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RESPONSE OF ANNUAL WORMWOOD (*ARTEMISIA ANNUA* L.) FOR NITROGEN FERTILIZER SOURCES AND SPRAYING WITH SALICYLIC ACID

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The objective of the study was to maximize the growth, production and quality of annual wormwood (*Artemisia annua* L.) plants, which are highly valued for their bioactive compounds, particularly artemisinin, known for its potent anti-malarial properties The field experiments were conducted in the Experimental Station of Desert Research Center in Ras Sudr, South Sinai Governorate, Egypt, during the two successive 2021 and 2022 seasons to investigate the effect of nitrogen sources (NS) [urea, ammonium nitrate (AN) and ammonium sulfate (AMS)] as a soil application at the rate of 160 kg N/ha additionally to control and salicylic acid (SA) (0, 1, and 2 mM) on *Artemisia annua* plant growth, productivity and chemical composition. Based on the results, AN combined with 2 mM SA treatment resulted in the greatest plant growth characteristics, chemical composition, and yield. Plant height, branch number, dry weight, and essential oil content were all significantly enhanced by this combination. The dry herb yield/ha reached 6822 and 8325 kg in the first season's cuts, and 6383 and 8262 kg in the second season's cuts, respectively. The essential oil yield/ha was notably enhanced, with the highest yields recorded at 76.61 and 89.15 kg/ha in the first season and 79.20 and 93.30 kg/ha in the second season across the cuts. These results indicate that the combined application of AN and 2 mM SA is the most effective treatment for enhancing the growth and productivity of the *Artemisia annua* plant under the conditions of South Sinai, Egypt.

Keywords: annual wormwood, *Artemisia annua*, nitrogen sources, salicylic acid, urea, ammonium nitrate, ammonium sulfate

INTRODUCTION

Annual wormwood (*Artemisia annua* L.) is a member of the family Asteraceae, a highly valued medicinal plant known for its rich composition of bioactive compounds and extensive medicinal benefits. This plant has garnered significant attention due to its primary active ingredient, artemisinin, widely recognized for its potent anti-malarial properties (Soni et al., 2022 and Ferreira et al., 2024). A notable increase in interest in the chemical makeup and biological activity of different species of the genus Artemisia has resulted from the 2015 Nobel Prize in Medicine being given for the discovery of the sesquiterpene lactone artemisinin, found in *A. annua*, and demonstrating the efficacy of its antimalarial action (Efferth et al., 2015). The plant produces artemisinin-based combination therapies the most popular antimalarial medicines (WHO, 2019 and Soni et al., 2022). In addition to its effectiveness against malaria, it also exhibits a broad spectrum of pharmacological activities including anti-inflammatory, enhancing wound healing, treating skin and having antibacterial properties (Mirbehbahani et al., 2020; Chebbac et al., 2023; Saunoriūtė et al., 2023 and Shaaban et al., 2024). Artemisia contains flavonoids such as quercetin and kaempferol, which have anti-inflammatory and antiviral activities, especially, COVID-19 (Dong et al., 2020; Nair et al., 2021; WHO, 2021 and Tang et al., 2022). Volatile oil is rich in compounds like artemisia ketone, which have shown significant antimicrobial activities against bacteria and fungi (Hong et al., 2023 and Ma et al., 2023).

Nitrogen (N) is a vital macro element for the growth and development of plants (Amin, 2011 and Dong et al., 2022). It gives plants the energy they need to grow and develop by constructing several organic substances, such as proteins, enzymes, chlorophyll, amino acids and nucleic acids. Plants can produce fewer essential oils when there is an N shortage, thus, increasing the rate at which N fertilizer is applied can boost the production of essential oils (Nurzyńska-Wierdak, 2013; Madandoust and Fooladchang, 2018 and Alhasan and Al-Ameri, 2021). Availability of N in soil fluctuates in both place and time because of environmental factors, including soil temperature, precipitation, wind, and soil type and pH. Therefore, the preferred N form in plants depends on plant adaptation to soil conditions. As an illustration, plants cultivated in acidic soil typically absorb ammonium (A), whereas plants produced in aerobic soil typically absorb nitrate (Masclaux-Daubresse et al., 2010). Urea (46.5% N) was usually used as the source of N because it has stable chemical properties, is commonly used in agriculture. According to recent research, urea greatly increases the growth and productivity of therapeutic plants, including the natural sweetener of *Stevia rebaudiana* (Śniegowska et al., 2024). Most of the other studies recorded the enhanced effect of urea on plant growth and productivity (Dong et al., 2022; Swify et al., 2022 and Sun et al., 2023). Ammonium nitrate (AN), 33.5% N is a universal and agrochemically valuable N fertilizer that provides N in a form that is immediately available to plants. Nitrate is the most mobile form of N and easily dissolves in water. It is effective on all types of soils and for almost all crops (Jabborova et al., 2024). Previous studies showed that, applying AN fertilizer can increase different plant parameters, including plant height, number of branches, essential oil content, total phenolic and flavonoids (Basal et al., 2019; Ahmadi et al., 2020; Zayed and Dawayati, 2021 and Sun et al., 2023). Ammonium sulfate (AMS), 21% N has a number of environmental and agricultural advantages compared to urea and AN. These include the absence of potential plant toxicity from aqueous ammonia and nitrite in alkaline soils, the prevention of N loss in acidic or neutral soils through ammonia volatilization, and the improvement of saline soils' N supply by decreasing the detrimental effects of NaCl on plant growth. A study conducted on flax showed that N and AMS were more effective than urea in increasing seed yield and quality in dry environments (Emam, 2019). Furthermore, by reducing nitrate leaching and increasing soil phosphorus and micronutrient availability through soil acidification, AMS enhances soil structure (Chien et al., 2011). The previous studies revealed the effect of AMS on medicinal and aromatic plants (Aziz et al., 2010; Basal et al., 2019 and Berhe et al., 2019).

