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Hend M Swaefy
Faculty of Agriculture, Department of Ornamental Horticulture, Cairo University, Giza, Egypt

Shimaa A Shaaban
Faculty of Agriculture, Department of Agricultural Botany, Cairo University, Giza, Egypt

Corresponding Author:
Hend M Swaefy
Faculty of Agriculture, Department of Ornamental Horticulture, Cairo University, Giza, Egypt

Improvement growth and chemical components of Cardoon plant through foliar application of nutrients and vitamins

Hend M Swaefy and Shimaa A Shaaban

Abstract
The study was carried out at the Department of Ornamental Horticulture, Faculty of Agriculture, Cairo University during the two successive seasons of 2017/2018 and 2018/2019 to investigate the effect of ascorbic acid at 50 and 100 ppm, folic acid at 10 and 25 ppm, and nutrients mixture at the rate of 10 ml/3L on the growth and chemical composition of *Synara cardunculus* var. *altilis* plant. The results showed that ascorbic acid had more enhancement effect on the leaf parameters compared with folic acid. Ascorbic acid at 50 ppm had a superior effect on the average leaf fresh weight (g), herb fresh weight (g), number of heads per plant, number of seeds per head, seed yield per plant (g), seed oil percentage and total phenols content compared to the other treatments. Supplying plants with the combination of nutrients mixture and vitamins (ascorbic or folic acid) had better improvement on total flavonoids than the vitamin alone. The identified components in fixed oil were Linoleic, Myristelaidic and Oleic as unsaturated fatty acids and Caprylic, Undecylic, Lauric, Tridecylic, Myristic, Palmitic and Stearic as saturated fatty acids. The highest total unsaturated fatty acids were found with the treatment of 10 ppm folic acid compared to the other treatments. The highest content of linoleic was found with the treatment of 25 ppm folic acid followed by the treatment of 10 ppm folic acid. Regarding the effect of foliar application with the best treatment (50 ppm ascorbic acid) on the anatomical structure, it could be stated that this treatment caused a marked increase in measurements and counts of the leaf internal tissues.

Keywords: *Cynara cardunculus* var. *altilis*, ascorbic acid, folic acid, flavonoids, seed oil

1. Introduction

The plant (*Cynara cardunculus* L.), commonly named "cardoon" is a perennial herbaceous plant belongs to family Asteraceae and includes three varieties namely; *sylvestris* (Lamk) fiori (wild cardoon), *scolymus* (L.) fiori (artichoke), and *altilis* (cultivated cardoon) [1]. *Cynara cardunculus* var. *sylvestris* (Lamk) is considered the grandfather of both the globe artichoke and the cultivated cardoon. Globe artichoke is grown for large edible non-spiny heads. Cultivated cardoon (*Cynara cardunculus*, syn. *C. cardunculus* var. *altilis* DC) is grown for its large edible non-spiny leaf stalks. Leafy Cardoon (cultivated cardoon) is of Mediterranean origin where it is used as a vegetable crop. Nowadays, countries such as France, Spain, and Italy still use leaf stalk of cardoons for culinary. Cardoons also are common vegetables in northern Africa, often used in Algeria and Tunisia [2]. Leafy Cardoon has been offered as an alternative source for producing cellulose biomass and oil [3].

Leafy Cardoon is a great addition to the ornamental garden for its decorative large silvery foliage. The plant height is about 1-1.5 m and has purple inflorescence. Moreover, cardoon has medicinal applications due to the polyphenol content. The different parts of Cardoon have many phenolic components and flavonoids, so this plant is a source of antioxidant phenolic compounds such as cynarin, silymarin, luteolin, and apigenin. The primary compounds in the cardoon leaf are flavones and the caffeoylquinic acid in the floral stem. Also, *C. cardunculus* extract has effect as antimicrobial and anticancer potential against breast cancer [1]. Seeds of cardoon plant are rich in oil which used for food and industry moreover both heads and seeds of cardoon are good source of fatty acids [4].

Natural phenols can be divided into two categories, flavonoids and non-flavonoids. Flavonoid compounds play an important protective role against outrageous ultraviolet light [5]. Flavonoids are considered principal phytochemical compounds in cardoon leaf including silymarin, luteolin, and apigenin. Silymarin is essential to manage hepatic disorders including non-alcoholic fatty liver disease. Luteolin possesses anti-cancer and anti-oxidant properties [6].

Also, it has anti-inflammatory effects in brain injury, spinal cord injury [7]. Apigenin is considered as a natural bioactive compound shown to have anti-cancer properties, particularly cancers of the breast, digestive tract, skin, prostate, and certain hematological malignancies [8].

The seeds of cardoons showed the highest oil content (25.2%) among the *Cynara spp*. The cardoon oil profile was 10.7% palmitic, 3.7% stearic, 25.0% oleic and 59.7% linoleic acids. The seeds of the cardoon can be involved as a sustainable energy source in the production of biofuels. The stalks could be used as a fiber source (17% lignin) for paper pulps production with high strength properties [9].

The nutrients mixture components contain macro elements as N, P, K and micro elements as Fe, Mn, Mo, and B. Nitrogen is considerable key component of amino acids, which form the building block of protein and enzymes. N is a vital component of the chlorophyll molecule, which helps plants to absorb energy from the light. Application of N in recommended doses increases yield through the increasing of plant growth and photosynthesis pigments. Phosphorus nutrition enhances fresh yield of leaves, seeds and flavonoids content in artichoke plant due to its important in metabolic process and producing strong roots [10].

Ascorbic acid (AsA) or vitamin C is an antioxidant and plays a crucial role in the growth and development of plants. It has a role in controlling cell expansion and cell division. Also (AsA) prevents plants from reactive oxygen species (ROS) damage and considers a cofactor of many enzymes and electron donor in the electron transport chain [11, 12].

