, 3.2 and 4.7 dS m<sup>-1</sup> treatments at both ST. The 4.7 dS m<sup>-1</sup> treatment reported the highest values (0.42 or 0.49 g/plant) of EO yield. 7.9 and 5.3 dS m<sup>-1</sup> treatments recorded the highest values of major components [camphor (38.67 and 38.52%) and trans caryophyllene (26.67 and 36.26)] under the both soils, on the other hand, different changes were found in monoterpene hydrocarbons (MCH), oxygenated monoterpenes (MCHO), sesquiterpene hydrocarbons (SCH) and oxygenated sesquiterpenes (SCHO). **Conclusion:** Different variations were found in VGC, PP and EO composition of *A. annua* plant under SIW, ST and their interactions.

**Key words:** *Artemisia annua* L., saline irrigation water, photosynthetic pigments, camphor, trans caryophyllene, oxygenated monoterpenes, sesquiterpene hydrocarbons

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**Competing Interest:** The authors have declared that no competing interest exists.

**Data Availability:** All relevant data are within the paper and its supporting information files.

## INTRODUCTION

*Artemisia annua* L. (*A. annua*) plant belongs to family Asteraceae<sup>1</sup>. It is commonly known as Sweet Annie, wormwood or Qing Hao and used in Chinese herbal medicine (CHM) for various treatments such as fever and malaria<sup>2</sup>. Important bioactive constituents were isolated from *A. annua* such as artemisinin, endoperoxide sesquiterpene lactone and essential oil (EO)<sup>2,3</sup>. Previous literature indicated that the leaves of *A. annua* have several biological properties i.e. antifungal, antimicrobial, antimalarial, antibacterial, anti-inflammatory, antitumor and antiallergenic<sup>3-5</sup>.

Salt stress is the major environmental factors, which has a very high impact on growth, yield and metabolism of medicinal and aromatic plants<sup>6</sup>. Plants treated with salt stress were threatened by drought, photosynthetic performance and ion imbalance. Changes in ionic homeostasis lead to certain damages at the molecular and cellular levels of plant, further causing growth arrest. Decline in photosynthetic capacity negatively affected the accumulation of dry matter<sup>7-9</sup>.

High stress levels caused significant reductions in growth characters, yield, photosynthetic attributes and total chlorophyll content of *A. annua* plants<sup>10-13</sup>. Significant reductions were reported in vegetative growth characters (VGC), yield and photosynthetic pigments (PP) under stress factors of sweet basil, American basil, apple geranium, calendula; lemon balm, black cumin and oregano herbs<sup>14-21</sup>.

Aromatic plants respond to stress conditions by different physiological defense mechanisms, for example by producing secondary metabolites, which are toxic to insects, micro-organisms and/or herbivore repellent. It was found that stress factor resulted in different changes in essential oil (EO) composition of some aromatic plants such as mint, oregano and basil<sup>4,22-24</sup>. *Artemisia annua* L. plants treated with stress doses produced higher values of EO (%) and EO constituents [ $\alpha$ -pinene, camphene, coumarin and dihydro-epi-deoxyarteannuin B]<sup>25</sup>. Essential oil (%) isolated from chamomile, lemon verbena and peppermint were increased under salinity stress levels<sup>26</sup>. The major constituents of EO extracted from sweet basil, American basil, apple geranium, calendula; lemon balm, black cumin and oregano herbs were increased under stress factors<sup>15-21</sup>.

The VGC, yield and active principals (EO) of aromatic plants differ with soil types (ST); and aromatic plants require suitable soil for greater yield and better quality<sup>27-31</sup>. Significant variations were found in VGC and EO composition of *A. annua*, thyme, calendula and roselle cultivated with different ST<sup>32-35</sup>.

More than 19% of different agricultural soils in Egypt have salt in soil or irrigation water<sup>36</sup>. In such conditions,

cultivation of resistant medicinal and aromatic plants is one way to utilize these soils. Therefore, the selection of suitable plants, which could cope with these conditions, is a necessity. In this investigation, the possible effects of saline irrigation water (SIW), ST and their interactions on VGC, yield, photosynthetic pigments (PP) and EO composition of *A. annua* were evaluated.