Salicylic acid (SA) is a vital phenolic compound with a molecular formula of $C_7H_6O_3$ extensively involved in plant growth, development and stress responses. SA a signaling molecule and plant hormone, regulates a variety of physiological processes, including plant growth and development, nutrients uptake, photosynthesis, germination and defensive responses (Okuma et al., 2014; Filgueiras et al., 2019 and El-Naggar et al., 2024). Studies support the use of SA in agriculture to improve plant resistance to abiotic challenges such as salt, drought and high temperatures (Alamer and Fayez, 2020 and Ali, 2021). Plants treated with SA have higher levels of antioxidant activity and stress tolerance as a result of the accumulation of hydroxyl cinnamic acid and flavonoids, especially in temperate areas where environmental conditions may restrict the formation of these compounds (Janda et al., 2017 and Salem et al., 2021). SA application enhances crop yield and quality and contributes to the sustainable management of agricultural systems by increasing the generation of secondary metabolites and fortifying plant defense mechanisms (Skrypnik et al., 2022). Previous research on medicinal and aromatic plants has shown that SA enhances plant growth features and boosts plant production (Es-sbihi et al., 2020; Gahory et al., 2022; Nasseralla and Almukhtar, 2023 and El-Naggar et al., 2024). This study aimed to evaluate the impact of N fertilizer sources and spraying with SA on annual wormwood (*Artemisia annua*) under South Sinai Governorate, Egypt conditions.

MATERIAL AND METHODS

1. Site Description, Plant Materials and Treatments

This study was conducted in the Experimental Station of Desert Research Center in Ras Sudr, South Sinai, Egypt (29°37'28.0"N 32°42'46.0"E), during the two successive summer seasons 2021 and 2022 to investigate the effect of N fertilizer sources and spraying with SA on annual wormwood (*Artemisia annua*). In the experimental field, the soil has a sandy loam texture. Its pH is 7.5, its EC is 4.9 ds cm^{-1}, its $CaCO_3$ concentration is 52.35%, and 0.89% organic matter with 1.51 g kg^{-1} of organic carbon in the 0-30 cm soil layer. Selected seedlings of *Artemisia annua* were obtained from Ornamental, Medicinal and Aromatic Plants Farm, Department of Horticulture, Faculty of Agriculture, Benha University, and the planting process was achieved on the 13th and 14th of March in the 1st and 2nd seasons, respectively. The seedlings were cultivated at 30 cm distances between plants and 60 cm between rows, (55500 plant/ha), approximately. Day after day, all plants received 30-minute irrigations with salty irrigation water (EC = 6.23 ds cm^{-1}). NS (urea 46.5% N and AN 33.5% N and AMS 21% N) were used as a soil application at the rate of 160 kg N/ha additionally to control and three rates of SA (control, 1 mM and 2 mM) were done as a foliar application, the control treatment was sparing with tap water. Sulfur was added to other N fertilization treatments in an equivalent quantity to AMS. Monthly, foliar application of SA was randomly in distributed in main plots, while the NS were randomly arranged in the sub main plots. The quantity of N fertilizers was divided into two equal portions as side dressing at 30 and 60 days after planting for the 1st cut and after cutting for the 2nd cut beside plants. All agricultural practices were carried out as usually recommended for *Artemisia annua* production in Egypt. The plants were cut twice in each season at the stage of full-blooming by cutting the vegetative parts at 10-15 cm above the soil surface. The 1st cut was done on the 17th of July, while, the 2nd cut was done on the 8th of October for the 2021 and 2022 seasons.

2. Experiment Layout

This experiment used a Randomized Complete Block Design (RCBD) layout with two factors, a split plot design, and three replicates. The first factor was three SA spraying treatments and the second was four nitrogen sources (NS) treatments, all treatments had three replicates, and each replicate contained 10 plants.

3. Data Recorded

3.1. Vegetative growth

The plant height (cm), branches number/plant, dry weight per plant (g) and per hectare (kg), dry weight of leaves (g) per plant and per hectare (kg).

3.2. Chemical composition

The N, P, K, and total carbohydrates were determined in the dried leaves at the flowering stage according to A.O.A.C. (1970), Murphy and Riley (1962), Cottenie et al. (1982), Chaplin and Kennedy, (1994), respectively. Furthermore, the essential oil percentage was determined as described in the British Pharmacopoeia (1963). The determination of artemisinin was according to the method described by Bilia et al. (2006).

