

Chemical Constituents, Growth and Flowering of *Coreopsis tinctoria* Nutt Plants as Affected by Glutamic Acid and Irrigation by Different Wastewater Treated Effluents

Nahed, G. Abdel Aziz¹; Hussein, I. Abdel- Shafy^{2*} and Bedour H. Abou Leila³

¹Ornamental and Woody Trees Dept., National Research Centre, Dokki, Cairo, Egypt.

^{2*}Water Research and Pollution Control Dept., National Research Centre, Dokki, Cairo, Egypt.

³Water Relation and Field Irrigation Dept., National Research Centre, Dokki, Cairo, Egypt.

THIS study deals with the effect of glutamic acid and the reuse of chemically treated wastewater on the chemical constituents of *Coreopsis tinctoria* Nutt Plants in a batch-pot experiment. This chemical constituent includes the nutrient elements (N, P, K), micronutrient elements (Fe, Mn), sodium, as well as heavy metals (Cd, Cu, Zn). In this study different doses of glutamic acid were examined in combination with different types of treated wastewater to investigate the environmental stress on *Coreopsis tinctoria* Nutt Plants. The study was conducted during the two successive seasons of (2015/2016– 2016/2017) in the greenhouse of National Research Centre, Cairo, Egypt, aimed to study the response of *Coreopsis tinctoria* plants to glutamic acid application at four chemical concentrations (0, 50, 100 and 150 ppm) as well as the effect of treated wastewater as a source of irrigation. Besides, this investigation includes the impact on growth and flowering of *Coreopsis tinctoria* plants. The physical and chemical characteristics of the treated wastewaters were investigated and reused to determine the beneficial advantages as an additional water resource. It was concluded that Glutamic acid and irrigation with three types of treated wastewater effluents individually induced favorable changes in *Coreopsis tinctoria* Nutt plants in terms of chemical constituents, vegetation and flowering. Thus, the effluent of the chemically treated municipal wastewater can be reused safely without any side effect. Meanwhile, type of treatment has an important effect on the final chemical characteristics of the treated wastewater, particularly the nutrient elements and heavy metals. The reuse of treated wastewater is the most important challenge in the near future to increase the water budget in Egypt as the population escalating and the stringent need for more cultivated land. Consequently, the need for extra safe water resources should be supplied.

Keywords: Glutamic acid, chemical Constituents of Nutt plants, Chemical characteristics of treated wastewater, Heavy metals accumulation, Chemical nutrient elements.

Plants can be used to treat and/or prevent several diseases is on the basis of many traditional treatments. Additionally, they are regarded as important sources of essential nutrients and health - beneficial components that are crucial for human life due to their importance as source of vitamins, minerals, phenolic compounds, volatiles and fatty acids [1]. Meanwhile, the presence of fatty acids in plants is important due to the fact that they protect the plants from the biological and environmental stress constitute. In addition, the presence of fatty acids in plants is considered energy storage [2]. Fatty acids in the human organism act like hormones or their precursors. They are a source of metabolic energy besides; they help the digestion process [3].

The *Coreopsis tinctoria* Nutt Asteraceae, is a native from North America. Presently, it spread worldwide. Its flowering tops infusion

is; traditionally; used in several countries to control hyperglycemia [4]. Despite these of these advantages, the *C. tinctoria* flowering tops constitute is a poorly studied matrix. However, few studies were conducted recently have in terms of its chemical composition with the promotion of glucose tolerance [5].

On the other hand, the importance of glutamic acid lies on the synthesizing of the auxin and fruit set [6]. Several investigations proved that amino acids can either directly or indirectly influence the physiological activities on the growth and the development of plants. According to these investigations, the foliar application of amino acids induces an enhancement in plant growth and fruit yield. Besides, the maintenance of protein component is regulated in garlic [7], cucumber [8] and sweet pepper [9]. In a study by Neeraja et al. [10], they noticed an increase in the

*Corresponding author email: hshafywater@yahoo.com, waterbiotech@yahoo.com;

Mobile: +201224018017 & +201124170008

DOI : 10.21608/ejchem.2017.1726.1145

©2017 National Information and Documentation Center (NIDOC)

number of flowers, fruit setting and fruit yield of tomatoes due to the amino acids application. On the other side, addition of amino acids did not affect the fresh weight of the shoot and root of the plant [11, 12]. It was then concluded that spraying amino acids is effective on growth and yield of strawberry. The requirement of amino acids nitrogen in essential quantities for the purpose to increase the growth and the yield for all crops is well documented for all crops. In addition, amino acids and/or nitrogen are the essential ingredients for the protein synthesizing process. The importance of amino acids or nitrogen is attributed to their widely use for the bio-synthesizing of great varieties of non-protein nitrogenous materials i.e., pyrimidine bases, pigments, coenzymes, Purine and vitamins [9].

The purpose of the present study is to investigate the effect of glutamic acid on the chemical constituents, growth, and flowering of *Coreopsis tinctoria* nutt plants. In addition, the effect of water reuse in irrigation on the studied plant was also investigated. For this purpose, three types of wastewater treated effluents were employed for irrigating this *Coreopsis tinctoria* Nutt Plants.

Materials and Methods

The present experiments were conducted at the "Training Demonstration Center (TDC)", National Research Center, Cairo, Egypt. The study was carried out in pot experiments, and the experimental work continued from 20 November 2015 till 20 July 2016 and from 25 November 2016 till 25 July 2017. The aim of this investigation is to study the response of *Coreopsis tinctoria* plants to glutamic acid grown under the influence of irrigation with treated municipal wastewater.

Real municipal wastewater is separated into black (B), grey (G) and yellow (Y) water segregated and collected from one house across the Training Demonstration Center (TDC) site in the National Research Center (NRC), Cairo, Egypt. This house comprises of two separated sides. Each side consists of five apartments. One side is presently connected to the TDC site as separated B, G and Y water manholes. The source of blackwater (B) is the toilet including faeces, urine and the flushing water. On the other hand, the collected greywater (G) includes wastewater from bath, showers, hand wash basins, washing machines, dishwashers and kitchen sinks. Description of the different treated wastewaters

Egypt.J.Chem. **60**, No.6 (2017)

that are used in this study is as follow:

Black water (1) [treated wastewater 1] is the treated black water using membrane bioreactor (MBR). Full description of the procedure is given in Abdel-Shafy and El-Khateeb [13]

Black water (2) [treated wastewater 2] is the treated black water using hybrid (namely horizontal followed by vertical flow) constructed wetlands. Details of the experiments and results are shown in Abdel-Shafy et al. [14]

Greywater (1) [treated wastewater 3] is the treated greywater using different hybrid treatment processes for handling the grey water for unrestricted reuse. The treatment system consists of sedimentation that was enhanced by Effective Micro-organism (EM) followed by aeration. The purpose is to reach the unrestricted water reuse. The final treated effluent could cope with the permissible limits of the unrestricted water reuse according to "Egyptian Guideline" regulations. Details of this study, the experiments and results are shown in Abdel-Shafy et al. [15]

The physical and chemical characteristics of the three types of the treated wastewater are given in Table 1.

For cultivation of the plants, plastic pots size 30 cm in diameter and 30 cm in depth were used. Each pot was filled with media containing peat and sand at the ratio of (1:1) by volume. Seedlings of *Coreopsis tinctoria* were kindly supplied by the Ministry of Agriculture, Ornamental Plants Research Centre, Cairo, Egypt. The experimental work was conducted in two seasons; each season consists of 16 treatments. In addition, four different treatments with glutamic acid were examined namely (0, 50, 100 and 150 ppm). Two months homogeneous seedlings of *Coreopsis tinctoria* were selected and transplanted on 20 and 25 November, respectively, for the studied two seasons. Phosphorus fertilizers as calcium superphosphate were added. The contents of this fertilizer are (16% P₂O₃) at the rate of 4.0 g/pot, calcium 4.0 g/pot as calcium nitrate (15.5% N) and 2.0 g/pot as potassium sulphate (48.5% K₂O). This fertilizer was added in four different intervals namely, 4, 8, 16 and 20 weeks from transplanting seedlings. The irrigation by the treated wastewater was applied 15 days from time of transplanting. The quantity of the added water was adjusted to bring the soil moisture to field capacity by weighting pots and daily loss of water

was supplemental every day. After 15 days and 30 days from transplanting the plants were sprayed with glutamic successively.

