Caspian Journal of Environmental Sciences
Online ISSN: 1735-3866
Print ISSN: 1735-3033

Application of *Eruca sativa* Mill. for organic fertilization and spraying with arginine, their effects on growth characteristics and seed yield along with identifying their content of some medically effective compounds

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ABSTRACT

A field experiment was conducted during the 2018-2019 season in one of the private fields in Karma City, Anbar Governorate, Iraq to study the effects of organic fertilizer levels (0, 5 and 10 tons ha^{-1}) and spraying with amino acid arginine at concentrations of 0, 100, 200 mg L^{-1} on growth and also yield characteristics in addition to some medicinally-active compounds in watercress seeds, *Eruca sativa* Mill. The study factors were arranged as split-plot experiment according to the randomized complete block design (RCBD), with three replications. According to the results, the fertilizer level at 10 tons ha^{-1} exhibited a significant elevation in the characteristics of growth, yield, oil rate (%), and seed contents of glucosinolate glycoside and Astragalin alkaloid, whereas spraying arginine at 200 mg L^{-1} showed significant differences in the characteristics of yield, oil rate (%), and seed content of glucocinolate glycoside and Astragalin alkaloid. In addition, the interaction of the studied characteristics displayed a significant effect. So that, the plants given 10 tons ha^{-1} of organic fertilizer under the influence of spraying with 200 mg L^{-1} arginine, revealed the highest rate of 1000-seed weight, oil rate (%), in addition to the seed content of glucocinolate glycoside and Astragalin.

Keywords: Eruca sativa, Fertilizer, Plant, Growth, Medicinal plant.
Article type: Research Article.

INTRODUCTION

Watercress plant, *Eruca sativa* Mill., one of the family Brassicaceae, is known as fresh green leaves. Its seeds contain oil that is promising to be medicinal oil through its effects on improving liver function, elevating sperm fertility, enhancing the effectiveness of sex hormones such as progesterone and estrogen as well as the sex glands (Merza *et al.* 2000). The negative effects of chemical fertilizers on environmental pollution come through the risk of toxic accumulation of chemicals in the soil (Rai *et al.* 2015). So, agricultural specialists have been urged to rely on organic fertilizers in addition to high economic costs. Consequently, global interest in finding alternatives was launched from the application of chemical fertilizers, hence, organic agriculture spread to many field crops and vegetables, since it acquired wide acceptance in most countries of the world (Rakhimova *et al.* 2021; Valentinovna 2021; Kamali Omidi *et al.* 2022; Al-Dulaimy *et al.* 2022). So that, an approved agricultural system was higher than 31 million hectares distributed in almost 120 countries (Willer & Yusessefi 2006). One of the most important organic sources that have been used in the past and present is animal fertilizer, which is rich in source of nutrients. In addition, it is considered as one of the fertilizers that preserve nutrients from deterioration and loss, help the soil to retain water, increase soil revival activity, and improve its physical and chemical properties (Fageria 2012). Havlin *et al.* (2005) stated that using cow manure as an organic fertilizer increases soil acidity, leading to an elevation in the availability of macronutrients in the soil such as N, P, K, Mg, Ca and improving its properties. Mheidi *et al.* (2018) reported the highest significant rise in plant height and number of branches of the anise plant

Caspian Journal of Environmental Sciences, Vol. 21 No. 1 pp. 161-167 Received: April 02, 2022 Revised: Aug. 27, 2022 Accepted: Sep. 19, 2022
DOI: 10.22124/CJES.2023.6209

Publisher: University of Guilan, Iran

Pimpinella anisum L. when adding organic fertilizer at the level of 5 tons ha^{-1}, reaching 84.77 cm in height and 14.53 branches compared to those with no additional organic fertilizer. The lowest was reported as 73.92 cm and 11.74 branches for each of the traits respectively. Amino acids are the basic compounds for building protein in the plant, as they are composed of inorganic compounds consisting of the fission processes and the stress cycle, and also in the presence of inorganic nitrogen (NH_3; Verma & Vetma 2008). The amino acid compounds help and stimulate the plant to efficiently absorb nutrients from fertilizers added to the soil. It is very important for its function as a natural chelating substance that plays an important role in plant's resistance to the encountered abnormal conditions, including high or low temperatures and poor nutrition, like arginine deficiency, in addition to its role in helping the plant to grow in a balanced way and elevating its resistance to diseases (Abdel-Hafez 2006), followed by the role of arginine in increasing cell division and chlorophyll as well as upraising the activity of enzymes, which is positively reflected in the characteristics of yield and the quality of the seeds (Hozayn *et al.* 2010). In contrast, a study on the barley to determine the effect of arginine, the higher concentration of 400 mg L^{-1} exhibited a significant increase in plant height and dry weight (Shalaby *et al.* 2018). Due to the importance of this crop in many medicinal and nutritional uses, this study was conducted, in order to determine the best amount of organic fertilizer and appropriate concentration of arginine for plant growth and those which may be reflected in the amount and quality of seed yield and oil.