4. Statistical Analysis

The means of all obtained data from the studied factors were subjected to analyses of variance (ANOVA) as a factorial experiment in a complete randomized block design. For means' comparison, Duncan's multiple range test ($p < 0.05$ significance level) was used for both factors (NS and SA spraying) using the MSTAT-C statistical software package (Duncan, 1955) where each examined characteristic's means of treatment were distinguished using capital letters.

RESULTS AND DISCUSSION

1. Plant Height (cm) and Number of Branches/Plant of *Artemisia annua*

The findings indicated that, SA significantly impacted plant height and the number of branches in *Artemisia annua* plants across both cuts in each season (Table 1). In both cuts during two seasons, plants treated with 2 mM SA exhibited the tallest heights and the most branches per plant, outperforming the control treatments. Among the NS, AN was the most effective in increasing plant height, in the case of the number of branches per plant the effect of AN and AMS had no significant differences in both seasons across two cuts. AN when combined with 2 mM SA resulted in the tallest plants and the highest number of branches per plant in both seasons and across both cuts. The interaction between SA and NS showed that the combination of AN and 2 mM SA was the most effective treatment for maximizing plant height and the number of branches per plant across both cuts and both seasons..

2. Dry Weight and Dry Leaves of *Artemisia annua*

Data recorded in Tables (2 and 3) obviously indicate that, SA significantly impacted the dry weight per plant and per hectare, as well as dry leaves per plant and per hectare in *Artemisia annua* plants across both cuts during two seasons. The application of 2 mM SA consistently resulted in the highest values for these parameters, significantly outperforming the control treatments. Regarding NS, AN was the most effective in increasing dry weight per plant and per hectare, as well as the highest dry leaves per plant and per hectare in both seasons and across both cuts without significant differences between it and AMS in the 2nd cut only in both seasons.

It is obvious from the data that, SA and NS interaction showed that, the combination of AN and 2 mM SA was the most effective treatment for maximizing dry weight and dry leaves yield. The recorded values of dry weight per hectare in the 1st season were 6822 and 8325 kg and the corresponding values were 6383 and 8262 kg for the 2nd season during the first and second cuts, respectively, with significant differences among most treatments. For dry leaves per hectare, the maximum values in the 1st season were 1770 and 2405 kg and the corresponding values were 1739 and 2168 kg for the 2nd season during the first and second cuts, respectively, with a significant difference among most treatments. The combined application of AN and 2 mM SA consistently produced the best results, for optimizing the productivity of *Artemisia annua*

Table (1). Effect of salicylic acid, nitrogen sources and their interaction treatments on plant height (cm) and number of branches *Artemisia annua* L. plant during 2021 and 2022 seasons.

SA / NS	Plant height (cm)								Number of branches/plant							
	1st season				2nd season				1st season				2nd season			
	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean
1st cut																
Control	171.2f	177.3f	179.8f	176.1B	172.3d	173.0d	178.3d	174.6D	28.67g	32.33f	34.33f	31.78C	28.00g	31.33fg	34.67ef	31.33C
Urea	181.2ef	182.7ef	184.4def	182.9B	181.7d	183.3d	182.0d	182.3C	37.67e	41.33d	43.67d	40.89B	38.33de	41.67d	40.67d	40.22B
AN	196.6bcd	209.2ab	214.2a	207.7A	198.0c	213.0ab	222.7a	211.2A	49.00c	54.33a	52.67ab	52.00A	47.67c	54.67a	55.00a	52.44A
AMS	194.7cde	208.0abc	207.7abc	20.35A	197.0c	203.7bc	204.3bc	201.7B	48.33c	53.00ab	51.00bc	50.78A	20.55bc	53.67ab	52.33ab	52.00A
Mean	186.0B	194.3A	197.3A		187.3A	193.3A	196.8A		40.92B	45.25A	45.42A		40.00B	45.33A	45.67A	
2nd cut																
Control	175.7f	183.0ef	184.7ef	181.1C	173.7h	180.7h	179.3h	177.9D	57.00f	60.67ef	66.00e	61.22C	56.33f	59.00f	68.33e	61.22C
Urea	187.0e	190.de	191.0de	189.3B	189.0g	188.7g	192.3fg	190.0C	73.67d	79.33cd	85.67bc	79.56B	72.67e	78.33d	86.33c	79.11B
AN	198.7cd	216.0b	228.7a	214.4A	200.0de	2013.7b	224.2a	212.6A	92.67b	104.0a	107.7a	101.4A	91.00bc	107.3a	109.0a	102.40A
AMS	200.3c	213.0b	218.3b	210.6A	196.3ef	205.7cd	208.7bc	203.6B	92.67b	102.0a	104.3a	99.67A	93.67b	107.3a	108.7a	103.20A
Mean	190.4B	200.5A	205.7A		189.8C	197.2B	201.1A		79.00C	86.50B	90.92A		78.42C	88.00B	93.08A	

SA = Salicylic acid, NS = Nitrogen source, AN = Ammonium nitrate, AMS = Ammonium sulfate

Table (2). Effect of salicylic acid, nitrogen sources and their interaction treatments on dry weight/plant (g) and dry weight/ha (kg) of *Artemisia annua* L. plant during 2021 and 2022 seasons.