This study was conducted in complete randomized design with three replicates for each treatment. The irrigation was carried out with the 3 types of treated wastewater alone or in combination with the variable concentrations namely (1, 50, 100 and 150 ppm) of glutamic acid.

Eight months after transplanting and during the flowering stage, plant samples were collected random wise in three replicates from each set. The following data were recorded:

The present study was concerning also with the following: Plant height (cm), number of branches, number of leaves, stem diameter (mm), root length (cm), fresh and dry weight of shoots and roots (gm), number of flowers, flower diameter (mm), fresh and dry weight of flowers (gm). Meanwhile, fresh leaves (gm/plant) were collected to determine photosynthetic pigments (chlorophyll a, b and carotenoids). Furthermore, leave samples were collected and were oven dried at 70°C for the determination of nitrogen, phosphorus and potassium contents. In addition,

metals including sodium, cadmium, copper, manganese, zinc and iron contents in *Coreopsis tinctoria* plants were determined according to Abdel-Shafy et al. [16]. The mean values of the determined data were statistically analysis using the completely randomized design in factorial arrangement [17]. The mean values of the studied treatments were compared using L.S.D. test at 0.05 level of probability [18].

Results and Discussion

Physical and chemical characteristics of Reused treated wastewater

The characteristics of the three types of treated water are given in Table 1. These results showed that the treatment systems were efficient to remove any hazard components from wastewaters. However, treated blackwater (1) contains higher level of nitrates and organic nitrogen. The rest of parameters were much closed to each other (Table 1).

Growth parameters

Effect of glutamic acid

Data presented in Table 2 showed that foliar application of glutamic acid on *coreopsis tinctoria* plants increased plant height, number of branches and leaves, diameter of stem and length of root significantly as correlated with control plants

TABLE 1. Physical and chemical characteristics of the three types of water that were used for irrigation.

Parameters	Number of samples	Treated water No. (1) Blackwater treated with MBR (average)	Treated water No. (2) Blackwater treated with Hybrid constructed wetlands (average)	Treated water No. (3) Greywater enhanced sedimentation followed by aeration (Average)	Permissible limits 3 rd group for (Advanced Treatment)
pH	7	7.8 (0.3)	7.8(0.3)	6.8 (0.2)	6.5 – 9.0
TSS (mg/l)	7	2.3	7.6(2.1)	10.50	20
COD (mg/l)	7	34- 76 (43)	18(5.1)	38.17	40
BOD (mg/l)	7	6-18 (10)	18.0 (8.2)	19.58	20
TP (mg/l)	7	3 – 7 (5.8)	8.9(1.1)	0.80	---
NO ₃ (mg/l)	7	270-335 (302)	0.01(0.01)	0.30	---
Oil & greaser (mg/l)	7	5.08	4(3.8)	5.08	5
TKN (mg/l)	7	12	23.6(2.72)	10.89	---
SAR (%)	7			14.23	20

particularly at the 100ppm. The increments were (21.7, 68.7, 53.9, 35.3 and 31.4%) respectively compared with control plants. The positive effect of glutamic acid is that plant growth regulators may produce their effects within the part in which they were synthesized. It was stated that certain messengers called plant growth substances are generally like hormones in their action, controlled utilization of nutritional substances for a balanced coordinated development in plant body [19]. Growth regulators are used as natural compounds that are applied directly to the target plant for altering their life process or structure. The purpose is to improve quality and productivity of the plants. Furthermore, it facilitates harvesting. Similar increases in growth parameters resulted from amino acid treatments have been previously observed by several investigators and was exhibited by several plant species. Examples are on *Codiaeum variegatum* L. [19], on *Pelargonium graveolens* plants [20], on *Salvia farinacea* plants [21] and on *Gladiolus grandiflorum* [22]. These investigators found that amino acids significantly increased vegetative growth. Table 2 exhibited the glutamic acid that affected on fresh and dry weight of shoots and roots /plant. The lowest (52.55 and 6.52 gm) fresh weight of both shoots and roots and (14.18 and 2.24 gm) dry weight of shoots and roots were obtained by the control plants. The highest values of these characters were obtained when 100 ppm glutamic acid was used. The values of fresh and dry weight were (123.02 and 36.17 gm) and (10.00 and 3.36 gm) for shoots and roots respectively. It was pointed out by Thom *et al.* [18] that amino acids provide plant cells with an immediately available source of nitrogen, which can be up taken by the cells more rapidly than inorganic nitrogen. In addition, Bidwell [23] stated that amino acids are known as the building blocks of proteins. In plants, amino acids serve in number of additional functions in regulating the metabolism, transport and storage of nitrogen [24]. In particular, it was observed in many systems that increased RNA synthesis accompanied growth, indicating that continuous protein synthesis was necessary during the growth. On the other hand, the response of vegetative growth to the application of amino acids were studied and their results are in good agreement with El-Fawakhry and El-Tayeb [25] on *Chrysanthemum* and Abdel-Aziz *et al.* [26] on *Thuja orientalis* plants and Abd El Al *et al.* [27] on squash plants.

Effect of water type

Table 3 represents the effect of using different quality of water on the vegetative stage as indicated by the mean values of the growth parameters. The results show that water surpassed both black water (2) and gray water on plant height, branches number, root length, fresh and dry weight of shoots and roots/ plant. However, stem diameter did not exhibit any significant differences. On the other hand, black water (2) was more effective in increasing flower number followed by black water (Blk-MBR). Treated greywater (Gry-CW) was more effective in increasing flower diameter compared with other types and untreated "control" plants. Meanwhile, Table 3 shows that the three types of treated wastewater induced an increase in both fresh and dry weight of the flowering stage significantly.

Effect of interaction

The interaction between glutamic acid and the use of different types of treated wastewater on certain growth parameters were presented in Table 4. In this respect the interaction exhibited the difference between the three types of water interacted with the chosen concentration of glutamic acid. The percentage was increased in plant height and branches number attained under the combined effect of 100 ppm glutamic acid and treated black water (1). The increments reached 19.7 and 72.9%, respectively over the control plants (Table 4). Meanwhile, the reduction percentage on the previously mentioned parameters reached 19.5 and 45.1%, respectively in correlation with the control. The study was extended to record the effect of irrigation with greywater in combination with 30 ppm glutamic acid, in one hand, and with blackwater (2) in combination with 150 ppm glutamic acid, on the other hand. In addition, the combined effect of 100 ppm glutamic acid and blackwater (2) reached the highest increments of leaves, stem diameter, root length, and fresh and dry weights of shoot, namely (151.5, 47.2, 68.9, 215.9 and 253.7%), respectively in comparison with the control one. The lowest values were exhibited by the single treatment of gray water.

Flowering

Effect of glutamic acid

Glutamic acid exhibited positive effect on the flower parameters (number of flowers, flower diameter, fresh and dry weights of flowers). The data recorded in Table 2 indicated that foliar application of glutamic acid significantly

TABLE 2. Effect of glutamic acid on vegetative and flowering stages of Coreopsis tinctoria plants during 2015 / 2016 -and 2016 / 2017. seasons.