Folic acid (vitamin B) is one of the most important vitamins due to its role in amino acids metabolism and nucleic acids synthesis. Folic acid in the plant is active when it occurs in reduced form, [13]. Folic acid has an effect in increasing yield by increasing vegetative growth and chlorophylls content in potato plant. Moreover this increase due to its role in many cellular reaction as metabolism of amino acids, formation of lignin, chlorophyll, as well as synthesis of methionine [14].

The objective of the present study is to evaluate the effect of nutrients mixture, ascorbic acid and folic acid on growth, yield and flavonoid compounds content in cardoon plants.

2. Materials and Methods

2.1. Field Experiment: This study was conducted at the experimental nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, during the two successive seasons of 2017/2018 and 2018/2019 to study the effect of two organic acids and nutrients on the growth and chemical composition of *Synara cardunculus* var. *altilis* plant. The seeds were sown on 1st of November in the two seasons in nursery beds. Seeds took ten days to germinate and thinning was done three weeks later. Seeds were cultivated in (1.5 x 1.5 m) plots, in rows 50 cm apart at 50 cm between plants (four plants/ m^2), each treatment consisted of 3 replicates and each replicate contained 9 plants.

Ascorbic and folic acids were added as foliar application three times per season, the first one started 45 days after planting then 30 days interval. Ascorbic acid was applied at the rates of 0, 50 and 100 ppm. Folic acid was applied at the rates of 0, 10 and 25 ppm. Commercial nutrients mixture named (Nufatrien) was added two times per season at the rate of 10 ml/3L as foliar application, the first one started 45 days after planting and the second was done two months later. The nutrients mixture components were 5% N, 5% P, 5% K, 1.5% Fe, 1.5% Zn, 1% Mn, 0.1% Mo and 0.5% B. Phosphorus as calcium super-phosphate (15.5% P_2O_5) was added as a basal dose for all plants during the preparation of the soil at the rate of 15 g/ plant/season.

Data on leaf length (cm), number of leaves/plant, average leaf fresh weight (g) and herb fresh weight (g) were recorded 5 months after sowing (on 1st of April in the two seasons). Two months later flower stem height (cm) and number of heads/plant were recorded. Harvesting was done after maturity and data about the mature heads were measured including seeds weight /head (g), head dry weight (g), number of seeds/ head, seed yield/plant (g), and oil%.

The layout of the experiment was randomized complete design with one factor, with three replications of ten treatments in each season. The treatments were as the following: Control, Nufatrien at the rate of 10 ml/3L, Ascorbic acid at the rate of 50 ppm, Nufatrien + Ascorbic acid at the rate of 50 ppm, Ascorbic acid at the rate of 100 ppm, Nufatrien + Ascorbic acid at the rate of 100 ppm, Folic acid at the rate of 10 ppm, Nufatrien + Folic acid at the rate of 10 ppm, Folic acid at the rate of 25 ppm and Nufatrien + Folic acid at the rate of 25 ppm.

2.2. Anatomical study

The plant material prepared for the anatomical study was taken after 5 months of planting, and micro technique procedure was done according to [15]. Transverse sections which were cut on a rotary microtome to a thickness of 15µ were stained with crystal violet-erythrosine combination before mounting in Canada balsam. Sections were examined and the data were recorded. Photomicrographs of the selected treatments were taken.

2.3. Soil and Chemical Analyses

The physical and chemical characteristics of the soil experiment field were determined according to [16]. Soil has the following mechanical characteristics: sand 56.4%, clay+ silt 43.6% and chemical characteristics are shown in Table (1).

Table 1: Chemical analysis of the experimental soil

Chemical Analysis													
pH	E.C. (dS/m)	OM%	Soluble Cations (meq. /L.)				Soluble Anions (meq. /L.)			Available Macronutrients (mg/kg)		
7.39	5.57	0.6	Na+	K+	Ca++	Mg++	HCO3--	Cl-	SO4--	N	P	K
			23.7	1.3	25	11	4	28	29	149.6	6.2	420

At 5 months old plants, total flavonoids (mg/g DW) in the both seasons were determined in dried leaves according to [17]. Total phenols (mg/g FW) were determined using the Folin-Ciocalteau colorimetric method according to [18].Total sugars (mg/g FW) were determined by phosphor-molybdic method according to [19].

Total free amino acids (mg/g FW) were determined using Ninhydrin reagent according to [20].

Cardoon oil from seeds was extracted at the Laboratory of Medicinal and Aromatic Plants, National Research Center-Giza using a Soxhlet apparatus and the oil percentage was calculated. Cardoon oil in the second season was analyzed using GC analysis, to determine the main constituents. The use of GC in the quantitative determinations was performed using the methods described by [21] using (Gas Liquid Chromatography Trace GC Ultra Thermo Scientific) apparatus stands at the Laboratory of Cairo University Research Park, CURP - Faculty of Agriculture.

2.4. Statistical analysis
For statistical analysis of the data, randomized complete design with one factor was used with three replications for each parameter. Duncan's Multiple Range Test [22] was applied to detect the significant differences among treatments means, when the F-test for these treatments was significant at 5% probability level using Assistat program.

3. Results and Discussion
3.1. Leaf parameters
Cardoon leaf parameters were studied due to its importance for industrial purposes as it is used in the manufacture of high-quality paper pulp. Also, blanched cardoon stalks are used to prepare many wonderful dishes in European restaurants. The data presented in (Table 2) illustrated the effect of nutrients mixture, ascorbic acid and folic acid treatments on average leaf fresh weight (g), number of leaves/plant and leaf length (cm) of *Cynara cardunculus* var. *altilis* plant.