## MATERIALS AND METHODS

**Experimental:** Experiments were carried out in the experimental farm, Faculty of Agriculture, Cairo University and National Research Centre (NRC), Egypt, during 2014 and 2015 seasons. *Artemisia annua* L. seedlings were obtained from the Institute of Medicinal and Aromatic Plants (IMAP), Egypt. Uniform seedlings (20 cm height) were transplanted into plastic pots (30 cm diameter and 50 cm height). In the first week of March during both seasons, the pots were adjusted to natural conditions. Each pot was filled with 10 kg of air-dried soil. Three weeks after transplanting, the seedlings were thinned to three plants/pot. Pots were divided into two main groups. The first group was subjected to different levels of SIW: 0.0, 1.6, 3.2, 4.7, 5.3 and 7.9 dS m<sup>-1</sup> under clay soil (CLS) whereas the second group had been treated with the same SIW levels under sandy loam soil (SALS). All agricultural practices such as fertilization and weed control were conducted according to the main recommendations by the Ministry of Agriculture, Egypt. The soils were salinized by sea water (highly soluble sea salts were used). Content of sea water are: Cations such as Na<sup>+</sup> (55%), Mg<sup>++</sup> (3.7%), Ca<sup>++</sup> (1.2%), K<sup>+</sup> (1.1%); Anions such as Cl<sup>-</sup> (19.9%), HCO<sub>3</sub><sup>-</sup> (0.4%), CO<sub>3</sub><sup>-</sup> (0.7%), Br<sup>-</sup> (0.2), SO<sub>4</sub><sup>-</sup> (7.7%), B<sup>3-</sup> (0.1%), F<sup>-</sup> (traces). Physical and chemical properties of the soil used in this study were determined according to Margenot *et al.*<sup>37</sup> and Carter and Gregorich<sup>38</sup> and are presented in Table 1.

**Harvesting:** At full bloom, the plants were harvested. Plant height, PH (cm/plant), branch number, BN (per plant), fresh mass (FM) (g/plant) and dry mass (DM) (g/plant) were recorded.

**Determination of PP:** Chlorophyll (Chl) a, Chl b and total carotenoids (TC) in leaves collected at the first and second seasons of each treatment were determined using the methods described by the Association of Official Agricultural Chemists<sup>39</sup>.

**EO isolation:** FM (aerial part) was collected from each treatment during both seasons; air dried and weighed to

Table 1: Physical and chemical properties of soil types used in this study

Property	Soil type	
	A	B
<b>Physical</b>		
Sand (%)	32.80	68.80
Silt (%)	34.00	14.00
Clay (%)	33.20	17.20
Soil texture	Clay	Sandy loam
<b>Chemical</b>		
pH (1:2.5)	8.10	7.90
EC (dS m <sup>-1</sup> )	2.26	1.02
<b>Soluble cations (mEq L<sup>-1</sup>)</b>		
Na	11.40	2.50
K	18.13	0.20
Ca	4.50	5.50
Mg	2.50	2.00
<b>Soluble anions (mEq L<sup>-1</sup>)</b>		
Cl	26.50	3.00
HCO <sub>3</sub>	2.30	3.45
CO <sub>3</sub>	0.00	0.00
SO <sub>4</sub>	7.90	3.80
N (ppm)	666.7	24.20
P (ppm)	518.0	2.40
K (ppm)	1487.1	581.90

extract the EO, then 100 g from each replicate of all treatments was subjected to hydro-distillation (HD) for 3 h using a Clevenger-type apparatus<sup>40</sup>. The EO content was calculated as a relative percentage (v/w). In addition, total EO as mL/plant was calculated by using the DM. The EOs extracted from *A. annua* were collected in both seasons from each treatment and dried over anhydrous sodium sulfate to identify the chemical constituents.

**Gas chromatography-mass spectrometry (GC-MS):** The GC-MS analysis was carried out with an Agilent 5975 GC-MSD system. DB-5 column (60 m×0.25 mm, 0.25 mm film thickness) was used with helium as carrier gas (0.8 mL min<sup>-1</sup>). GC oven temperature was kept at 60°C for 10 min and programmed to 220°C at a rate of 4°C/min that was kept constant at 220°C for 10 min and followed by elevating the temperature to 240°C at a rate of 1°C/min. Split ratio was adjusted at 40:1. The injector temperature was set at 250°C. Mass spectra were recorded at 70 eV. Mass range was m/z 35-450.