Treatments	Vegetative stage										Flowering stage			
	Plant height (cm)	No. of branches/plant	No. of leaves/plant	Stem diameter (cm)	Root length (cm)	F.W of shoots(gm/plant)	F.W of roots (gm/plant)	D.W of shoots(gm/plant)	D.W of roots(gm/plant)	No. of flowers/plants	Flower diameter (cm)	F.W of flowers (gm)/plant	D.W of flowers(gm) /plant	
Control	55.2	10.48	47.43	0.34	5.25	52.55	6.52	14.18	2.24	6.6	3.7	22.6	3.80	
50	57.9	15.08	67.68	0.44	6.30	77.30	7.72	21.81	2.55	9.3	4.5	34.5	6.28	
100	67.2	17.68	73.0	0.46	6.90	123.02	10.00	36.17	3.36	10.2	4.1	31.3	5.87	
150	59.8	13.73	52.83	0.39	6.30	81.77	6.87	23.19	2.10	9.2	4.2	31.3	5.86	
L.S.D at 5%	0.79	0.55	0.64	N.S	0.47	5.68	0.73	0.69	0.43	0.42	0.31	0.60	0.39	

TABLE 3. Effect of waste water treated affluent on vegetative and flowering stages of Coreopsis tinctoria plants during 2015 / 2016 -and 2016 / 2017.

Treatments	Vegetative stage										Flowering stage			
	Plant height (cm)	No. of branches/plant	No. of leaves/plant	Stem diameter (cm)	Root length (cm)	F.W of shoots (gm/plant)	F.W of roots (gm/plant)	D.W of shoots (gm/plant)	D.W of roots(gm/plant)	No. of flowers (gm/plant)	Flowers diameter (cm)	F.W of flowers (gm)/plant	D.W of flowers (gm)/plant	
Control	60.6	14.40	50.13	0.40	6.55	67.60	7.25	18.69	2.35	8.0	4.0	18.3	3.2	
Black 1	64.2	17.07	62.83	0.42	6.77	107.70	11.82	31.34	4.07	9.0	3.1	41.7	8.0	
Black 2	58.9	13.23	65.60	0.45	6.35	85.90	5.23	24.70	1.99	10.6	4.1	34.9	6.4	
Gray	56.3	12.25	62.38	0.35	5.03	73.75	5.82	20.60	1.85	7.7	4.4	24.8	4.2	
L.S.D at 5%	0.79	0.55	0.64	N.S	0.47	5.68	0.73	0.69	0.43	0.42	0.31	0.60	0.39	

TABLE 4. effect of interaction between glutamic acid and west water treated effluent on vegetative and flowering stages of *Coreopsis tinctoria* plants during two seasons (2015 / 2016 -and 2016 / 2017).

Treatments	Vegetative stage										Flowering stage				
	Plant height (cm)	No. of branches	No. of leaves	stem Diameter	roots Length	F.W of shoots	D.W of shoots	F.W of roots	F.D of roots	No. of flowers	Diameter of flower	F.W of flowers	D.W of flower		
Control	59.3	13.3	39.6	0.36	5.8	50.8	13.61	7.0	1.90	6.7	2.7	10.8	1.72		
G. 50	56.3	12.0	49.6	0.46	6.3	55.3	14.98	4.3	1.28	6.7	4.9	16.5	2.78		
G. 100	68.4	13.7	66.7	0.39	5.8	85.3	24.22	11.7	4.00	9.7	3.3	18.1	3.09		
G.150	58.6	18.6	44.6	0.37	8.3	79.0	21.96	6.0	2.25	9.0	5.0	27.6	5.30		
Black 1	60.0	13.0	56.3	0.41	7.2	81.0	22.84	9.0	4.19	5.3	3.0	40.5	7.29		
Black 1+G. 50	63.3	14.3	83.7	0.44	7.3	89.9	25.80	14.8	5.18	15.3	4.3	49.8	9.46		
Black1 +G. 100	71.0	23.0	61.3	0.43	6.5	127.3	37.55	11.4	3.87	11.3	4.7	40.5	8.10		
Black1 +G. 150	62.6	18.0	50.0	0.42	6.3	131.4	39.15	12.1	3.03	4.3	3.8	36.0	7.12		
Black 2	49.7	7.3	54.7	0.40	3.5	39.1	10.04	6.9	1.66	9.0	5.0	19.5	3.17		
Black 2+G. 50	64.3	18.3	63.4	0.51	6.3	99.6	28.88	4.7	1.46	7.0	3.7	49.5	9.20		
Black2 +G. 100	67.0	20.0	99.6	0.53	9.8	160.5	48.15	8.0	2.64	12.3	4.0	34.4	6.70		
Black2 +G. 150	54.7	7.3	44.4	0.38	5.8	44.4	11.76	5.3	2.20	14.3	4.0	36.2	6.41		
Gray	51.7	8.3	39.1	0.19	4.5	39.3	10.21	3.2	1.20	5.3	4.0	19.6	3.05		
Gray +G.50	47.7	15.7	73.7	0.37	5.3	64.4	17.58	7.1	2.31	8.3	5.2	22.2	3.68		
Gray + G. 100	62.3	14.0	64.4	0.47	5.5	119.0	34.74	8.9	2.96	7.7	4.4	32.2	5.60		
Gray +G. 150	63.3	11.0	72.3	0.37	4.8	72.3	19.88	4.1	0.92	9.3	4.0	25.2	4.61		
L.S.D at 5%	1.59	1.12	1.29	N.S	0.94	11.47	1.38	1.48	0.88	0.86	0.62	1.22	0.79		

increased flower parameter in correlation with control plants. The concentration of 50ppm gave the highest values of these parameters except number of flowers. The flowers were increased when plants treated with 100ppm glutamic acid. The favorable effect of such treatment may be due to its effect on branches number which increased due to this treatment (Table 2). In this respect, the stimulatory effect of the amino acids were found to be correlated with the increase in content and activity levels of endogenous promoters particularly gibberellins and IAA. The later are known to promote linear growth of plants organs [28, 29]. The present results are compatible with those obtained by Wahba et al. [30] on *Antholyza acthypoica* and El-Fawakhry and El-Tayeb [25] on chrysanthemum. They found that foliar application of amino acids led to increment of flowering parameters and produced a high quality of inflorescences.

Effect of water type

Data in Table 3 showed that at flowering stage, the blackwater (2) surpassed both blackwater (1) and graywater (GW) in number of flowers/ plant. When GW was used for irrigation, the highest values of flower diameter namely 4.4cm were obtained. With regard to the effect of three water type for irrigation on fresh and dry weight of flowers, it is clear from data that irrigation with blackwater (1) increased fresh and dry weight of flowers/ plant significantly. The increments were (127.8 % and 150.0%) respectively, as compared with control plants.

Effect of interaction

The effect of interaction on number of flowers, flowers fresh and dry weight was studied. Results in Table 4 showed that employing treated blackwater (1) combined with the 50 ppm glutamic acid was more effective in increasing the studied parameters to 128.3, 361.1 and 450.0% respectively, compared with untreated plants (Table 4). The flower diameter increased significantly when plants were treated with treated greywater (Gry-CW) combined with 50ppm glutamic acid. It reached 92.5% as compared with control plants (Table 4).

Chemical constituents

Photosynthetic pigments

Effect of glutamic acid

Data in Table 5 showed that photosynthetic pigments were affected by glutamic acid application on *Coreopsis tinctoria* plants.

Increasing concentration from 50ppm up to 100 ppm gave higher values for chlorophyll a, total chlorophylls and carotenoid, namely 1.572, 2.026 and 0.235 mg/g, respectively. While the highest concentration decreased chlorophyll a, b and total chlorophylls. Nevertheless, the concentration of 150 ppm exhibited the highest carotenoids mean value. The previous studies proved that amino acids can directly or indirectly influence the physiological activities of the plant. These results are in good agreement with that reported by other investigators [7, 31].