Data showed that generally, all studied treatments increased leaf parameters compared to control plants. Plants treated with nufatrien lonely had heavier leaf fresh weight and had bigger number of leaves per plant than plants treated with folic acid at 10 or 25 ppm with or without nufatrien. On the other hand, leaf length of plants treated with 10 ppm folic acid with or without nufatrien was more than treated with nufatrien only.

Ascorbic acid had more enhancement effect on the leaf parameters compared with folic acid. Ascorbic acid at 50 ppm had a superior effect on the average leaf fresh weight compared to other treatments with values of 308.33 and 300.0 g in the first and second seasons, respectively. In both seasons, the biggest number of leaves per plant (16.0) was recorded with the treatment of ascorbic acid at the rate of 100 ppm. The longest leaf was recorded in plants treated with Nufatrien plus 100 ppm ascorbic acid with values of 158.0 and 149.67 cm in the first and second seasons, respectively.

The increasing in leaf parameters could be attributed to the role of ascorbic acid as coenzymes and controlling cell expansion and cell division. The results were in agreement with findings on Sunflower plants by [23] who found that there was a significant increase in the number of leaves per plant as a result of ascorbic acid at the rate of 1 and 2 mmol as a foliar application. Also, [24] reported that ascorbic acid showed improvement in plant growth because it can influence on ions uptake and transport, membrane permeability and photosynthesis.

Table 2: Effect of nutrients mixture, ascorbic acid and folic acid on cardoon leaf parameters

Treatments	Aver. leaf fresh weight (g)		Number of leaves/plant		Leaf length (cm)	
	First season	Second season	First season	Second season	First season	Second season
Control	111.67f ±20.2	120.00g ±13.2	11.67bcd ±2.5	10.00b ±2.6	110.00e ±15.0	112.67b ±29.1
Nufatrien	260.00bcd ±13.2	263.33bc ±20.8	15.00abc ±4.6	14.33ab ±5.5	136.67bcd ±14.4	128.33ab ±22.3
Asc50	308.33a ±20.2	300.00a ±13.2	15.67ab ±4.0	15.67ab ±5.0	148.33ab ±7.6	145.00a ±8.7
Nufatrien+Asc50	271.67abc ±54.8	290.00ab ±15.0	15.67ab ±4.7	15.67ab ±4.0	147.33ab ±10.3	144.00a ±11.5
Asc100	303.33ab ±22.5	295.00ab ±13.2	16.00a ±3.0	16.00a ±3.6	146.67abc±23.1	143.00a ±18.1
Nufatrien+Asc100	241.67cde ±27.5	238.33cd ±27.5	13.33abcd ±3.8	14.33ab ±3.2	158.00a ±3.5	149.67a ±22.4
Fol10	226.67de ±22.5	226.67de ±28.9	12.67abcd ±4.6	13.67ab ±3.5	141.67abcd ±12.6	143.33a ±24.6
Nufatrien+Fol10	225.00de ±5.0	221.67de ±17.6	12.33abcd ±6.7	13.67ab ±6.0	141.67abcd ±14.4	136.67ab ±11.5
Fol25	208.33e ±15.3	203.33ef ±20.2	11.00cd ±2.0	12.33ab ±1.2	130.00cd ±10.4	130.67ab ±8.1
Nufatrien+Fol25	203.33e ±20.8	180.00f ±13.2	10.33d ±1.5	11.67ab ±2.1	126.33de ±1.2	123.33ab ±5.8

Different letters indicate significant differences according to Duncan's multiple range test ($P = 0.05$)
Nufatrien= macro and micronutrients mixture at the rate of 10 ml/3L Asc50= ascorbic acid at the rate of 50 ppm, Asc100= ascorbic acid at the rate of 100 ppm Fol10= folic acid at the rate of 10 ppm Fol25= folic acid at the rate of 25 ppm

3.2. Leaf anatomical structure
Measurements (µ) and counts of certain anatomical features in transverse sections through the median portion of the leaf blade on the main stem of Cardoon (*Cynara cardunculus* var. *altilis*) plant treated with foliar application of 50 ppm ascorbic acid and those of untreated plant are given in Table (3) and Figure (1). Transverse section at the midvein shows Uniseriate epidermis, below the epidermal cells in every corner there is collenchyma tissue followed by parenchyma tissue. The midvein consists of eight elongated vascular bundles. Mesophyll tissue in *Cynara cardunculus* var. *altilis* leaf lamina consists of palisade and spongy tissues. According to the morphological results, the plants treated with 50 ppm ascorbic acid as a foliar application were the best.

This was consistent with the anatomical results of the leaf as it caused a higher increase in leaf lamina thickness (45.46%) relative to the control. The increase in leaf lamina thickness was attributed to an increase in the thickness of the upper and the lower epidermis, the palisade and spongy tissues being; 46.10, 66.67, 43.10 and 66.65%, respectively, compared to control. Also, foliar application of 50 ppm ascorbic acid achieved another increment in the leaf midvein thickness. The average dimensions of midvein recorded 6365.66 and 4535.75 µ in length and width, by 112.82 and 65.57% increase over the control, respectively. The increases that occurred in midvein due to the applied 50 ppm ascorbic acid were linked with another increase in collenchyma tissue thickness, large and small vascular bundle dimensions, xylem and phloem thickness in small and large bundles as well as their xylem vessels diameters and numbers.