SA / NS	Dry weight/plant (g)								Dry weight/ha (kg)							
	1st season				2nd season				1st season				2nd season			
	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean
1st cut																
Control	71.00f	79.79e	83.00de	77.93C	73.33g	79.00fg	84.33ef	78.89C	3941f	4428e	4607de	4325C	4070g	4385fg	4681ef	4378C
Urea	85.67de	89.67d	90.34d	88.49B	88.00e	90.00de	89.33e	89.11B	4755de	4965d	5014d	4911B	4884e	4995de	4958e	4946B
AN	99.31c	115.0ab	122.9a	112.4A	103.7bc	108.7ab	115.0a	109.1A	5512c	6382ab	6822a	6239A	5754bc	6031ab	6383a	6056A
AMS	98.93c	113.4b	119.0ab	110.4A	98.33cd	106.7abc	107.7ab	104.2A	5491c	6292b	6606ab	6130A	5458cd	5920abc	5976ab	5784A
Mean	88.73B	77.40A	103.8A		90.83B	96.08A	99.08A		4924B	5517A	5762A		5041B	5333A	5499A	
2nd cut																
Control	75.33h	83.67gh	89.50fg	82.83D	76.33g	84.33fg	90.83ef	83.83D	4181h	4643gh	4967fg	4597D	4237g	4681fg	5041ef	4653D
Urea	97.67ef	105.7de	116.3cd	106.6C	98.17de	107.0cd	121.5b	108.9C	5421ef	5865de	6457cd	5914C	5448de	5937cd	6745b	6043A
AN	125.0c	139.0ab	150.0a	138.0A	119.3bc	143.9a	148.8a	137.4A	6938c	7715ab	8325a	7659A	6623bc	79.88a	8262a	7624A
AMS	108.0de	122.0c	126.3bc	118.8B	113.3bc	120.9b	124.7b	119.6B	5994de	6771c	7012bc	6592B	6288bc	6712b	6921a	6640B
Mean	101.5B	116.6AB	120.5A		101.8B	114.0A	121.5A		5633B	6248AB	6690A		5649B	6329A	6742A	

SA = Salicylic acid, NS = Nitrogen sources, AN = Ammonium nitrate, AMS = Ammonium sulfate

Table (3). Effect of salicylic acid, nitrogen sources and their interaction treatments on dry leaves/plant (g) and dry leaves/ha (kg) of *Artemisia annua* L. plant during 2021 and 2022 seasons.

SA / NS	Dry leaves/plant (g)								Dry leaves/ha (kg)							
	1st season				2nd season				1st season				2nd season			
	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean
1st cut																
Control	14.61g	17.72fg	18.12efg	16.82C	15.33f	16.95ef	17.50ef	16.59C	811g	984fg	1006efg	933.4C	851f	941ef	971ef	921C
Urea	16.88fg	20.60def	21.68cde	19.72B	15.90f	20.17de	22.50cd	19.52B	937fg	1143def	1203cde	1064B	883f	1119de	1249cd	1083B
AN	25.10bc	29.22a	30.90a	28.74A	22.00d	27.33ab	31.33a	26.89A	1393bc	1622a	1770a	1595A	1221d	1517b	1739a	1492A
AMS	23.47cd	28.21ab	29.10a	26.93A	24.00bcd	27.00b	26.33bc	25.78A	1303cd	1565ab	1615a	1459A	1332bcd	1499b	1462bc	1431A
Mean	20.02B	23.94A	25.20A		19.31B	22.86A	24.42A		1111B	1328A	1399A		1072B	1269A	1355A	
2nd cut																
Control	16.67g	18.83g	20.17g	18.56D	16.27f	19.07ef	23.33e	16.56C	925g	1045g	1119g	1030D	903f	1058ef	1295e	1085C
Urea	25.17f	28.17ef	32.33cde	28.56C	24.73de	30.73c	31.17c	28.88B	1397f	1563ef	1795cde	1585C	1373de	1706c	1730c	1603B
AN	35.10bcd	38.00b	43.33a	38.81A	34.07abc	37.63ab	39.07a	36.92A	1948bcd	2109b	2405a	2154A	1891abc	2089ab	2168a	2049A
AMS	31.67de	36.33bc	39.33ab	35.78B	30.10cd	33.22bc	37.77ab	33.69A	1758de	2017bc	2183ab	1986B	1671cd	1844bc	2096ab	1870A
Mean	27.15C	30.33B	33.79A		26.29B	30.16A	32.83A		1507C	1684B	1875A		1459B	1674A	1822A	

SA = Salicylic acid, NS = Nitrogen sources, AN = Ammonium nitrate, AMS = Ammonium sulfate

followed by the treatment of the combination between AMS and 2 mM SA. These findings are consistent with previous studies that examined the effect of SA and N on different plants by enhancing photosynthetic efficiency and nutrient uptake (Ghasemzadeh et al., 2012; Hayat et al., 2012; Pacheco et al., 2013; Sarrou et al., 2015; Basal et al., 2019 and Ilyas et al., 2021).