Effect of water type

Data (Table 6) indicated that blackwater (1) gave the highest mean values for chl. "a" and total chlorophylls followed by graywater. Meanwhile, chl. b and carotenoids increased to 0.483 and 0.252 mg/g respectively as watered with treated greywater. The lowest values for photosynthetic pigments were recorded as the system was watered with treated blackwater (2).

Effect of interaction between water type and glutamic acid

Results are illustrated in Fig 1. It is evident that both blackwater (1) and blackwater (2) in combination with 100ppm glutamic acid were affected positively on chl. a, b and total chl. The control one showed lesser effect (Fig.1). On the other hand, the interaction between Blackwater (1) and 150ppm glutamic acid was more effective (Table 7).

Macronutrients percentage and uptake

Effect of glutamic acid

Figure 2 showed the chemical properties of N, P, K and Na% and the uptake of *Coreopsis tinctoria* plants. Figure 3 illustrates the effect of variable glutamic acid concentration in combination with reusing different types of treated wastewater on the plant uptake of N, P, K and Na as (g/plant). The given results showed that the uptake of N, P, K and Na% were influenced by the foliar spraying of glutamic acid during the two studied seasons (Table 7). The highest concentrations of glutamic acid increases the percentage and uptake of N and P compared with untreated plants. The uptake of N and P increments were (34.4 and 163.6%) and (117.8 and 350.0%), respectively compared with controlled plants. The application of glutamic acid at rate of 100 ppm induced a promotion effect (i.e. increasing the uptake of K). The lowest concentrations of glutamic acid led to an increase of the Na percentage. On the contrary, the uptake

of Na increased when plants were treated with glutamic acid at 100 ppm. The accumulation of photosynthetic pigments as a result of these nitrogen compounds may be attributed to the important role of nitrogen in the biosynthesis of chlorophyll molecules according to Meyer *et al.* [32]. These results (Table 7) are in agreement with those obtained by other investigators [33]. They found that foliar application of amino acids increased the level of total nitrogen, total phosphorus and total potassium on rosemary plants. Such increments led to the quantitative changes in amino acids and specific proteins that are affected positively in cell division and cell elongation on carnation plants [33]. Meanwhile, Davis [34] clarified that amino acids as organic nitrogenous compounds are the building blocks in synthesis of proteins which formed by a process in which ribosome catalyze the polymerization of amino acids.

Effect of water type

Data in Tables 3 and 4 showed that the highest values of N and P % were obtained by using blackwater (2). On the other hand, the use of treated grey-water for irrigation led to the increase the K and Na % compared with control plants. Furthermore, the uptake of N, P and Na was increased by the irrigation with blackwater (1). Nevertheless, the highest values of K uptake was obtained when plants was irrigated with treated grey-water. All results were compared with control plants.

Effect of interaction

It is clear from Fig. 2 & 3 that the highest uptake of N and P% was achieved when plants were irrigated with Blk-MBR water in combination with 150 ppm glutamic acid and Blk-CW water combined with 100 ppm glutamic acid compared with the controlled one. Meanwhile, irrigating the plant with Gry-CW in combination with 150 ppm or 100ppm glutamic acid led to increase the uptake of K % in the plant compared with the control plants. Furthermore, irrigating plants with Blk-CW in combination with 50 ppm glutamic acid caused an increase in the up of Na%. The same founding was obtained when Gry-CW combined with 100 ppm glutamic acid was examined.

Uptake of heavy metals by Coreopsis tinctoria plants as affected by the level of Glutamic acid

Effect of glutamic acid

The effect of different glutamic acid concentration on the level of heavy metals namely *Egypt.J.Chem.* **60**, No.6 (2017)

TABLE 5. Effect of glutamic acid on photosynthetic pigments and mineral nitration of *Coreopsis tinctoria* plants during two seasons(2015 / 2016 -and 2016 / 2017).

Treatments	Photosynthetic pigments (mg/g f.w)					Mineral nutrition								
	Chl. a		Chl. b		A+b	N%	P%	K%	Na%	Cd mg/kg	Cu mg/kg	Mn mg/kg	Zn mg/kg	Fe mg/kg
	1.138	0.393	0.455	1.747	1.531	2.31	0.12	0.92	0.54	3.04	10.7	54.7	42.6	134.8
Control	1.293	0.455	0.249	1.747	1.531	2.66	0.13	1.04	0.55	3.33	10.8	61.0	43.6	138.4
50	1.596	0.508	0.281	2.104	2.104	3.51	0.15	1.20	0.51	3.07	11.7	63.5	44.9	139.4
100	1.346	0.417	0.252	1.763	1.763	3.97	0.16	1.35	0.47	3.49	13.5	69.9	48.6	142.5

TABLE 6. Effect of waste water treated effluent on photosynthetic pigments and mineral nitration of *Coreopsis tinctoria* plants during two seasons (2015 / 2016 -and 2016 / 2017).

Treatments	Photosynthetic pigments					Mineral content								
	Chl. a		Chl. b		A+b	N%	P%	K%	Na%	Cd mg/kg	Cu mg/kg	Mn mg/kg	Zn mg/kg	Fe mg/kg
	1.268	0.422	0.480	1.996	1.690	1.65	0.19	1.07	0.42	3.12	11.9	58.2	47.9	127.9
Control	1.516	0.480	0.311	1.996	1.690	3.73	0.09	1.07	0.48	3.06	11.2	61.4	43.1	146.4
Black 1	1.285	0.438	0.206	1.723	1.723	3.85	0.20	1.14	0.57	3.42	12.3	59.3	45.2	139.4
Black 2	1.303	0.433	0.252	1.736	1.736	3.21	0.08	1.23	0.60	3.33	11.1	70.4	43.5	141.8

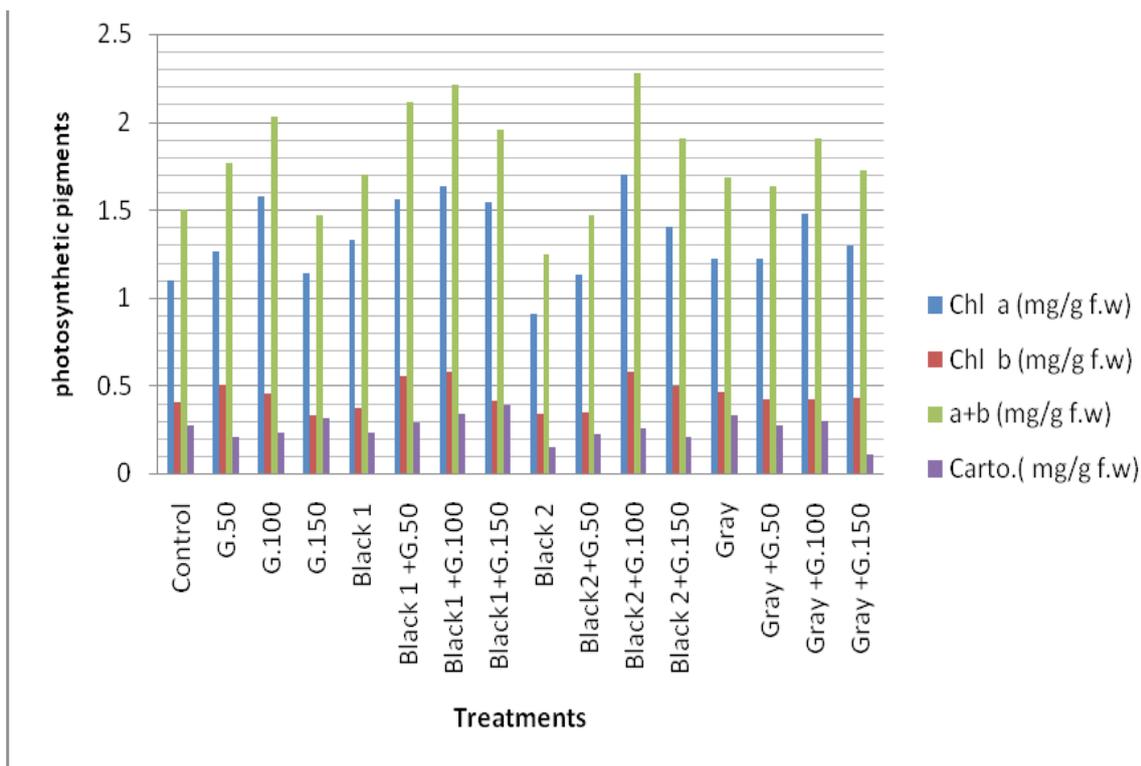


Fig. 1. Effect of glutamic acid, waste water treated effluent and their interaction on photosynthetic pigments (mg/g f.w) of *Coreopsis tinctoria* plants during two successive seasons.