Relative to control, the average increase percentages in large and small vascular bundle dimensions were 84.15 and 9.88% for a large bundle and 58.91 and 51.31% for a small bundle in length and width, respectively. While the percentage of increment in thickness of xylem and phloem tissues, xylem vessels diameters and No. of xylem vessels were 63.21, 94.41, 82.70 and 64.71% in a large vascular bundle and 42.81, 109.56, 69.37 and 155.56% in a small vascular bundle, respectively, compared with untreated plants. In this respect [25] on sorghum plant as well as [26] on basil plant found that ascorbic acid treatment caused an increase in lamina and midvein thickness and dimensions. From the above mentioned results, it is obvious that 50 ppm ascorbic acid showed a marked superiority over the control in most anatomical features of the leaf.

These results were consistent with the positive effects of foliar application of ascorbic acid which may be due to its role as an enzyme cofactor, detoxifies reactive oxygen species generated through photosynthesis process so it considers as multifunctional supporting plant in growth and development [27].

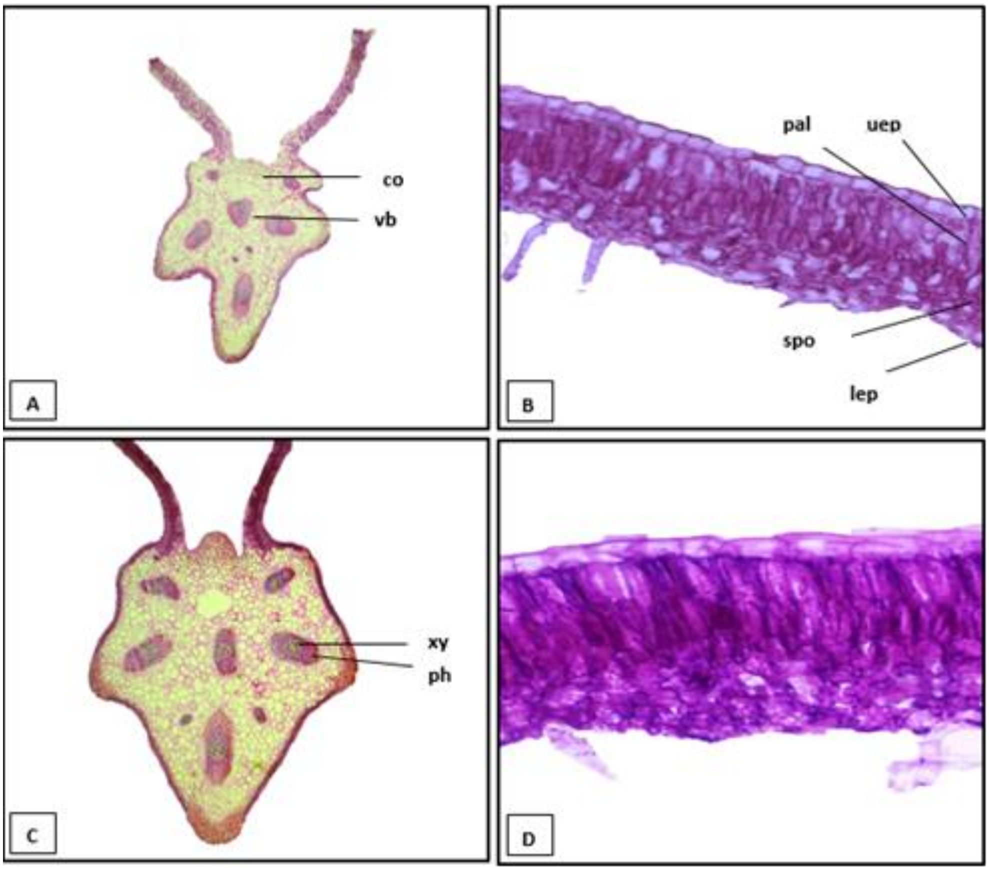

Fig 1: Transverse sections of the middle leaf blade developed at the median portion of the main stem of Cardoon (*Synara cardunculus* var. *altilis*) plant as affected by foliar application with ascorbic acid.

(A & B) from untreated plant, (C & D) from plant treated with 50 ppm ascorbic acid
Details: co, collenchyma tissue; lep, lower epidermis; pal, palisade tissue; PH, phloem; SPO, spongy tissue; UEP, upper epidermis; VB, vascular bundle; XY, xylem.

Table 3: Average measurements (µ) and counts of transverse sections through the middle portion of Cardoon (*Synara cardunculus* var. *altilis*) plant leaf at 5 months age as affected by foliar application of ascorbic acid

Characters	Control	Ascorbic (50 ppm)	% over the control
Average thickness of leaf lamina (µ)	183.84	267.41	45.46
Thickness of upper epidermis (µ)	24.23	35.40	46.10
Thickness of lower epidermis (µ)	3.84	6.40	66.67
Av. Thickness of palisade tissue (µ)	108.55	155.33	43.10
Av. Thickness of spongy tissue (µ)	39.88	66.46	66.65
Av. dimensions of the midvein:			
Length (µ)	2998.77	6365.66	112.28
Width (µ)	2739.52	4535.75	65.57
Av. Thickness of collenchyma tissue (µ)	88.50	267.41	202.16
Av. No. of vascular bundles (µ)	8.00	8.00	0.00
Av. large vascular bundle dimensions:			
Length (µ)	450.91	830.33	84.15
Width (µ)	287.47	315.88	9.88
Xylem tissue thickness (µ)	156.90	256.07	63.21
Phloem tissue thickness (µ)	137.12	266.57	94.41
Xylem vessel diameter (µ)	21.39	39.08	82.70
Av. No. of xylem vessels	17.00	28.00	64.71
Av. small vascular bundle dimensions:			
Length (µ)	334.22	531.12	58.91
Width (µ)	201.08	304.25	51.31
Xylem tissue thickness (µ)	132.25	188.87	42.81
Phloem tissue thicken (µ)	95.52	200.17	109.56
Xylem vessel diameter (µ)	20.37	34.50	69.37
Av. No. of xylem vessels	9.00	23.00	155.56

3.3. Herb fresh weight (g), flower stem height (cm) and head dry weight (g)

Data in Table (4) revealed that in both seasons, the treatment of ascorbic acid with or without nufatrien was most effective on the herb fresh weight than all the other treatments. Whereas, the rate of 50 ppm ascorbic acid produced the heaviest herb fresh weight with values of 2946.67 and 2888.33 (g) in the first and second seasons, respectively. These findings were in harmony with those mentioned by [28] who improved the vegetative growth of geranium plants by spraying with ascorbic acid at the rate of 200 mg/l.