MATERIALS AND METHODS

The experiment was carried out during the 2018-2019 agricultural season in one of the private fields in Al-Karma City, Anbar Governorate, Iraq in soil with physical and chemical characteristics shown in Table 1 to study watercress plant's response to organic fertilizer and spraying with arginine and their effects on some growth characteristics, yield and content of some of the effective compounds. The experiment land was prepared from plowing, levelling and modification, then it was divided into plots with 2.5 × 2 m in dimension. The seeds were planted on 10/15/2018 in the lines; the distance between one line and another was 30 cm while between one plant and another 10 cm. All agricultural processes were carried out including irrigation and hoeing when needed. The experiment was conducted with a split plot system according to a Randomized Complete Block Design (RCBD) with three replications. Where the concentrations of the first factor in the main plot were three concentrations of arginine (0, 100 and 200 mg L^{-1}). Each level was sprayed on the leaves of the plant until completely wet in the early morning time. Ten-litre hand-held two sprays were carried out during the growing season as follows:

• The first spray: in the vegetative growth stage.
• The second spray: when the plants reach 100% flowering
• As for the comparison treatment, it was only sprayed with water.

The second factor levels in the sub plots included three levels of organic fertilizer (decomposed cow manure; Table 2) with three levels (0, 5 and 10 tons ha^{-1}). Before planting, the surface layer mixed with the soil well. All data were subjected to statistical analysis of variance (ANOVA), and the means of the factors were compared based on the Least Significant Difference (LSD) test at a significant level of 5% (Al-Rawi & Khalaf Allah 2000).

Data collection

1- Plant height (cm).
2- The number of leaves of a plant.
3- The number of plant flowers.
4- Weighing 1000 seeds.
5- The oil content (%) was measured according to the method of Stahl (1969).
6- Glucosinolate seed content (mg g^{-1} dry weight) was measured according to the method of Jezek et al. (1999)
7- Seed content of the alkaloid Astragalin was assayed according to the method of Stahl (1969).

Table 1. Some physical and chemical characteristics of experiment soil.

Parameter	Value
EC (dS m^{-1})	2.81
pH	7.21
Organic matter (%)	1.25
Available N (mg kg^{-1})	110.5
Available P (mg kg^{-1})	34.3
Available K (mg kg^{-1})	124.9

Ca^{++} (mg kg^{-1})	3.02
Mg^{++} (mg kg^{-1})	1.9
SO_4^{--} (mg kg^{-1})	3.53
Na^{+} (mg kg^{-1})	0.8
Clay (g kg^{-1})	304
Silt	552
Sand	140
Texture	Silty-Clay

Table 2. Some chemical analysis of organic manure used.

Parameter	Value
EC (dS m^{-1})	12.22
pH	6.35
Organic Carbon (mg kg^{-1})	302
Available N (mg kg^{-1})	22.5
Available P (mg kg^{-1})	10.4
Available K (mg kg^{-1})	23.6
C/N	10.5
Ca^{++} (%)	2.95
Fe	0.52
Zn	0.07

RESULTS AND DISCUSSION

Parameters of growth

It is evident from Table 3 that adding organic fertilizer exhibited a significant effect on increasing the height of the plant and the number of leaves in it. The effect increases once the level of added fertilizer elevates, so that, the fertilized plants at level of 10 tons ha^{-1} recorded the highest rate for these two characteristics, reaching 56.00 cm and 34.58. The plant experienced a significant elevation of 27.50% and 8.65% over the comparison treatment (without fertilization), which recorded the lowest rate of 43.92 cm and 31.78 leaves for both characteristics respectively. This increase may be attributed to the role of organic fertilizers (cow manure) in elevating the nutrient concentrations in the soil, especially nitrogen, which plays a direct role in enhancing the formation of leaf principles, and is positively reflected in building nucleic acids and the production of necessary and stimulating proteins to increase cell division, in addition to the role of nitrogen in the production of auxin, which increases cell division and elevates the plant height and number of leaves. These results are consistent with the findings of Darzi & Hadi (2012) on dill, and EL-Dewiny *et al.* (2006) on spinach, as they indicated that organic fertilizers have a significant effect in enhancing the growth and plant height characteristics. Arginine and the interaction between the study factors, were similar to the behaviour of organic fertilizer, as the rate of plant height and the number of leaves in it increased by the elevated concentrations of spraying, however, it did not reach the significant limits (Table 3).