The application of SA and NS significantly enhanced the vegetative growth of *Artemisia annua* plants. SA, a key phytohormone, modulates several plant physiological processes, including cell division, expansion, and differentiation, which are crucial for plant growth (Vanacker et al., 2001 and Abreu and Munné-Bosch, 2008). The application of 2 mM SA resulted in substantial increases in plant height, number of branches, dry weight per plant and hectare and dry leaves per plant and hectare. SA acts as a signaling molecule that regulates plant growth and defense mechanisms, it improves photosynthetic efficiency, enhance nutrient absorption and adjusts antioxidant defense mechanisms, all of which improve plant growth and productivity (Scott et al., 2004 and Wang et al., 2010). In addition, SA is vital for stress tolerance because it increases the expression of genes that respond to stress and makes plants more resilient to abiotic stress like salt and drought (Ghasemzadeh et al., 2012 and Ahmad et al., 2020). Because of its many functions in encouraging plant physiology and metabolic efficiency, SA is a crucial part of agricultural methods meant to increase crop yield and quality. N plays a significant role in the production of the components of plants, through the action of several enzymes and protein synthesis (Jones et al., 1991) it was reflected in the growth metrics of the plants of sweet fennel, coriander and anise. N is a necessary component of several vitamins. N enhances the quantity and quality of dry matter in leafy plants (Silva and Uchida, 2000 and Khalid, 2012). N Fertilization enhanced the vegetative development of *Nigella sativa* L. (Khalid, 2001). Arabaci and Bayram (2004) found that, N fertilizer increased the amount of fresh herb yield, dry herb yield and dry leaves of *Ocimum basilicum* L. and increased the dry herb of *Mentha. piperita*, linalool chemo-type (Luciana et al., 2010).

3. Carbohydrates, N, P, K, Essential Oil Percentages, and Essential Oil Yield Per Plant and Hectare of *Artemisia annua*

SA significantly enhanced the chemical composition of *Artemisia annua* across both seasons and cuts. Specifically, SA at concentrations of 1 mM and 2 mM improved the total carbohydrates content, N, P, K and essential oil percentages, and essential oil yield per plant and hectare (Tables 4, 5, 6 and 7). Treatment with SA at 2 mM recorded the highest essential oil yield, reaching 37.16 kg and 39.63 kg in the first season, and 37.93 kg and 39.83 kg per hectare for the first and second cuts, respectively.

Table (4). Effect of salicylic acid, nitrogen sources and their interaction treatments on total carbohydrates % and N % of *Artemisia annua* L. plant during 2021 and 2022 seasons.

SA / NS	Total carbohydrates %								N %							
	1st season				2nd season				1st season				2nd season			
	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean
	1st cut															
Control	13.60h	14.60g	15.63f	14.61C	12.67f	15.10ef	15.63de	14.47C	1.90h	2.12gh	2.18g	2.07D	1.93h	2.13g	2.26g	2.11D
Urea	16.23ef	17.00e	18.07d	17.10B	16.43cde	18.00bcd	19.07ab	17.83B	2.98f	3.21de	3.28cde	3.16C	3.00f	3.24de	3.3de	3.18C
AN	18.60cd	19.37abc	20.00a	19.32A	19.70ab	20.93a	21.23a	20.62A	3.38bcd	3.57ab	3.66a	3.54A	3.37cd	3.60ab	3.68a	3.55A
AMS	19.10bc	19.40abc	19.80ab	19.43A	19.50ab	18.83abc	19.90ab	19.41A	3.15ef	3.28cde	3.45abc	3.29B	3.20e	3.30de	3.49	3.33B
Mean	16.88C	17.59B	18.38A		17.08B	18.22A	18.96A		2.85B	3.04A	3.14A		2.87B	3.07A	3.18A	
	2nd cut															
Control	13.97h	15.50g	16.60fg	15.36C	12.80g	14.03fg	15.77ef	14.20C	1.94g	2.19f	2.21f	2.11D	1.96h	2.22g	2.23g	2.14D
Urea	16.63fg	17.53ef	17.83de	17.33B	16.30de	16.93cde	17.67bcd	16.97B	2.94e	3.25d	3.31	3.17C	2.95f	3.27e	3.34de	3.19C
AN	17.93de	19.33bc	20.50a	19.26A	18.60abc	18.77ab	19.63a	19.00A	3.43bc	3.16a	3.62a	3.55A	3.45bc	3.64a	3.66a	3.58A
AMS	18.97cd	19.60abc	20.13ab	19.57A	18.83ab	19.20ab	20.00a	19.34A	3.24d	3.36cd	3.53ab	3.37B	3.26e	3.40cd	3.56ab	3.41B
Mean	16.88B	17.99AB	18.77A		16.63B	17.23AB	18.27A		2.89B	3.10A	3.17A		2.91B	3.13A	3.20A	

SA = Salicylic acid, NS = Nitrogen sources, AN = Ammonium nitrate, AMS = Ammonium sulfate

Table (5). Effect of salicylic acid, nitrogen sources and their interaction treatments on P and K of *Artemisia annua* L. plant during 2021 and 2022 seasons.