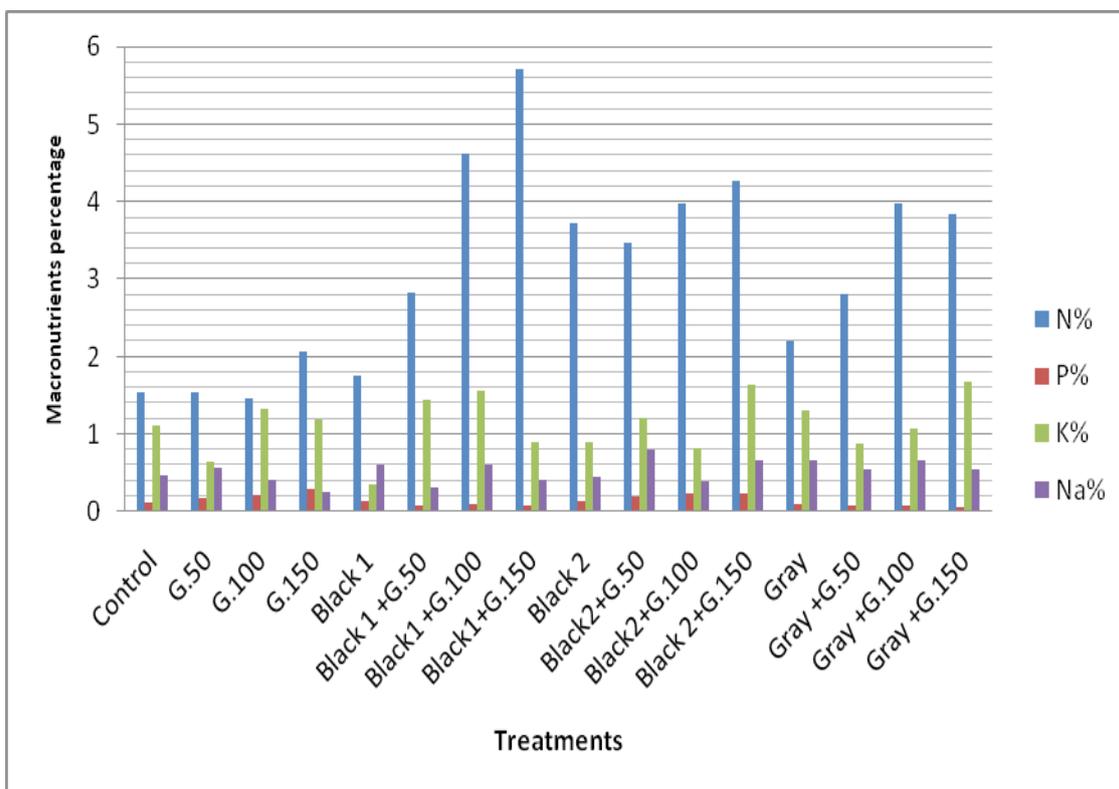


Fig.2. Effect of glutamic acid, waste water treated effluent and their interaction on macronutrients percentage of *Coreopsis tinctoria* plants during two successive seasons.

TABLE 7. Effect of interaction between glutamic acid and wastewater treated effluent on photosynthetic pigments and mineral nutrients of *Corcopsis tinctoria* plants during two seasons (2015 / 2016 -and 2016 / 2017).

Treatments	Photosynthetic pigments				Mineral content										
	Chl a (mg/g f.w)	Chl b mg/g f.w)	A+b mg/g f.w)	carot mg/g f.w)	N%	P%	K%	Na%	Cd mg/ kg	Cu mg/ kg	Mn mg/ kg	Zn mg/ kg	Fe mg/kg		
Control	1.098	0.405	1.503	0.275	1.54	0.11	1.11	0.46	2.78	12.1	46.4	42.8	138.3		
G.50	1.264	0.504	1.768	0.209	1.54	0.18	0.65	0.57	3.40	10.5	57.4	51.6	113.1		
G.100	1.572	0.454	2.026	0.235	1.46	0.21	1.33	0.40	3.18	12.0	60.4	49.2	123.9		
G.150	1.139	0.326	1.465	0.314	2.07	0.29	1.18	0.26	3.12	13.1	68.0	48.0	136.2		
Black 1	1.328	0.368	1.696	0.234	1.76	0.13	0.36	0.60	2.98	10.5	50.0	46.0	148.22		
Black 1 +G.50	1.558	0.556	2.114	0.287	2.83	0.07	1.45	0.31	3.26	10.9	65.4	43.8	139.7		
Black1 +G.100	1.632	0.580	2.212	0.340	4.62	0.09	1.56	0.60	3.14	12.3	61.3	37.4	147.7		
Black1+G.150	1.544	0.415	1.959	0.384	5.71	0.07	0.90	0.40	2.86	10.9	68.7	45.0	150.1		
Black 2	0.905	0.337	1.242	0.147	3.72	0.14	0.90	0.44	3.26	11.3	53.2	45.2	130.7		
Black2+G.50	1.129	0.343	1.472	0.222	3.47	0.20	1.20	0.79	3.47	11.9	57.4	44.0	155.4		
Black2+G.100	1.699	0.576	2.275	0.254	3.98	0.24	0.82	0.39	3.10	12.0	62.4	40.2	128.5		
Black 2+G.150	1.406	0.497	1.903	0.203	4.26	0.23	1.63	0.66	3.84	13.7	64.2	51.4	143.0		
Gray	1.219	0.462	1.681	0.332	2.21	0.09	1.31	0.66	3.13	8.9	69.2	36.4	122.0		
Gray +G.50	1.220	0.417	1.637	0.276	2.80	0.08	0.87	0.55	3.19	9.9	63.8	34.8	147.2		
Gray +G.100	1.480	0.423	1.903	0.295	3.98	0.08	1.07	0.66	2.86	10.3	69.8	52.8	157.4		
Gray +G.150	1.296	0.429	1.725	0.107	3.84	0.06	1.67	0.54	4.12	15.5	78.9	49.8	140.5		

Cd, Cu, Mn, Zn and Fe is presented in Fig. 4. These results revealed that spraying *Coreopsis tinctoria* plants with glutamic acid stimulated the uptake of heavy metals compared to the control, namely Cd, Cu, Mn, Zn, and Fe. On the contrary, increasing the concentrations of glutamic acid decreases

the percentage of Na. The highest uptake values of Cu, Mn and Fe were obtained when the dose of 150 ppm glutamic acid was implemented. These results are in good agreement with Mazher et al. [19] on *Cymbopogon citratus* and *Codiaeum variegatum* L. plants. Nevertheless,