Yield components

According to Table 4, the increased level of added organic fertilizer exhibited a significant effect on the elevated number of pods per plant and the weight of 1000 seeds. The effect rises as the level of added fertilizer increases, specially if the plants were given a high level of organic fertilizer (10 tons ha^{-1}). The highest rates of these two characteristics were 84.67 pods and 0.6711 g per 1000 seeds respectively, compared to the comparison treatment that recorded the lowest rate (80.74 pods and 0.5822 g per 1000 seeds respectively). These effects may be attributed to the fact that adding organic fertilizer improves the physical and chemical characteristics of the soil and the release of nutrients in the vicinity of the roots of plants, which is accompanied by an elevation in the biological processes inside the plant and the transfer of their products to the components of the yield. These processes are considered as the final downstream for the collection of these products, which is positively reflected in the elevated number of pods in the plant and the upraised weight of its seeds. This is consistent with those found by EL-Desuki *et al.* (2010) and the role of humic acid in improving vegetative growth and reducing food competition between pods, thus the increased number of pods in the plant along with the colour of its seeds. The spraying of plants with different concentrations of arginine has significant differences between plants in the number of pods and the weight of 1000 seeds.

Table 3. Effects of organic fertilization and spraying with arginine, and the interaction between them on plant height and number of leaves in the plant.

Factors	Concentrations Arginine (mg L^{-1})	Plant height (cm)	The number of plant leaves
Organic fertilization levels (tons ha^{-1})	0	43.92	31.78
	5	48.81	33.74
	10	56.00	34.53
	LSD = 0.05	1.88	0.914
Arginine concentrations (mg L^{-1})	0	48.66	33.14
	100	49.95	33.54
	200	50.12	33.37
	LSD = 0.05	NS	NS
Interactions			
0	0	43.42	31.51
	100	44.04	32.40
	200	44.31	31.42
5	0	48.38	33.47
	100	49.33	33.65
	200	48.73	34.09
10	0	54.18	34.45
	100	56.49	34.56
	200	57.31	34.58
LSD = 0.05		NS	NS

Note: LSD = Least Significant Difference; NS = Not significant.

So that, the sprayed plants with a high concentration of arginine (200 mg L^{-1}) exhibited the highest rate of these two characteristics, reaching 84.12 pods per plant and 0.6556 g per 1000 seeds, in comparison with the control (spraying plants with water), which recorded the lowest rate of 81.67 pods and 0.5978 g per 1000 seeds (Table 4). The elevation in the most of characteristics as a result of spraying with arginine is due to the fact that this acid plays an important role in many vital processes, whether in its free form or as a component of proteins. So, it is important and effective in all stages of plant growth, including encouraging vital activities, especially the two processes of division and the expansion of plant cells. In addition, it increases the activity of enzymes that work on the decomposition of organic compounds and work to liberate elements from them, upraises their readiness and in turn elevates the plant growth rates (Claussen 2004; Nur *et al.* 2006.) Moreover, the free amino acids are an essential nitrogen sources in building proteins and enzymes and also energy processing which encourage vegetative growth and increase yield components (Abdel- Aziz & Balbaa 2007).

The interaction between organic fertilizer and spraying with arginine did not display a significant effect on the characteristic of the number of pods, while the effect was significant in the weight of 1000 seeds. So that, the plants with a high level of organic fertilizer and sprinkled with a high concentration of arginine (5 tons ha^{-1} + 200 mg L^{-1} exhibited the highest seed weight of 0.7200 g, compared to non-fertilized plants and sprayed with water, which recorded the lowest average of 0.5367 g (Table 4).