SA / NS	P %								K %							
	1st season				2nd season				1st season				2nd season			
	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean
1st cut																
Control	0.201k	0.220j	0.242i	0.224D	0.218l	0.234k	0.240j	0.231D	1.38g	1.46fg	1.50bc	1.45D	1.34g	1.46fg	1.52ef	1.44D
Urea	0.323h	0.328g	0.336f	0.329C	0.306i	0.328h	0.348f	0.327C	1.58de	1.68cd	1.70bc	1.65C	1.48ef	1.59cdef	1.66cd	1.58C
AN	0.359e	0.379b	0.407a	0.382A	0.342g	0.373d	0.398a	0.371B	1.79b	1.91a	1.97a	1.89B	1.54def	1.61cde	1.82ab	1.66B
AMS	0.372d	0.361e	0.374c	0.369B	0.376c	0.391b	0.371e	0.379A	1.97a	1.94a	1.98a	1.96A	1.66cd	1.70bc	1.87a	1.74A
Mean	0.316A	0.322A	0.340A		0.311C	0.332B	0.339A		1.68B	1.75A	1.79A		1.50B	1.59B	1.72A	
2nd cut																
Control	0.203l	0.217k	0.248j	0.223D	0.181e	0.230de	0.258cd	0.223C	1.40g	1.44fg	1.49f	1.44D	1.30h	1.39gh	1.49efg	1.39C
Urea	0.317i	0.328h	0.342g	0.329C	0.295bc	0.312bc	0.329ab	0.312B	1.55e	1.60e	1.67d	1.61C	1.43fg	1.57cde	1.62cd	1.54B
AN	0.352d	0.363c	0.396a	0.370A	0.340ab	0.348ab	0.375a	0.355A	1.77c	1.88ab	1.92ab	1.86B	1.52def	1.59vde	1.80ab	1.64A
AMS	0.345f	0.365b	0.349e	0.353B	0.339ab	0.342ab	0.341ab	0.340AB	1.87b	1.93a	1.89ab	1.90A	1.58cde	1.68bc	1.85a	1.70A
Mean	0.304C	0.318B	0.334A		0.229B	0.308AB	0.326A		1.65B	1.71A	1.74A		1.46B	1.56AB	1.69A	

SA = Salicylic acid, NS = Nitrogen sources, AN = Ammonium nitrate, AMS = Ammonium sulfate

Table (6). Effect of salicylic acid, nitrogen sources and their interaction treatments on oil % and oil yield per plant (g) of *Artemisia annua* L. plant during 2021 and 2022 seasons.

SA \ NS	P %								K %							
	1st season				2nd season				1st season				2nd season			
	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean	Control	1 mM SA	2 mM SA	Mean
1st cut																
Control	0.62d	0.82c	0.81c	0.75C	0.63g	0.85de	0.82ef	0.77D	0.44d	0.65c	0.67c	0.59C	0.45e	0.68d	0.68d	0.60D
Urea	0.68d	0.93b	0.94b	0.85B	0.71fg	0.97bc	0.96bcd	0.88C	0.58cd	0.83b	0.86b	0.76B	0.61d	0.87c	0.87c	0.78C
AN	0.94b	1.15a	1.13a	1.07A	0.96bcd	1.19a	1.16a	1.10A	0.93b	1.32a	1.38a	1.21A	0.95c	1.37a	1.43a	1.24A
AMS	0.86bc	1.08a	1.11a	1.02A	0.88cde	1.01b	1.14a	1.01B	0.85b	1.23a	1.32a	1.13A	0.87c	1.15b	1.36a	1.13B
Mean	0.77B	1.00A	1.00A		0.80B	1.00A	1.02A		0.70B	1.01A	1.06A		0.72B	1.02A	0.09A	
2nd cut																
Control	0.60d	0.83bc	0.80c	0.74C	0.62d	0.86bc	0.82c	0.77C	0.46g	0.70f	0.71f	0.62D	0.47f	0.71e	0.73e	0.64D
Urea	0.66d	0.91b	0.90bc	0.83B	0.67d	0.94b	0.92b	0.84B	0.65f	0.97de	1.05de	0.89C	0.65e	0.99cd	1.07cd	0.90C
AN	0.89bc	1.14a	1.07a	1.03A	0.91bc	1.17a	1.12a	1.07A	1.11cd	1.58a	1.61a	1.43A	1.14c	1.63a	1.68a	1.48A
AMS	0.85bc	1.03a	1.05a	0.98A	0.87bd	1.11a	1.09a	1.02A	0.92e	1.26bc	1.33b	1.17B	0.94d	1.36b	1.38b	1.23B
Mean	0.75B	0.98A	0.97A		0.77B	1.02A	0.99A		0.78B	1.13A	1.18A		0.80B	1.18A	1.22A	

SA = Salicylic acid, NS = Nitrogen sources, AN = Ammonium nitrate, AMS = Ammonium sulfate

Table (7). Effect of salicylic acid, nitrogen sources and their interaction treatments on oil yield/ha (kg) of *Artemisia annua* L. plant during 2021 and 2022 seasons.