The tallest flower stem was found with the plants received the combination of nufatrien and 50 ppm ascorbic acid with values of 147.0 and 137.67 cm followed by the treatment of 50 ppm ascorbic acid lonely with values of 142.33 and 135.0 cm in the first and second seasons, respectively. Also in both seasons, the treatment of nufatrien plus 10 ppm folic acid caused enhancement in the flower stem height with no significant differences from the previous treatments of ascorbic acid.

Concerning the head dry weight the obtained results found in Table (4), showed that the best head dry weight found with the treatments nufatrien plus 100 ppm ascorbic acid and nufatrien plus 10 ppm folic acid with no significant differences between them.

These results were in accordance with [29] who found gradually increasing in growth parameters of *Codiaeum variegatum* L. as a result of increasing the foliar application of ascorbic acid from 100 to 200 ppm. On *Solanum melongena*, it was observed that morphological and physiological parameters were significantly improved with application of 200 ppm ascorbic acid [24].

Table 4: Effect of nutrients mixture, ascorbic acid and folic acid on herb fresh weight (g), flower stem height (cm) and head dry weight (g) of cardoon plant

Treatments	Herb fresh weight (g)		Flower stem height (cm)		Head dry weight (g)	
	First season	Second season	First season	Second season	First season	Second season
Control	1330.00^{e}±194.7	1413.00^{d}±158.4	92.67c ±37.2	99.67c±21.7	10.40^{c}±0.0	10.00^{d} ±0.0
Nufatrien	2181.67^{c}±134.8	1813.33^{bcd}±278.9	112.67abc±19.4	114.00abc±18.5	19.51^{bc}±1.4	19.98^{bcd}±2.6
Asc50	2946.67^{a}±37.5	2888.33^{a}±187.5	142.33ab±9.5	135.00ab±31.6	22.70^{ab}±11.5	23.43^{abc}±16.4
Nufatrien+Asc50	2918.33^{a}±74.2	2876.67^{a}±509.3	147.00a±6.6	137.67a±24.0	19.50^{bc}±4.2	19.63^{bcd}±4.5
Asc100	2613.33^{b}±110.3	2261.67^{b}±165.1	109.33bc±17.2	119.67abc±14.0	15.84^{bc}±4.8	18.47^{cd}±6.8
Nufatrien+Asc100	2811.67^{ab}±192.8	2863.33^{a}±283.0	130.00ab± 21.8	130.67abc±28.3	30.51^{a}±10.1	32.63^{a}±1.4
Fol10	2261.67^{c}±71.8	2246.67^{b}±302.9	94.67c±13.3	100.67c±20.2	14.63^{bc}±0.8	19.47^{cd}±4.9
Nufatrien+Fol10	1908.33^{d}±185.4	2198.33^{bc}±100.0	133.67a±14.4	135.00ab±17.8	30.03^{a}±1.7	30.38^{ab}±3.7
Fol25	1696.67^{d}±196.2	1733.33^{cd}±293.8	108.00bc±32.8	103.33bc±20.8	10.91^{c}±0.8	10.63^{d}±1.5
Nufatrien+Fol25	1145.00^{e}±67.3	1365.00^{d}±273.7	117.00abc±11.3	122.67abc±5.5	22.09^{ab}±3.3	22.85^{abc}±2.1

Different letters indicate significant differences according to Duncan's multiple range test (P = 0.05)

Nufatrien= macro and micronutrients mixture at the rate of 10 ml/3L Asc50= ascorbic acid at the rates of 50 ppm, Asc100= ascorbic acid at the rates of 100 ppm Fol10= folic acid at the rates of 10ppm Fol25= folic acid at the rates of 25 ppm

3.4. Number of heads/plant, number of seeds/head and seeds weight /head (g)

The data presented in Table (5) showed that in most cases, the number of heads per plant was significantly improved by nutrients mixture, ascorbic acid, and folic acid treatments. The greatest number of heads per plant was found with plants received 50 ppm ascorbic acid with values of 14.67 and 12.67 in the first and second seasons, respectively. Similar result recorded by [28] who found that the number of inflorescence per plant was improved by sprayed geranium plants with ascorbic acid at the rate of 200 mg/l.

Cardoon seeds have the power of antioxidants due to their phenolic contents. Cardoon seeds are a source of fixed oil which can be consumed as an alternative vegetable oil [4]. For this importance, seed parameters were studied and discussed in Table (5). In both seasons, the biggest number of seeds per head (196.0 and 211.0) produced from plants sprayed with 50 ppm ascorbic acid followed by (179.33 and 177.0) from plants sprayed with nufatrien plus10 ppm folic acid.