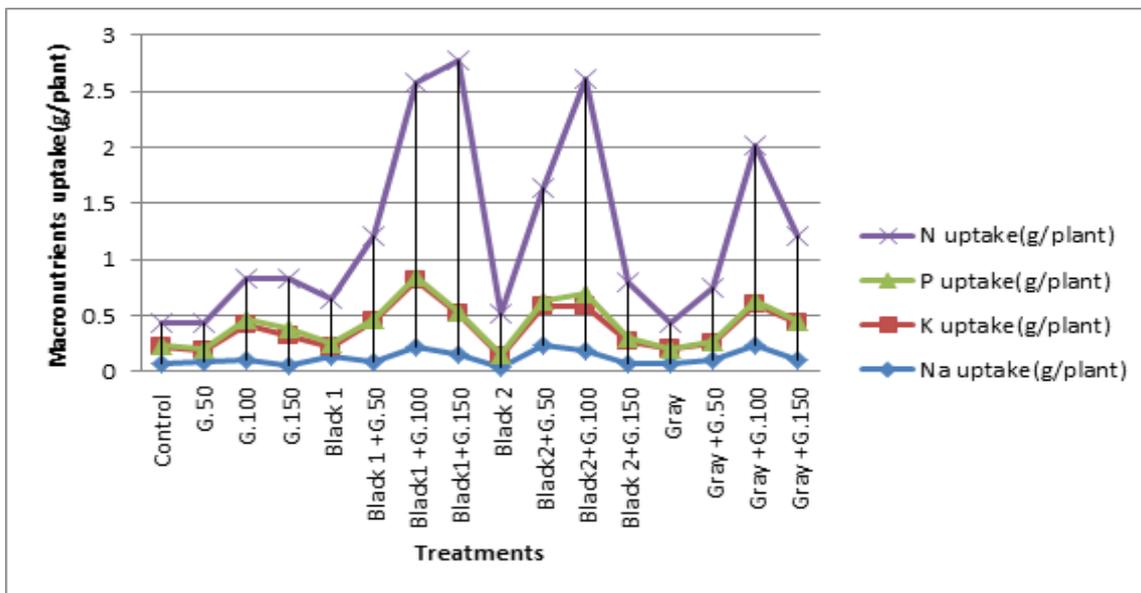


Fig. 3. Effect of glutamic acid, waste water treated effluent and their interaction on macronutrients uptake (g/plant) of *Coreopsis tinctoria* plants during two successive seasons.

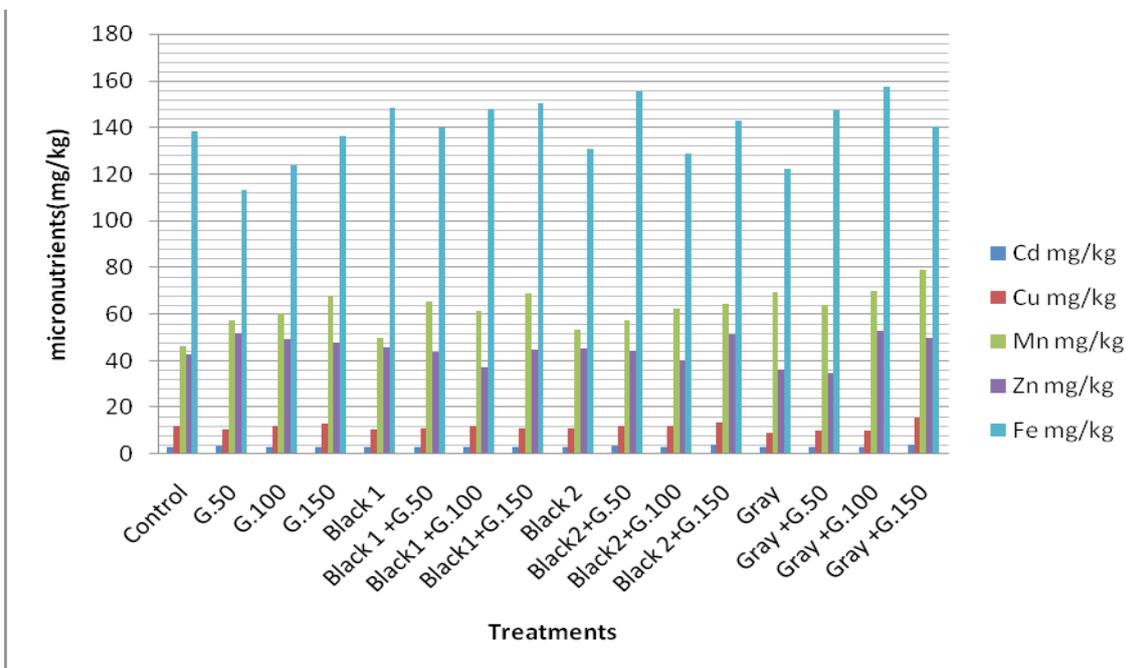


Fig. 4. Effect of glutamic acid, waste water treated effluent and their interaction on micronutrients (mg/kg) of *Coreopsis tinctoria* plants during two successive seasons.

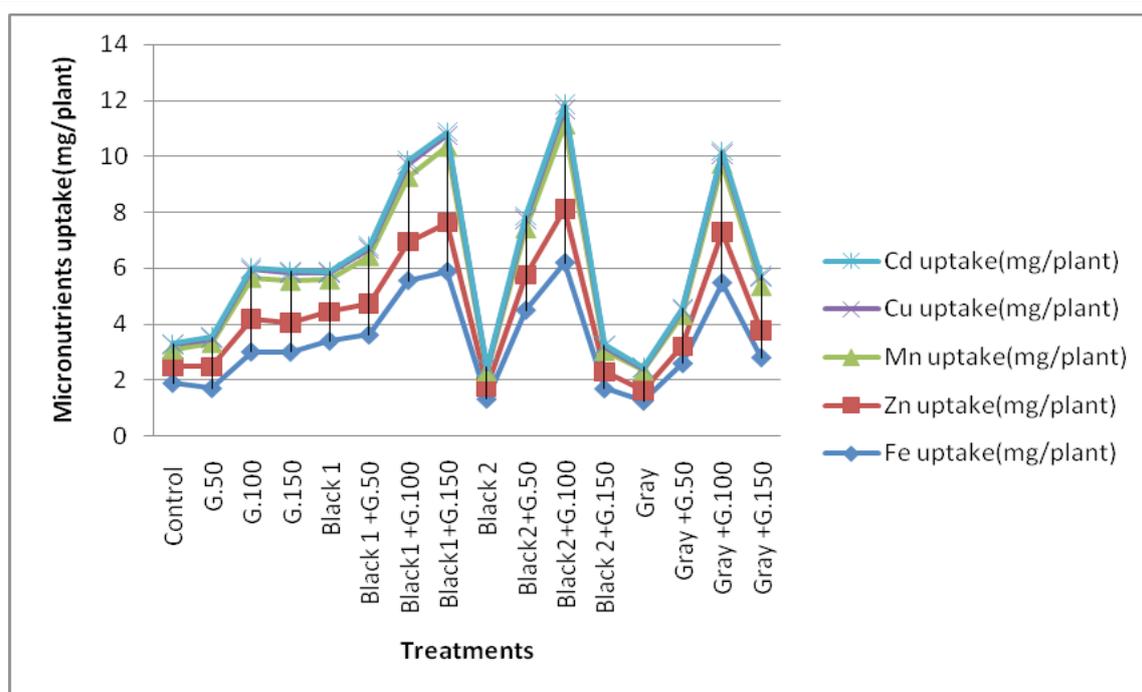


Fig.5. Effect of glutamic acid , waste water treated effluent and their interaction on micronutrients uptake (mg/plant) of *Coreopsis tinctoria* plants during two successive seasons.

available information for the relationship between amino acids and macro and micronutrients are limited. While micronutrients like (Fe, Mn and Zn) were reported to improve roots growth leading to greater absorbing capacity of the root surface and consequently increasing the nutrient uptake from the soil [35]. Such improvement could be reflected on increasing the dry matter accumulation through the improvement of photosynthesizing production and mobilization of plants [17, 36].