**GC analysis:** The GC analysis was carried out using an Agilent 6890N GC system using FID detector temperature of 300°C. To obtain the same elution order with GC-MS, simultaneous auto injection was done on a duplicate of the same column at the same operational conditions. Relative percentage amounts of the separated compounds were calculated from FID chromatograms.

**Identification of components:** Identification of the EO components was carried out by comparison of their relative retention times with those of authentic samples or by comparison of their retention index (RI) to series of n-alkanes. Computer matching against commercial (Wiley GC/MS Library, Mass Finder 3 Library)<sup>41-42</sup> and in-house "Baser Library of EO Constituents" built up by genuine compounds and components of known oils. Additionally, MS literature data<sup>43-44</sup> was also used for the identification.

**Statistical analysis:** In this experiment, 2 factors were considered; i.e. saline irrigation water (0.0, 1.6, 3.2, 4.7, 5.3 and 7.9 dS m<sup>-1</sup>) and two soil types. For each treatment there were 4 replicates, each of which had 10 pots; in each pot 3 individual plants were planted. The experimental design followed a complete random block design. According to De Smith<sup>45</sup>, the averages of data were statistically analyzed using 2-way analysis of variance (ANOVA). Significant values were determined according to LSD (0.05). The applications of that technique were according to the STAT-ITCF program<sup>46</sup>.

## RESULTS

**Effect of SIW, ST and their interaction on VGC:** SIW, various ST and their interactions affected VGC [PH (cm/plant), BN (per plant), FM (g/plant) and DM (g/plant)] of *A. annua* plants (Table 2). Thus the different VGC were decreased under the different doses of SIW of both ST. Control treatment recorded the greatest VGC with the values of 112.75, 90.00; 32.42, 36.17; 83.15, 106.86; 45.43, 64.85 for various ST. SALS recorded the highest values of PH (95.69) while CLS produced the greatest values of BN (31.13), FM (79.95) and DM (46.77). Changes in all VGC were significant for SIW or ST. SIW×ST resulted in insignificant changes in all VGC except the DM (were significant).

**Effect of SIW, ST and their interaction on PP:** Data in Table 2 revealed that applying SIW caused a reduction in the accumulation of PP (Chl a, Chl b and TC). Control treatment resulted in the highest accumulation in PP with the values of 3.69, 3.58; 1.81, 1.51; 3.36, 3.13 under different ST. SALS reported higher amounts of Chl a or Chl b (3.21 and 1.58) than CLS (3.09 and 1.47) while CLS recoded higher accumulation of TC (3.01) than SALS (3.00). Changes in all PP were significant for SIW but it were insignificant for ST or SIW×ST.

**Effect of SIW, ST and their interaction on EO contents:** The EO contents (% or mL/plant) were affected at various SIW treatments or different ST (Table 2). The highest EO