Concerning seeds weight per head, data in Table (5) revealed that using nutrients mixture alone or in combination with ascorbic acid or folic acid caused a noticeable improvement in seeds weight per head with one exception when using nutrients mixture combined with 100 ppm ascorbic acid caused an insignificant decrease in seeds weight per head compared with control. The heaviest seeds weight per head was recorded from plants supplied with nufatrien combined with 10 ppm folic acid with values of 5.91 and 5.57 g/head due to the great number of seeds per head with values of 179.33 and 177.00 in the first and second seasons, respectively. This result was in agreement with that reported by [13] who found that weight of pea seeds and yield significantly were enhanced by the folic acid application because it has a positive effect on growth, yield, and quality of plants. Also, it was found that folic acid significantly increased seed number per head of the sunflower plant, maybe due to its biochemical function in the synthesis of the amino acid. Also, it can be used to increase the performance of the plant and maintain its nutrients [30].

3.5. Seed yield/plant (g), seed oil% and total flavonoids (mg/g DW)

From data presented in Table (6) it could be expected that seed yield per plant is correlated with the number of heads per plant, the number of seeds per head and seeds weight /head. The obtained result showed that, seed yield per plant was in the same trend of parameters mentioned previously in Table (5). In both seasons, the highest seed yield per plant was obtained from plants treated with 50 ppm ascorbic acid alone or in combination with nufatrien and plants treated with nufatrien plus10 ppm folic acid with insignificant differences between them compared with the other treatments.

Table 5: Effect of nutrients mixture, ascorbic acid and folic acid on number of heads/plant, number of seeds/head and seeds weight /head (g) of cardoon plant

Treatments	Number of heads/plant		Number of seeds/head		Seeds weight/head (g)	
	First season	Second season	First season	Second season	First season	Second season
Control	6.00e±1.0	5.33d±1.5	110.33de±25.7	108.33de±13.5	2.94bcd±1.7	2.71cde±0.6
Nufatrien	9.33bcd±1.2	9.00bc±2.6	157.67abc±29.2	159.67bc±32.2	5.21abc±1.5	5.34a±0.7
Asc50	14.67a±3.5	12.67a±1.5	196.00a±25.4	211.00a±34.8	4.22abcd±3.6	4.94ab±0.3
Nufatrien+Asc50	10.00bcd±1.0	9.67abc±1.5	174.67ab±26.3	165.00b±32.8	5.64ab±0.5	5.36a±0.6
Asc100	8.00de±1.0	7.67cd±3.1	117.33cde±34.5	112.00de±33.3	3.86abcd±1.6	3.73bc±1.9
Nufatrien+Asc100	11.00b±0.0	11.33ab±0.6	113.33de±21.2	107.67de±9.1	2.39d±1.0	2.06e±0.4
Fol10	10.67bc±2.1	10.67abc±2.3	80.67e±22.0	69.33e±3.1	2.47cd±0.1	2.19de±0.4
Nufatrien+Fol10	8.67bcd±0.6	9.00bc±1.0	179.33ab±32.5	177.00ab±16.8	5.91a±1.4	5.57a±0.8
Fol25	8.33cde±0.6	8.67bc±1.5	118.00cde±15.6	118.67cd±17.6	3.00bcd±0.4	3.13cde±0.4
Nufatrien+Fol25	9.67bcd±0.6	8.67bc±1.5	144.67bcd±18.8	145.33bcd±31.6	3.55abcd±0.8	3.57bcd±0.3

Different letters indicate significant differences according to Duncan's multiple range test (P = 0.05)
Nufatrien= macro and micronutrients mixture at the rate of 10 ml/3L Asc50= ascorbic acid at the rates of 50 ppm, Asc100= ascorbic acid at the rates of 100 ppm Fol10= folic acid at the rates of 10ppm Fol25= folic acid at the rates of 25 ppm

In the first season, there were insignificant differences in oil % between the studied treatments except with the treatment of 50 ppm ascorbic acid which gave the highest oil% (30.25%). In the second season, the treatment of 50 ppm ascorbic acid ranked first in the seed oil percentage (30.78%) and the treatment of 100 ppm ascorbic acid ranked second (26.75%) while the treatment of 10 ppm folic acid ranked third (26.02%). These results were in accordance with [31] on flax and [32] on geranium, who found that ascorbic acid as a foliar application significantly increased essential oil percent of the plants.

Concerning the leaf content of total flavonoids (mg/g DW) which are major bioactive compounds of the cardoon leaf, having many benefits against incurable diseases, data in Table (6) indicated that all the studied treatments improved the cardoon leaf content of total flavonoids compared with the control plants. Supplying plants with the combination of nutrients mixture and vitamins (ascorbic or folic acid) had better improvement on total flavonoids than the vitamin alone.

In both seasons, the highest total flavonoids content was obtained with nufatrien alone (2.83 and 3.17 mg/g DW) followed by the treatment of nufatrien plus 100 ppm ascorbic acid (2.46 and 2.35 mg/g DW) compared with other treatments. It was noticed an improvement in cardoon and artichoke leaf quality (total polyphenols, phenolic acids and flavonoids) with use lower fertilizer concentration in the nutrient solution and cardoon plant has a potential use as valuable sources of phenolic acids and flavonoids than artichoke [33].

These results were in accordance with [34] who found that spraying the "Red spur" apple tree with ascorbic acid significantly enhanced the total antioxidant activity and total flavonoids that in turn increase fruit quality.