Effect of water type

The data is illustrated in Fig. 4 as mg/kg. From these results it can be concluded that, during the study of the two seasons, blackwater (2) induced a marked increase in the level of Cd and Cu compared to the control plants. Meanwhile, Mn exhibited the highest level by irrigation with greywater. However, the observed increase in Zn and Fe resulted from blackwater (1). The effect of water type on micronutrients uptake is presented in Fig.5 as mg/plant. These results indicated that the highest values of Cd, Cu, Mn, Zn and Fe were obtained by irrigation with blackwater (1). The increments were (83.7, 45.7, 80.9, 80.4 and 79.8%), respectively compared with control plants (Table 7).

Effect of interaction

From Fig. 4 and 5, it can be concluded that employing 150 mg/l glutamic acid and irrigating

with greywater caused the highest values of Cd, Cu and Mn. Moreover, irrigating plants with gray water type and + 100mg/l glutamic acid caused increase in Zn and Fe during the two studied both seasons. Furthermore, the combinations between glutamic acid at 100 mg/l and blackwater (2) were almost positive for the uptake of Cd, Cu, Mn, Zn and Fe (Table 7). The increments were 302.7, 251.8, 376.0, 232.4 and 228.7, respectively compared with control plants. These results confirmed that reuse of treated municipal wastewater can be considered as an additional water resources and safe for irrigation as confirmed by several investigators [37-44].

Coculusion

Glutamic acid and irrigation with three types of treated wastewater effluents individually induced favorable changes in *Coreopsis tinctoria* Nutt plants in terms of vegetation, flowering and chemical constituents.

Effluent of the treated municipal wastewater can be reused for irrigation safely without any side effect. Meanwhile, type of treatment has an important effect on the final characteristics of the treated wastewater, particularly the nutrient elements for the plant.

Thus it is recommended to treat the municipal

wastewater in a way to preserve the nutrient elements as much as possible in case of reusing for irrigation.

Recommendation

Adequate treated wastewater can be reused safely as an additional water resources. The quality of the reused water depends mainly on the implemented treatment technology.

Glutamic acid is an efficient chemical to enhance the growth of plants.

The combination between treated wastewater and glutamic acid possess a great advantage for the uptake of nutrient elements by plants.

Acknowledgment: The authors wish to express their deep appreciation and gratitude to the facilities provided by the project titled "Sustainable Development for Wastewater Treatment and Reuse via Constructed Wetlands in Sinai (SWWTR)" that is funded by STDF of Egypt.

References

1. Grusak M.A., Phyto chemicals in plants: genomics-assisted plant improvement for nutritional and health benefits. *Curr Opin Biotech* **13**, 508-511 (2002).
2. Ohlrogge J.B., Regulation of fatty acids synthesis. *Annu. Rev. Plant Phys.*, **48**, 109-136 (1997).
3. Burtis C.A. and Ashwood E.R. *Tietz Fundamentals of Clinical Chemistry*, 4th ed. Philadelphia: W.B. Saunders Company (1996).
4. Oliveira D. and Feijão R., *Medicina pelas Plantas*. Lisboa: Livraria Progresso Editora. (1973).
5. Dias T., Bronze, M.R., Houghton P.J., Mota-Filipe, H. and Paulo, A., The flavonoid-rich fraction of *Coreopsis tinctoria* promotes glucose tolerance regain through pancreatic function recovery in streptozotocin-induced glucose intolerant rats. *J Ethnopharmacol* **132**, 483-490 (2010a).
6. Taiz L. and Zeiger E. *Plant physiology*. Sunderland: Sinauer. p. 690. (2002).
7. Awad E.M.M., Abd El-Hameed A.M. and Shall Z.S., Effect of glycine, lysine and nitrogen fertilizer rates on growth, yield and chemical composition of potato. *J. Agric. Sci. Mansoura Univ.*, **32**(10), 8541-8551 (2007).
8. El-Shabasi M.S., Mohamed S.M and Mahfouz S.A., Effect of foliar spray with amino acids on growth, yield and chemical composition of garlic plants. *The sixth Arabian Conf. for Hort., Ismailia, Egypt*. (2005).
9. Kamar M.E. and Omar A., Effect of nitrogen levels and spraying with aminal-forte (amino acids salvation) on yield of cucumber and potatoes. *J. Agric. Sci. Mansoura Univ.*, **12**(4), 900-907(1987).
10. Neeraja G., Reddy I.P. and Gautham B., Effect of growth promoters on growth and yield of tomato cv. Marutham. *J. Res. A.N.G.R.A.U.*, **33**(3), 68-70 (2005).
11. Cerdan M., Sanchez S.A., Oliver M., Juarez M. and Sanchez A.J.J., Effect of foliar and root application of amino acid on iron uptake by tomato plants. *Acta Hort.*, **830**, 481-4488 (2006).
12. Shehata S.A., Gharib A.A., El-Mogy M.M., Abdel G.K.F. and Shalaby E.A., Influence of compost, amino and humic acids on the growth, yield and chemical parameters of strawberries. *J. Med. Plants Res.*, **5**(11), 2304-2308 (2011).
13. Abdel-Shafy Hussein I. and El-Khateeb M. A., Membrane Bioreactor for the Treatment of Municipal Blackwater in Egypt. *J. Desalination and Water Treatment*, **29**(1-3), 56-62 (2011). doi: 10.5004/dwt.2011.1746
14. Abdel-Shafy Hussein I., El-Khateeb M.A. and Shehata M., Blackwater treatment via combination of sedimentation tank and hybrid wetlands for unrestricted reuse in Egypt. *J. Desalination and Water Treatment, Jan.*, **71**, 145-151 (2017).
15. Abdel-Shafy Hussein I., Makki Al-Sulaiman A. and Mansour M.S.M., Greywater treatment via hybrid integrated systems for unrestricted reuse in Egypt. *J. Water Process Eng.*, **1**, 101-107 (2014), <http://dx.doi.org/10.1016/j.jwpe.2014.04.001>
16. Abdel-Shafy Hussein I., Mohamed R. Farid and Aly M. Shams El-Din, Water-hyacinth from Nile River: chemical contents, nutrient elements and heavy metals. *Egyptian. J. Chemistry*, **59**(2), pp.131-143 (2016).