Table 2: Effect of SIW, ST and their interactions on VGC, EO and PP

Treatments	VGC					EO	Yield	PP		
	SIW (dS m <sup>-1</sup> )	PH (cm/plant)	BN (per plant)	FM	DM			Chl a	Chl b	TC
				(g/plant)						
SALS	0.0	112.75	32.42	83.15	45.43	0.86	0.34	3.69	1.81	3.36
	1.6	97.75	25.17	71.85	41.25	0.79	0.41	3.33	1.74	3.17
	3.2	92.83	24.92	69.53	40.10	0.90	0.38	3.18	1.53	3.02
	4.7	89.67	24.75	68.72	39.10	1.41	0.42	3.08	1.47	2.91
	5.3	93.33	24.00	64.05	37.25	1.05	0.40	3.04	1.63	2.94
	7.9	87.83	23.50	58.73	35.10	0.61	0.23	2.92	1.30	2.57
Overall (SALS)		95.69	25.79	69.34	39.71	0.94	0.36	3.21	1.58	3.00
CLS	0.0	90.00	36.17	106.86	64.85	0.82	0.49	3.58	1.51	3.13
	1.6	87.50	35.17	79.56	48.85	0.87	0.39	3.40	1.50	3.07
	3.2	83.33	31.25	77.10	42.45	0.87	0.38	2.96	1.50	3.04
	4.7	86.67	29.00	75.55	41.05	0.87	0.35	2.87	1.42	2.99
	5.3	83.67	27.50	71.40	44.05	0.77	0.33	3.06	1.53	3.05
	7.9	74.67	27.67	69.20	39.35	0.71	0.29	2.64	1.38	2.79
Overall (CLS)		84.31	31.13	79.95	46.77	0.82	0.37	3.09	1.47	3.01
Overall (SIW)	0.0	101.38	34.30	95.01	55.14	0.84	0.42	3.64	1.66	3.25
	1.6	92.63	30.17	75.71	45.05	0.83	0.40	3.37	1.62	3.12
	3.2	88.08	28.09	73.32	41.28	0.89	0.38	3.07	1.52	3.03
	4.7	88.17	26.88	72.14	40.08	1.14	0.39	2.98	1.45	2.95
	5.3	88.50	25.75	67.73	40.65	0.91	0.37	3.05	1.58	3.00
	7.9	81.25	25.59	63.97	37.23	0.66	0.26	2.78	1.34	2.68
<b>LSD at 0.05</b>										
SIW		5.85	4.37	8.16	2.90	0.03	0.03	0.15	0.17	0.15
ST		3.38	2.52	4.71	1.68	NS	NS	NS	NS	NS
SIW×ST		NS	NS	NS	4.10	0.04	0.04	NS	NS	NS

SIW: Saline irrigation water, ST: Soil type, SALS: Sandy loam soil, CLS: Clay soil, VGC: Vegetative growth characters, EO: Essential oil, PP: Photosynthetic pigments, PH: Plant height, BN: Branch number, FM: Fresh mass, DM: Dry mass, PP: Photosynthetic pigments, Chl a: Chlorophyll a, Chl b: Chlorophyll b, TC: Total carotenoids

percentages (1.41 and 0.87%) were obtained with treatments of 1.6, 3.2 and 4.7 dS m<sup>-1</sup> of SIW under different ST. SALS recorded the higher EO percentage (0.94%) than CLS (0.82%). The greatest EO yield (0.42 or 0.49 g/plant) resulted from 4.7 dS m<sup>-1</sup> treatment or control under both ST. The variations of EO contents were significant for SIW treatments or SIW×ST while it were insignificant for both ST.

#### Effect of SIW, ST and their interaction on EO components:

Quantity and quality of the components determined with SIW, ST and their interactions in hydro distilled *A. annua* EO were investigated (Table 3-5). A total of 20 components ranging from 75.07-94.93% of the total EO were detected by GC/MS and divided into four chemical fractions such as monoterpene hydrocarbons (MCH), oxygenated monoterpenes (MCHO), sesquiterpene hydrocarbons (SCH) and oxygenated sesquiterpenes (SCHO) (Table 3-5). MCHO and SCH were the major fractions (more than 60%) followed by MCH fraction while SCHO formed as minor fraction. Camphor and trans caryophyllene were identified as major constituents which gave the greatest amounts (more than 50%) with all SIW, soil types and SIW×soil types.

The SIW×ST resulted in various changes in *A. annua* EO constituents, the major components were increased with the increasing of SIW levels for both ST (Table 3-5). The highest amounts of major components [camphor (38.67, 38.52%) and trans caryophyllene (26.67, 36.26)] were obtained from the treatments of 7.9 and 5.3 dS m<sup>-1</sup> for CLS and SALS respectively. The highest amounts of MCH (21.4, 9.23%) had been obtained at 4.7 dS m<sup>-1</sup> level, while 7.9 dS m<sup>-1</sup> gave the highest values (46.10, 42.33%) of MCHO. The highest amounts (40.02, 42.31%) of SCH were reported with 7.9 and 5.3 dS m<sup>-1</sup> treatments. Application of 3.2 dS m<sup>-1</sup> and control treatments reported the greatest values of SCHO (2.50, 3.09%).