Table 6: Effect of nutrients mixture, ascorbic acid and folic acid on seed yield/plant (g), seed oil% and total flavonoids (mg/g DW) of cardoon plant

Treatments	Seed yield/plant (g)		Seed oil%		Total flavonoids (mg/g DW)	
	First season	Second season	First season	Second season	First season	Second season
Control	17.64d±10.6	14.45c±8.7	21.85b±1.6	22.49e±0.9	1.42e±0.32	1.77c±0.30
Nufatrien	48.63abc±21.1	48.06abc±23.3	22.81b±0.4	23.03de±0.9	2.83a±0.44	3.17a±0.03
Asc50	61.89a±35.9	62.45a±47.4	30.25a±2.9	30.78a±1.3	1.98cd±0.14	1.99bc±0.02
Nufatrien+Asc50	56.00ab±31.2	48.24abc±33.5	23.58b±0.6	23.84cde±0.5	2.03cd±0.13	2.00bc±0.01
Asc100	30.88bcd±11.6	28.60abc±11.4	24.94b±7.0	26.75b±3.4	2.22bc±0.15	2.06bc±0.31
Nufatrien+Asc100	26.29bcd±20.7	23.35bc±1.2	21.88b±0.4	22.05e±1.0	2.46b±0.36	2.35b±0.42
Fol10	26.35bcd±11.7	23.36bc±21.9	24.94b±4.8	26.02b±2.2	2.11bc±0.14	1.98bc±0.21
Nufatrien+Fol10	51.22abc±12.3	50.13ab±20.1	22.76b±3.3	23.52cde±1.5	2.16bc±0.16	1.98bc±0.14
Fol25	25.00cd±7.5	27.13bc±22.6	24.90b±1.1	25.27bc±0.6	1.67de±0.20	1.83c±0.12
Nufatrien+Fol25	34.32abcd±19.6	30.94abc±8.2	24.47b±1.1	25.09bcd±0.8	1.90cd±0.13	1.97bc±0.18

Different letters indicate significant differences according to Duncan's multiple range test (P = 0.05)
Nufatrien= macro and micronutrients mixture at the rate of 10 ml/3L Asc50= ascorbic acid at the rates of 50 ppm, Asc100= ascorbic acid at the rates of 100 ppm Fol10= folic acid at the rates of 10ppm Fol25= folic acid at the rates of 25 ppm

3.6. Total sugars, phenols and free amino acids (mg/g FW)

The data presented in (Figure 2) illustrated that the control plants had the lowest total sugars, phenols, and free amino acids compared to the other studied treatments. Using nufatrien alone led to a noticeable increase in plant content of bio-compounds.

Using nufatrien combined with 50 or 100 ppm ascorbic acid or combined with 10 ppm folic acid caused a synergistic effect on the cardoon leaf content of total sugars and total free amino acids. The highest total free amino acid was observed with the treatment of nufatrien combined with 10 ppm folic acid with values of 13.8 and 14.6 mg/g FW in the first and second seasons, respectively.

On the other hand, the highest total phenols content was observed from plants treated with 50 ppm ascorbic acid alone compared to the other studied treatments with values of 21.0 and13.5 mg/g FW in the first and second seasons, respectively. Folic acid has an important biochemical role in amino acid metabolism and nucleic acids synthesis [14]. The obtained results concerning the effect of ascorbic acid on total soluble sugars were in agreement with the findings reported by [23, 29 and 32] who noticed that ascorbic acid enhanced the plants' contents of chlorophylls, total carbohydrates, and total soluble sugars.