Active compounds in seeds

It is evident from the results in Table 5 that the level of added organic fertilizer exhibited a significant effect on increasing the oil content (%) and the amount of glycosides and alkaloids in the seeds. So that, the seeds of plants fertilized with a high level of organic fertilizer (10 tons ha^{-1}) recorded the highest rates of oil content and the amount of glucocinolate and the alkaloid Astragalin (36.06%, 26.28 mg g^{-1}, 32.34 mg g^{-1} respectively), significantly superior to the rest of the levels. The comparison coefficients (without adding organic fertilizer) recorded the lowest rates for the abovementioned compounds (26.79%, 22.65 mg g^{-1}, 29.84 mg g^{-1} respectively). This may explain the role of organic fertilizer in releasing nutrients such as nitrogen, phosphorous and potassium, which play a direct role in elevating the activity of the photosynthesis process.

Table 4. Effects of organic fertilization and spraying with arginine and the interaction between them on the number of pods per plant and the weight of 1000 seeds.

Factors	**Arginine concentrations (mg L^{-1})**	**The number of pods per plant**	**Weight of 1000 Seeds**
Organic fertilization levels (tons ha^{-1})	0	80.74	0.5822
	5	83.08	0.6433
	10	84.67	0.6711
LSD = 0.05		0.83	0.021
Arginine concentrations (mg L^{-1})(mg L^{-1})	0	81.67	0.5978
	100	82.69	0.6433
	200	84.12	0.6556
	LSD = 0.05	1.39	0.023
Interactions			
0	0	78.93	0.5367
	100	80.62	0.6333
	200	82.66	0.5767
5	0	82.27	0.6100
	100	82.72	0.6500
	200	84.25	0.6700
10	0	83.82	0.6467
	100	84.73	0.6467
	200	85.46	0.7200
LSD = 0.05		NS	0.034

Note: LSD = Least Significant Difference; NS = Not significant.

It in turn enhances the pace of building amino acids, nitrogenous bases and some vitamins that contribute directly or indirectly to increase the effectiveness of building carbohydrates, especially disaccharides, especially sucrose, which are converted into oil in the lysosomes of the seed cells.

The upraised production of basic compounds such as carbohydrates and proteins leads to an elevation in the formation of secondary compounds as a concomitant result. It is also noted that spraying with arginine exhibits a significant effect in elevating these characteristics, such as arginine rise in the plant. This may be justified by the active role of arginine in the transfer of carbohydrates from the leaves to the seeds of the sink, as well as its role in elevating the activity of biological processes that support the upraised carbohydrate production, especially the glucose, which participates in uridine diphosphate glucose (UDPG) in the biosynthesis of the glucocinolate molecule.

It is also noticed that there is a significant difference in the interaction between organic fertilizer and spraying with arginine, as the plants that were fertilized at a level of 10 tons ha^{-1} and sprayed with arginine at a concentration of 200 mg L^{-1}, exhibited the highest rates (37.06%, 27.16 mg g^{-1} and 33.84 mg g^{-1}) of oil content, glycoside and also alkaloid contents respectively, compared to the lowest rates (26.20%, 21.31 mg g^{-1}, 25.44 mg g^{-1}) of relevant traits in the control groups that were not fertilized with organic fertilizer and sprayed with distilled water only (Table 5).

Table 5. Effects of organic fertilization and spraying with arginine and the interaction between them on the oil content (%), glycosides and alkaloids.

Factors	Concentrations Arginine (mg L^{-1})	The percentage of oil in the seeds (%)	Glucosinolate glycoside (mg g^{-1})	Astragalin alkaloid (mg g^{-1})
Organic fertilization levels	0	26.79	22.65	29.84
(tons ha^{-1})	5	31.57	24.68	31.35
	10	36.06	26.28	32.34
	LSD = 0.05	0.45	0.31	0.57
Arginine concentrations	0	30.03	23.64	27.92
(mg L^{-1})	100	31.61	24.77	32.18
	200	32.79	25.20	33.43
	LSD = 0.05	0.24	0.84	1.18
Interactions				
	0	26.20	21.31	25.44
0	100	26.65	23.18	31.43
	200	27.52	23.46	32.65
	0	29.18	24.39	27.73
5	100	31.74	24.67	32.50
	200	33.80	24.97	33.80
	0	34.70	25.23	30.58
10	100	36.43	26.46	32.59
	200	37.06	27.16	33.84
LSD = 0.05		0.65	0.84	1.25

Note: LSD = Least Significant Difference; NS = Not significant.

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Bibliographic information of this paper for citing:

Mheidi, UH, Abdulkafoor, AH & Ali, IM 2023. Application of *Eruca sativa* Mill. for organic fertilization and spraying with arginine, their effects on growth characteristics and seed yield along with identifying their content of some medically effective compounds. Caspian Journal of Environmental Sciences, 21: 161-167.