It was found that SALS resulted in the greatest contents of camphor, trans caryophyllene, MCHO, SCH and SCHO with the values of 35.90, 29.33, 40.11, 36.26 and 1.45%, respectively (Table 5). CLS gave the highest content of MCH (11.83%). Regarding to SIW the treatment of 7.9 dS m<sup>-1</sup> resulted in the greatest values of camphor (38.32%) and MCHO (44.22%). The highest amounts of trans caryophyllene (30.38%) and SCH (40.20%) were recorded for 5.3 dS m<sup>-1</sup> treatment. MCH (15.33%) and SCHO (1.75) were obtained as the highest values for 3.2 and 4.7 dS m<sup>-1</sup> treatments.

Table 3: Effect of SIW on EO constituents under CLS

Components (%)	RI	Class	SIW (dS m <sup>-1</sup> )					
			0.0	1.6	3.2	4.7	5.3	7.9
α-Thujene	931	MCH	1.03	0.53	0.78	6.53	6.08	2.09
α-Pinene	939	MCH	2.15	2.34	0.19	5.24	3.99	1.83
Camphene	953	MCH	0.72	7.50	0.32	3.28	1.42	1.26
β-Pinene	980	MCH	0.74	1.05	0.79	3.10	3.13	0.40
2-Hydroxy-1,8-cineole	1033	MCHO	3.04	1.90	0.80	1.31	2.27	3.23
γ-Terpinene	1062	MCH	0.82	0.61	1.48	1.68	1.03	0.96
β-Terpinene	1071	MCH	3.05	0.67	1.41	1.57	0.49	0.67
Camphor	1143	MCHO	31.30	32.79	32.80	31.77	34.59	38.67
Borneol	1165	MCHO	0.76	0.80	3.78	1.95	1.48	0.77
γ-Terpinen-4-ol	1177	MCHO	0.69	1.37	1.28	1.62	0.71	1.40
α-Cubebene	1351	SCH	0.61	1.31	0.38	1.15	1.03	0.34
Eugenol	1356	MCHO	2.06	0.88	0.47	1.06	1.16	2.03
β-Elementene	1375	SCH	0.71	1.0	1.48	1.34	0.96	0.68
α-Copaene	1376	SCH	0.97	0.82	2.10	7.39	6.55	7.43
Longifolene	1402	SCH	0.90	1.43	0.89	2.24	0.96	0.79
Trans caryophyllene	1418	SCH	21.76	21.82	22.88	19.14	24.49	26.67
E-β-farnesene	1458	SCH	1.61	0.97	0.29	3.05	2.69	2.25
β-Selinene	1485	SCH	0.71	0.77	0.24	0.04	0.54	0.24
γ-Cadinene	1513	SCH	0.83	0.57	0.50	0.04	0.77	1.62
Caryophyllene oxide	1581	SCHO	0.61	1.02	2.50	0.04	0.59	0.55
MCH			8.51	12.70	4.97	21.40	16.14	7.21
MCHO			37.85	37.74	39.13	37.71	40.21	46.10
SCH			28.10	28.69	28.76	34.39	37.99	40.02
SCHO			0.61	1.02	2.50	0.04	0.59	0.55
TI			75.07	80.15	75.36	93.54	94.93	93.88

SIW: Saline irrigation water, ST: Soil type, CLS: Clay soil, RI: Retention index, MCH: Monoterpene hydrocarbons, MCHO: Oxygenated monoterpenes, SCH: Sesquiterpene hydrocarbons, SCHO: Oxygenated sesquiterpenes, TI: Total identified