SA / NS	Oil yield / ha (kg)							
	1st season				2nd season			
	Control	**1 mM SA**	**2 mM SA**	**Mean**	**Control**	**1 mM SA**	**2 mM SA**	**Mean**
				1st cut				
Control	24.24d	36.30c	37.16c	32.57C	24.93e	37.48d	37.93d	33.45D
Urea	32.19cd	46.30b	47.44b	41.98B	33.83d	48.25c	48.46c	43.51C
AN	51.60b	73.45a	76.61a	67.22A	52.71c	75.87a	79.20a	69.26A
AMS	47.16b	68.33a	73.16a	62.88A	48.50c	63.59b	75.62a	62.57B
Mean	38.80B	56.10A	58.59A		39.99B	56.30A	60.30A	
				2nd cut				
Control	25.21g	38.66f	39.63f	34.50D	26.02f	39.74e	40.79e	35.52D
Urea	35.93f	53.80de	58.45de	49.39C	36.08e	55.15cd	59.46cd	50.23C
AN	61.63cd	87.74a	89.15a	79.51A	63.06c	90.52a	93.30a	82.29A
AMS	50.97e	70.03bc	73.88b	64.96B	52.35d	75.64b	76.15b	68.05B
Mean	43.43B	62.56A	65.28A		44.37B	65.26A	67.42A	

SA = Salicylic acid, NS = nitrogen sources, AN = Ammonium nitrate, AMS = Ammonium sulfate

Regarding N treatments, AN demonstrated the highest improvements compared to the control treatment, showing significant increases in all measured parameters, followed by AMS, which also showed substantial improvements but was slightly less effective than AN. Both AN and AMS were the most effective NS, outperforming Urea and the control in all measured parameters across both seasons and cuts. Throughout both seasons and cuts, total carbohydrates content, N percentage, and oil yield per hectare significantly increased with AN, followed by AMS. AN recorded the highest essential oil yield, reaching 76.61 kg and 89.15 kg in the first season, and 79.20 kg and 93.30 kg per hectare for the first and second cuts, respectively.

SA and AN interaction resulted in the most significant improvements of all studied characters. Specifically, the combination of 2 mM SA and AN produced the best overall results in total carbohydrate content, NPK, essential oil percentages, and essential oil yield per hectare across both seasons and cuts. This treatment improved the essential oil yield/ha which reached 76.61 kg/ha and 89.15 kg/ha in the first season's cuts, and 79.20 kg and 93.30 kg in the second season's cuts, respectively, compared with control. Overall, the combination of 2 mM SA and AN consistently demonstrated the highest improvements in essential oil yield per hectare across both seasons and cuts, with the peak essential oil yield per hectare recorded at 93.30 kg/ha in the 2nd cut of the 2nd season. These findings underscore the significant impact of the 2 mM SA and AN combination in enhancing the oil yield of *Artemisia annua*. The results were supported by the findings of Khalid (2001), Arabaci and Bayram (2004), Sarrou et al. (2015), Sarmadi et al. (2018) and Pirbalouti et al. (2019) on *Ocimum basilicum* and *Mentha piperita* and El-Ghawwas et al. (2011) on *Artimisia annua*.

4. Artemisinin Content

Fig. (1) illustrates the chemical structure of artemisinin, a vital compound extracted from *Artemisia annua* L. and Fig. (2) demonstrates the effect of different treatments on artemisinin content in the plants during the 2nd cut of the 2nd season. The highest artemisinin content was achieved with the combination of 2 mM SA and AMS, significantly surpassing other treatments. These results are consistent with the findings of Aidah et al. (2023) on *Artemisia annua*.

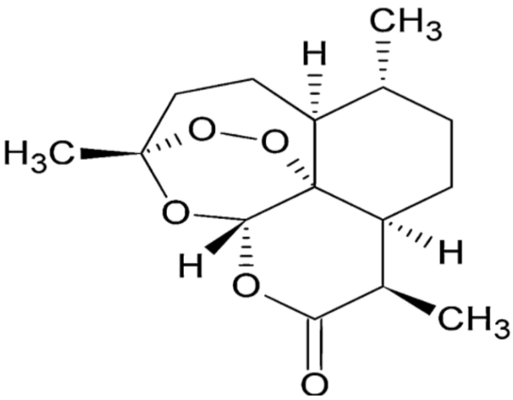

Fig. (1). The structure of artemisinin.

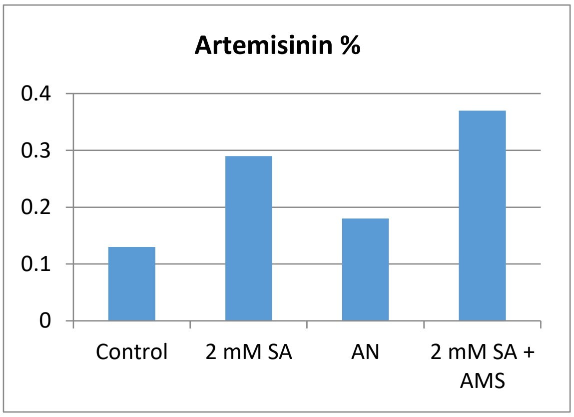

Fig. (2). Effect of SA, NS and their interaction treatments on artemisinin percentage of *Artemisia annua* L. plant at 2nd cut for 2nd season.