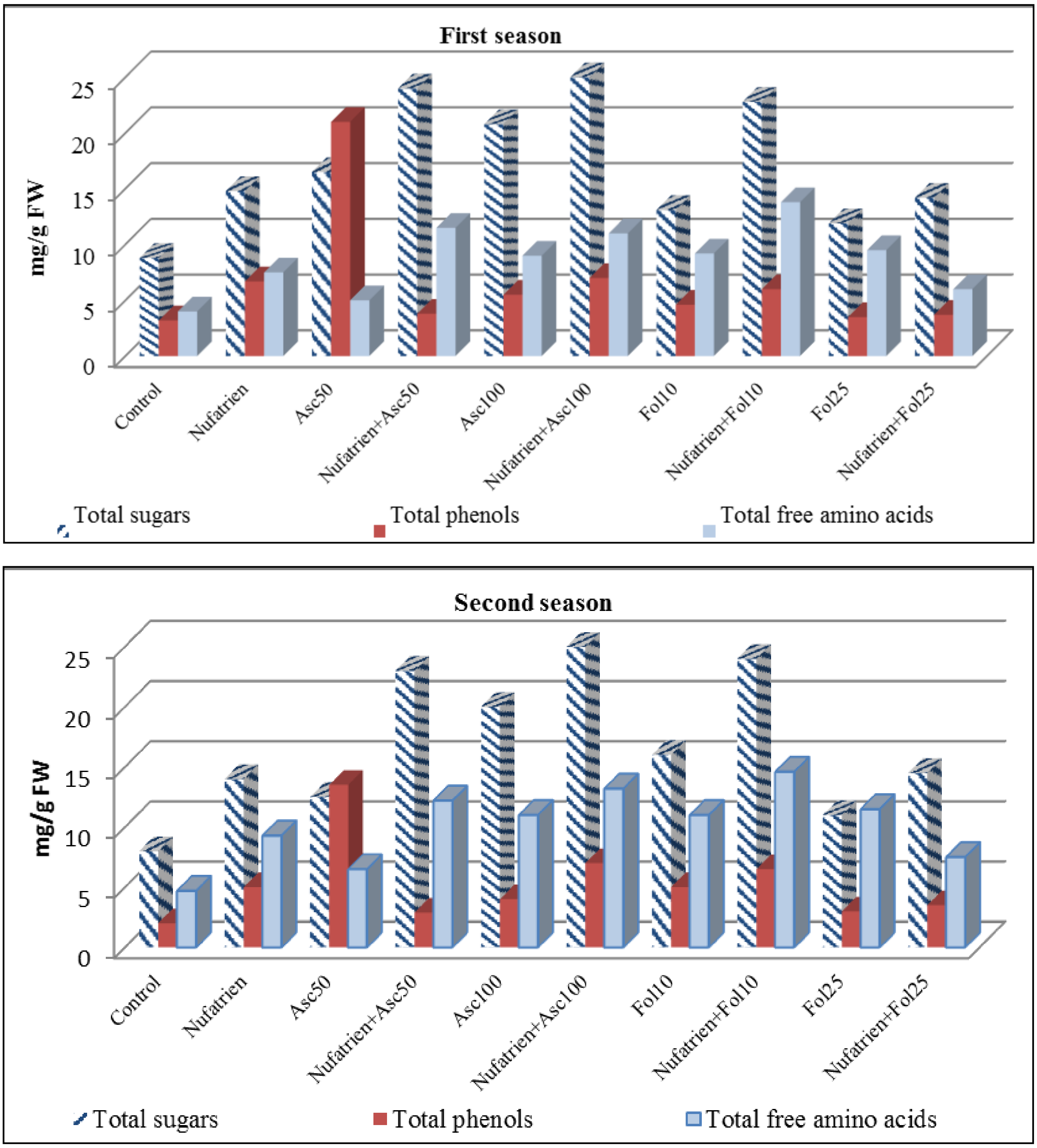

Fig 2: Effect of nutrients mixture, ascorbic acid and folic acid on total sugars, phenols and free amino acids (mg/g FW) of cardoon plant.

3.7. Oil constituents by GC

Data presented in Table (7) showed that cardoon oil obtained from different treatments had good quality because it contained higher quantity of total identified unsaturated fatty acids (Myristelaidic, Oleic and Linoleic) than quantity of total identified saturated fatty acids which caused stability against oxidation. Folic acid had a better effect on improving the content of total unsaturated fatty acids than ascorbic acid. The highest total unsaturated fatty acids (1056.9 mg/100g) found with the treatment of 10 ppm folic acid compared to other treatments.

It was observed that, from the detected unsaturated fatty acids, linoleic was ranked first then Myristelaidic acid ranked second. The highest content of linoleic (759.63 mg/100g) found with the treatment of 25 ppm folic acid followed by the treatment of 10 ppm folic acid with value (736.78 mg/100g). Linoleic acid is a major nutrient because the human body cannot synthesize it inside, so human have to consume it in the daily diet. Linoleic acid is capable of maintaining of the epidermis from losing moisture [35].

Myristelaidic acid is monounsaturated omega-5 long-chain fatty acid. It is isomer of myristoleic acid which is considered anti-cancer of prostate cells [36]. The highest content of Myristelaidic acid (291.9 mg/100g) found with the treatment of 10 ppm folic acid. On the flax plant, it was found a decrease in the saturated fatty acids (palmitic and stearic acids) and, increase linolenic acid. The increasing in linoleic acid might attribute to the acceleration of the biosynthetic pathway of linolenic [37]. Vitamins play an important role in lipid metabolism reactions and their present necessary for these reaction occur moreover folic acid and vitamin C influence lipid metabolism [38].

Table 7: Effect of nutrients mixture, ascorbic acid and folic acid on cardoon oil constituents by GC (mg/100g) in the second season

Oil constituents	Control	Nufatrien	Asc50	N+Asc50	Asc100	N+Asc100	Fol10	N+Fol10	Fol25	N+Fol25
(C8:0)Caprylic	2.38	0.00	1.41	0.84	0.00	0.00	0.00	0.00	0.00	3.66
(C11:0)Undecylic	13.13	17.23	2.16	7.39	15.87	2.26	24.60	9.78	11.44	15.69
(C12:0)Lauric	32.29	40.63	49.46	18.14	38.21	6.17	56.49	25.91	29.09	35.89
(C13:0)Tridecylic	103.91	121.39	151.16	56.13	114.67	18.60	178.23	76.97	87.55	111.4
(C14:0)Myristic	44.81	51.98	65.53	23.44	5.09	7.60	80.44	34.45	38.36	47.84
(C14:1)Myristelaidic	163.39	190.41	243.4	56.56	183.18	52.0	291.9	134.7	146.6	175.5
(C16:0)Palmitic	6.79	6.75	7.15	5.44	3.49	0.00	6.50	7.57	7.22	5.55
(C18:0)Stearic	1.83	1.88	1.67	1.04	0.79	1.97	1.71	1.75	2.00	2.87
(C18:1)Oleic	27.79	30.30	25.34	18.43	11.25	33.40	28.23	37.22	34.86	24.29
(C18:2)Linoleic	680.20	280.76	586.51	468.05	396.19	613.6	736.78	717.94	759.63	494.3
Total identified unsaturated fatty acids	871.4	501.5	855.3	543.0	590.6	699.0	1056.9	889.9	941.1	694.1
Total identified saturated fatty acids	205.1	239.9	278.5	112.4	178.1	36.6	348.0	156.4	175.7	222.9

Nufatrien= macro and micronutrients mixture at the rate of 10 ml/3L Asc50= ascorbic acid at the rates of 50 ppm, Asc100= ascorbic acid at the rates of 100 ppm Fol10= folic acid at the rates of 10ppm Fol25= folic acid at the rates of 25 ppm

4. Conclusion

From the previous results, we recommend supplying the Cardoon plant with 50 ppm ascorbic acid to obtain large decorative silver leaves for the ornamental garden as well as improve the plant material for medicinal and industrial purposes of this plant. This recommendation was based on that ascorbic acid at 50 ppm had a superior effect on the average leaf fresh weight, herb fresh weight, Number of heads per plant, number of seeds per head, seed yield per plant, seed oil percentage and total phenols content compared to other treatments. Also, this treatment caused a marked increase in anatomical features of cardoon leaf.

5. References

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