Table 4: Effect of SIW on EO constituents under SALS

Components (%)	RI	Class	SIW (dS m <sup>-1</sup> )					
			0.0	1.6	3.2	4.7	5.3	7.9
α-Thujene	931	MCH	0.92	0.87	0.73	1.44	0.57	0.64
α-Pinene	939	MCH	0.95	0.80	1.30	1.13	1.20	1.67
Camphene	953	MCH	0.66	0.83	0.89	1.56	0.82	0.54
β-Pinene	980	MCH	1.32	0.96	3.62	1.22	1.31	1.37
2-Hydroxy-1,8-cineole	1033	MCHO	0.61	0.93	1.35	1.41	0.65	0.67
γ-Terpinene	1062	MCH	1.20	0.59	0.65	3.19	0.66	0.56
β-Terpinene	1071	MCH	0.66	0.71	1.06	0.69	0.60	1.22
Camphor	1143	MCHO	32.60	33.68	35.87	36.75	38.52	37.96
Borneol	1165	MCHO	0.59	0.58	0.67	0.87	0.54	2.01
γ-Terpinen-4-ol	1177	MCHO	0.67	1.77	2.11	1.63	1.31	0.84
α-Cubebene	1351	SCH	0.65	1.00	1.27	0.77	0.56	0.57
Eugenol	1356	MCHO	0.90	0.88	1.05	1.00	1.30	0.85
β-Elementene	1375	SCH	2.32	1.47	1.16	1.78	0.98	1.57
α-Copaene	1376	SCH	0.90	0.64	0.77	1.05	1.56	1.12
Longifolene	1402	SCH	0.79	0.63	0.92	1.04	0.59	1.93
Trans caryophyllene	1418	SCH	24.86	25.92	30.29	30.82	36.26	27.85
E-β-Farnesene	1458	SCH	1.35	0.64	0.80	0.77	0.82	1.19
β-Selinene	1485	SCH	0.67	0.70	0.62	1.00	0.91	0.60
γ-Cadinene	1513	SCH	1.06	0.98	0.85	1.00	0.63	0.92
Caryophyllene oxide	1581	SCHO	3.09	1.91	1.01	0.90	0.63	1.13
MCH			5.71	4.76	8.25	9.23	5.16	6.00
MCHO			35.37	37.84	41.05	41.66	42.32	42.33
SCH			32.60	31.98	36.68	38.23	42.31	35.75
SCHO			3.09	1.91	1.01	0.90	0.63	1.13
TI			76.77	76.49	86.99	90.02	90.42	85.21

SIW: Saline irrigation water, ST: Soil type, SALS: Sandy loam soil, RI: Retention index, MCH: Monoterpene hydrocarbons, MCHO: Oxygenated monoterpenes, SCH: Sesquiterpene hydrocarbons, SCHO: Oxygenated sesquiterpenes, TI: Total identified

Table 5: Effect of SIW or ST on EO constituents

Components (%)	RI	Class	ST		SIW (dS m <sup>-1</sup> )					
			CLS	SALS	0.0	1.6	3.2	4.7	5.3	7.9
α-Thujene	931	MCH	2.84	0.86	0.98	0.70	0.76	3.99	3.33	1.37
α-Pinene	939	MCH	2.62	1.18	1.55	1.57	0.75	3.19	2.60	1.75
Camphene	953	MCH	2.42	0.88	0.69	4.17	0.61	2.42	1.12	0.90
β-Pinene	980	MCH	1.54	1.60	1.03	1.01	2.21	2.16	2.22	0.89
2-Hydroxy-1,8-cineole	1033	MCHO	2.09	0.94	1.83	1.42	1.08	1.36	1.47	1.95
γ-Terpinene	1062	MCH	1.10	1.14	1.01	0.60	1.07	2.44	0.85	0.76
β-Terpinene	1071	MCH	1.31	0.82	1.86	0.69	1.24	1.13	0.55	0.95
Camphor	1143	MCHO	33.65	35.90	31.95	33.24	34.34	34.26	36.56	38.32
Borneol	1165	MCHO	1.59	0.88	0.68	0.69	2.23	1.41	1.01	1.39
γ-Terpinen-4-ol	1177	MCHO	1.18	1.39	0.68	1.57	1.70	1.63	1.01	1.12
α-Cubebene	1351	SCH	0.80	0.80	0.63	1.16	0.83	0.96	0.80	0.46
Eugenol	1356	MCHO	1.28	1.00	1.48	0.88	0.76	1.03	1.23	1.44
β-Elemene	1375	SCH	1.03	1.55	1.53	1.24	1.32	1.56	0.97	1.13
α-Copaene	1376	SCH	4.21	1.01	0.94	0.73	1.44	4.22	4.06	4.28
Longifilene	1402	SCH	1.20	0.98	0.85	1.03	0.91	1.64	0.78	1.36
Trans caryophyllene	1418	SCH	22.79	29.33	23.31	23.87	26.59	24.98	30.38	27.26
E-β-Farnesene	1458	SCH	1.81	0.93	1.48	0.81	0.55	1.91	1.78	1.72
β-Selinene	1485	SCH	0.42	0.75	0.69	0.74	0.43	0.52	0.73	0.42
γ-Cadinene	1513	SCH	0.72	0.91	0.95	0.78	0.68	0.52	0.70	1.27
Caryophyllene oxide	1581	SCHO	0.89	1.45	1.85	1.47	1.75	0.47	0.61	0.84
MCH			11.83	6.48	7.12	8.74	6.64	15.33	10.67	6.62
MCHO			39.79	40.11	36.62	37.8	40.11	39.69	41.28	44.22
SCH			32.98	36.26	30.38	30.36	32.75	36.31	40.20	37.9
SCHO			0.89	1.45	1.85	1.47	1.75	0.47	0.61	0.84
TI			85.49	84.3	75.97	78.37	81.25	91.8	92.76	89.58