Chemical composition analysis, including carbohydrate, N, P, K content, and essential oil percentage and yield, showed notable improvements as a result of the AN and SA combination treatments. It is well known that SA up-regulates important genes involved in the manufacture of bioactive substances, boosting the output of essential oils and nutritional content (Ghasemzadeh et al., 2012 and Okuma et al., 2014). The results from Tables (4, 5, 6, and 7) showed that the greatest levels of nutrients were reported with the combined treatment of AN and 2 mM SA. This is in line with results from prior studies that showed SA increased nutritional absorption and metabolic activity, resulting in larger levels of carbohydrates, N, P, K, and essential oil contents (Gonçalves and Romano, 2013 and Okuma et al., 2014). SA treatments significantly increased essential oil content by enhancing the activity of biosynthetic pathways and promoting the accumulation of secondary metabolites (Sarrou et al., 2015; Sarmadi et al., 2018 and Pirbalouti et al., 2019). The enhancement of essential oil yield can be attributed to SA's role in up regulating genes involved in the biosynthesis of terpenoids, which are key components of essential oils (El-Ghawwas et al., 2011 and Rodrigues-Brandão et al., 2014).

N is an essential nutrient needed for the production of proteins, nucleic acids, amino acids, and chlorophyll. Its availability has a direct impact on the development, growth, and productivity of plants. Nitrogen is easily obtainable from AN, which is essential for biomass increase and vegetative development (Hao et al., 2012). Higher yields and better growth are the results of SA and AN working together to boost N absorption and assimilation. Research has demonstrated that the combination of N fertilizer with SA may greatly improve the efficiency of nutrient use and foster improved growth and productivity (Hayat et al., 2012; Sarrou et al., 2015 and Ilyas et al., 2021). Through the action of several enzymes and protein synthesis, N plays a significant role in the production of the components of plants (Jones et al., 1991) it was reflected in the chemical composition of the plants of sweet fennel, coriander, and anise. N is a necessary component of several vitamins. N enhances the quantity and quality of protein in grain crops in leafy plants (Silva and Uchida, 2000 and Khalid, 2012).

CONCLUSION

The best treatment for increasing the dry yield, leaf yield, and essential oil productivity per hectare in *Artemisia annua* plants was a combination treatment with AN and 2 mM SA. This combination produced considerable improvements in dry leaf yield and essential oil production, and it consistently produced the greatest dry weight across both cuts in both seasons. *Artemisia annua* crops may be grown with maximum productivity and quality due to the synergistic impact of AN and 2 mM SA, as demonstrated in this study. These results highlight the significant advantages of combining these two factors to increase biomass and essential oil yields for this plant's economic viability.

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استجابة الشيح الحولي لصور التسميد النتروجينى والرش بحمض السلسيليك

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الهدف من هذه الدراسة هو تعظيم نمو وإنتاج وجودة نبات الشيح الحولي (*Artemisia annua* L.)، والذي يحظى بإهتمام كبير نظرًا لإحتوائه على مركبات نشطة بيولوجيًا، وخاصة مادة الأرتيميسينين، المعروفة بخصائصها القوية المضادة للملاريا. أجريت تجارب حقلية بالمحطة التجريبية لمركز بحوث الصحراء برأس سدر بمحافظة جنوب سيناء بمصر خلال فصلي الصيف لعام ٢٠٢١ و٢٠٢٢ لدراسة تأثير مصادر النيتروجين (اليوريا ونترات الأمونيوم وكبريتات الأمونيوم) وحمض السلسيليك (٠، ١، ٢ مللى مول) على النمو والإنتاجية والمحتويات الكيميائية لنبات الشيح الحولي. بناءً على النتائج، أدى الجمع بين نترات الأمونيوم وحمض السلسيليك إلى أفضل خصائص نمو للنبات وتركيبه الكيميائي والمحصول. وقد أدت هذه المعاملة إلى تحسين كل من إرتفاع النبات وعدد الأفرع والوزن الجاف ومحتوى الزيت العطري بشكل ملحوظ. وبلغ محصول العشب الجاف في الموسم الأول ٦٨٢٢ كجم/هكتار في الحشة الأولى و٨٣٢٥ كجم/هكتار في الحشة الثانية، وفي الموسم الثاني كان محصول العشب الجاف ٦٣٨٣ كجم/هكتار في الحشة الأولى و٨٢٦٢ كجم/هكتار في الحشة الثانية. كما تحسن محصول الزيت العطري/هكتار بشكل ملحوظ، حيث سجلت أعلى محصول في الموسم الأول ٧٦,٦١ كجم/هكتار في الحشة الأولى و٨٩,١٥ كجم/هكتار في الحشة الثانية، وفي الموسم الثاني كان محصول الزيت العطري ٧٩,٢٠ كجم/هكتار في الحشة الأولى و٩٣,٣٠ كجم/هكتار في الحشة الثانية. وتشير هذه النتائج إلى أن التطبيق المشترك لنترات الأمونيوم وحمض السلسيليك بتركيز ٢ مللي مول هو المعاملة الأكثر فعالية لتعزيز نمو وإنتاجية نبات الشيح الحولي تحت ظروف جنوب سيناء، مصر.