17. Tarraf Sh., El-Sayed A.A. and Ibrahim M. E., Effect of some micronutrients on *Rosmarinus officinalis* L. *Egypt. J. Physiol. Sci.*, **18**, 201-211 (1994).
18. Thom M., Maretzki A., Kormer E. and Sokai W. S., Nutrient uptake and accumulation by sugar cane cell culture in relation to growth cycle. *Plant Cell Tissue and Organ Culture*. (1), 3-14 (1981).
19. Mazher A. M., Zaghoul S. M., Mahmoud S. A. and Siam H. S., Stimulatory effect of kinetin, ascorbic acid and glutamic acid on growth and chemical constituents of *Codiaeum variegatum* L. plants. *American-Eurasian J. Agric. & Environ. Sci.*, **10** (3), 318-323 (2011).
20. Mahgoub Mona, H. and Iman M. Talaat Physiological response of rose geranium (*Pelargonium graveolens* L.) to phenylalanine and nicotinic acid. *Annals of Agric. Sci. Moshtohor*, **43**(3), 807-822. (2005).
21. Abd El-Aziz Nahed, G. and Balbaa Laila, K., Influence of tyrosine and zinc on growth, flowering and chemical constituents of *Salvia farinacea* plants. *J. Appl. Sci. Res.*, **3**(11), 1479-1489. (2007).
22. Abd El-Aziz Nahed, G., Lobna T. Taha and Soad M.M. Ibrahim, Some studies on the effect of putrescine, ascorbic acid and thiamine on growth, flowering and some chemical constituents of *Gladiolus* plants at Nubaria. *Ocean J. Appl. Sci.*, **2**(2), 169-179. (2009).
23. Bidwell R. G. S., *Plant Physiology*, 2nd Ed. Collier Macmillan Publisher, London, New York. (1980).
24. Fowden L., Amino acids. In: *Phytochem.* Vol. 11, 1-29, Muller, L.P. Van Nostra and Reinhold. Co., New York. (1973).
25. El-Fawakhry F.M. and El-Tayeb H.F., Effect of some amino acids and vitamins on chrysanthemum production. *J. Agric. Res. Alex. Univ.*, **8**(4), 755-766. (2003).
26. Abdel Aziz Nahed, G., Mazher Azza A.M. and Farahat M.M., Response of vegetative growth and chemical constituents of *Thuja orientalis* L. plant to foliar application of different amino acids at Nubaria. *J. American Sci.*, **6**(3), 295-303. (2010).
27. Abd El-Aal Faten S., Shaheen A.M. Ahmed A.A. and Mahmoud Asmaa R., Effect of foliar application of urea and amino acids mixtures as antioxidants on growth, yield and characteristics of Squash plants. *Res. J. of Agric. and Biolog. Sci.*, **6**(5), 583-588. (2010).
28. Stoddart J.L. Gibberellin receptor. In *Hormones Receptors and Cellualr Interactions in Plants*. (Ed. Chadwick, C.M. and D.R. Carrod) Cambridge Univ. Press. Cambridge, London, New York. (1986).
29. Wilkins, M.B., *Advanced Plant Physiology*. Pitman Publishing Inc. London. (1989).
30. Wahba H.E., Safaa M.M., Attoa G.E. and Farahat A.A. Response of *Antholyza acthipoica* L. to foliar spray with some amino acids and mineral nutrition with sulphur. *Annals of Agric. Sci. Cairo Univ.*, **47**(3), 929-944 (2002).
31. Al-Said, M.A. and Kamal, A.M., Effect of folair spray with folic acid and some amino acids and some amino acids on flowering yield and quality of sweet pepper. *J. Agric. Sci. Mansoura Univ.*, **33**(10), 7403 - 7412 (2008).
32. Meyer, B. S., Anderson. D. B. and Bohning R. H., *Introduction of Plant Physiology*. 179-189. (1968).
33. Bekheta M.A. and Mahgoub M.H. Application of kinetin and phenylalanine to improve flowering characters, vase life of cut flowers as well as vegetative growth and biochemical constituents of carnation plants. *Egypt. J. Appl. Sci.*, **20**(6A), 234-246 (2005)
34. Davies D.D., Physiological aspects of protein turn over *Enycl. Plant Physiol. New Series*, 14 A (Nucleic acids and proteins: structure biochemistry and physiology of proteins). 190-288., Ed. Boulter, D. and Partheir, B. Spring Verlag. Berlin, Heidelberg and New York. (1982).
35. Wittwer S. H. and Bukovac M. J. The uptake of nutrients through leaf surface. In '*Handbuch der Pflanzenemanhung*'. Dungung, U. (Ed.) Hans LInser, 235-261. Springer- Veriag, Wien, N. Y. (1969).
36. El-Leithy A.S., Physiological studies on French marigold (*Tagets patula*, L.) plants. *Ph. D. Thesis*, Fac. Agric. Cairo Univ. (1987).
37. Abdel-Shafy Hussein I. and Aly R.O., Wastewater Management in Egypt" In "*Wastewater Reuse-Risk Assessment, Decision-Making and Environmental*

- Security*" Mohammed K. Zaidi (Ed) Springer Publisher, Netherland, pp.375-382. (2007).
38. Abdel-Shafy Hussein I. and Abdel-Sabour M.F., Wastewater reuse for irrigation on the desert sandy soil of Egypt: long-term effect" In *"Integrated Urban Water Resources Management"* P. Hlavinek et al. (Eds) Springer Publisher, Netherland, pp.301-312. (2006).
39. Abdel-Shafy Hussein I. and Abdel-Shafy Sally H., Membrane technology for water and wastewater management and application in Egypt: Review Article. *Egyptian. J. Chemistry*, **60**(3), 347-360 (2017).
40. Abdel-Shafy Hussein I. and Mansour Mona S.M., Integration of effective microorganisms and membrane bioreactor for the elimination of pharmaceutical active compounds from urine for safe reuse. *Journal of Water Reuse and Desalination*, **6.4**, 495-504. (Available online 13 January 2016).
41. Abdel-Shafy Hussein I., Al-Sulaiman Ahmed Makki and Mansour, Mona S.M. Anaerobic / aerobic treatment of greywater via UASB and MBR for unrestricted reuse. *J. Water Science and Technology*, **71**(4), 630-737 (2015). doi: [10.2166/wst.2014.504](https://doi.org/10.2166/wst.2014.504)
42. Abdel-Shafy Hussein I. and Al-Sulaiman Ahmed Makki, assessment of physico-chemical processes for treatment and reuse of greywater. *Egyptian J. Chemistry*, **57**(3), 215-231(2014).
43. Abdel-Shafy Hussein I., Al-Sulaiman Ahmed Makki and Mansour, Mona S.M., Greywater treatment via hybrid integrated systems for unrestricted reuse in Egypt, *J. Water Process Eng.*, 1,101-107 (2014), <http://dx.doi.org/10.1016/j.jwpe.2014.04.001>
44. Abdel-Shafy Hussein I., Hobus Inka and Hegemann, Werner, upgrading of decentralized ponds for municipal wastewater treatment and restricted reuse, *J. Water Reuse and Desalination*, **01.3**, 141-151 (2011). doi: [10.2166/wrd.2011.022](https://doi.org/10.2166/wrd.2011.022)
- (Received 25 / 9 / 2017;
accepted 31/10 /2017)

تأثير اعادة استخدام المياه ، وتركيزات مختلفة لحمض الجلوتاميك على المحتوى الكيميائي ، والنمو ، والتزهير للنبات

ناهد جلال عبد العزيز^١، حسين ابراهيم عبد الشافي^٢ ، بدور حلمي أبو ليله^٢
^١قسم نبات الزينة والاشجار الخشبية، ^٢قسم بحوث تلوث المياه ، قسم العلاقات المائية والرى الحقلى - المركز القومى للبحوث - القاهرة - مصر.

تتناول الدراسة الحالية تأثير كلا من التركيزات المختلفة لحمض الجلوتاميك، واعادة استخدام المياه على المحتوى الكيميائى لنبات " كينو كاريس " على العناصر المغذية (نترات ، فوسفات ، بوتاسيوم) والعناصر المغذية الصغرى (حديد ، منجنيز) و الصوديوم وكذلك تركيزات المعادن الثقيلة. حيث تمت الدراسة تحت تأثير الرى بالمخلفات السائلة المتباعدة فى الخواص التى تمت معالجتها بتقنيات مختلفة لمعرفة مدى تأثير تلك العوامل على النبات - حيث تمت الدراسة لفترة موسمين متتاليين (٢٠١٥-٢٠١٦ ، ٢٠١٦-٢٠١٧) فى مكان التجارب النصف صناعية - قسم تلوث المياه - المركز القومى للبحوث- حيث استخدمت الدراسة تركيزات مختلفة من حامض الجلوتاميك من (صفر ، ٥٠ ، ١٠٠ ، ١٥٠ مجم/ لتر) بالإضافة الى استخدام المخلفات التى سبق معالجتها كمصدر للرى ، كما تناول البحث الخواص الطبيعية والكيميائية للمخلفات السائلة بعد معالجتها بطرق مختلفة. وقد أظهرت النتائج أن كلا من حامض الجلوتاميك ، والمخلفات السائلة المعالجة لهم تأثير جيد على نمو النبات وتزهيره لما تحتويه المياه المعالجة على بعض العناصر الغذائية للنبات، ويستنتج من الدراسة أن اعاده استخدام المياه المعالجة جيداً كمصدر للرى لا يمكن تجاهله ويجب الاهتمام به كمصدر من مصادر مياه الرى بشكل آمن .