SIW: Saline irrigation water, ST: Soil type, SALS: Sandy loam soil, CLS: Clay soil, RI: Retention index, MCH: Monoterpene hydrocarbons, MCHO: Oxygenated monoterpenes, SCH: Sesquiterpene hydrocarbons, SCHO: Oxygenated sesquiterpenes, TI: Total identified

## DISCUSSION

The obtained results indicated that SIW caused an inhibition in VGC, PP and EO yield while the EO (%) and major components of EO were increased. The ST resulted in various changes in VGC, PP and EO composition. The reduction of VGC under SIW conditions may be due to exposure to harmful doses of SIW causing a reduction of turgidity leading to reduction of VGC and development of cells, especially in plant aerial part<sup>47</sup>. Salinity stress damages chloroplasts and the reduce values of PP<sup>48</sup>. With salt stress factors the amount of the proline increased. PP and proline are both synthesized from the same substrate, thus an increase in the synthesis of proline leads to reduction PP synthesis<sup>48</sup>, which leads to reduction in dry matter content of leaves and stems<sup>49</sup>. The increase in EO (%) and its major constituents under salinity stress may be due to the increase in activation of the biosynthesis of its content in the secretory and/or in the size of the glandular trichomes<sup>50</sup>. The reduction in EO yields under SIW levels may be explained by increasing osmotic pressure, which may affect the uptake of water and nutrients for the plant, thus changing the physiological and biochemical potential of the plant and hence the production of secondary

metabolites<sup>51</sup>. On the other hand, the changes in EO composition (contents and components) under the different treatments (SIW, ST and their interactions) may be due to the changes in enzymes activity and metabolites<sup>52</sup>. The variations in VGC, PP and EO composition may be due to soil properties which are very necessary for plant growth and chemical contents. Soil represents a complicated physiochemical and biological system by which the plant is supplied by the moisture, elements and oxygen it necessary for plant development and metabolites activities<sup>31</sup>. The obtained results are in accordance with those reported by some previous investigators on some medicinal plants (basil, geranium, calendula; lemon balm, black cumin and oregano) i.e. VGC, PP and EO yield (per plant) were significantly decreased under salinity stress conditions while EO (%) and its major constituents of EO were increased<sup>15-21</sup>. ST caused different changes in VGC, PP and EO composition of *A. annua*, thyme, calendula and roselle plants<sup>32-35</sup>.

## CONCLUSION

It may be concluded that SIW, ST and their interactions resulted in different variation in VGC, PP and EO composition

of *A. annua* plant. The highest values of VGC, PP and EO yield were recorded with control (0.0 dS m<sup>-1</sup>). The application of 1.6, 3.2 and 4.7 dS m<sup>-1</sup> resulted in the greatest values of EO (%). The highest value of PH was recorded with SALS while CLS gave the highest values of BN, FM and DM. Variations of the EO composition had been related to the different ST.

### SIGNIFICANCE STATEMENTS

Adapting *A. annua* to SIW factor under various ST is very necessary especially in arid and semi arid regions for increasing the natural resources in Egypt. *A. annua* used for various treatments such as fever and malaria. In the future, results of this study will be introduced to Ministry of Agriculture, farmers and producers to help them in production yield, EO and EO major components from *A. annua* (important medicinal herb) under various ST in Egypt.

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