

EFFECT OF GRAINS SOAKING WITH HUMIC ACID AND MICRONUTRIENTS FOLIAR SPRAY ON QUALITY AND PRODUCTIVITY OF RICE PLANT UNDER SALINE SOIL CONDITIONS

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ABSTRACT: *Two field experiments were carried out on a newly reclaimed saline soil during two successive summer seasons (2009 and 2010) at Sahl El- Hossinia, EL- Sharkia Governorate, Egypt to study the response of Egyptian rice variety namely Giza 178 to foliar spray with micronutrients mixture (Zn+ Mn+ Fe) at three rates (0.0, 0.3 and 0.6 g/l for each element) either singly or in combinations with humic acid which applied as soaking treatment at a rate of 100 mg/l for 24 hour before planting. Rice plants were sprayed two times with micronutrients after 45 and 70 days from sowing at a rate of 400 l/fed. in the form EDTA compounds i-e Zn-EDTA (14% Zn), Mn-EDTA (12%Mn) and Fe-EDTA (13% Fe). The obtained results showed the effective action of foliar application with Zn+ Mn+ Fe at a rate of 0.3 g/l for each nutrient in increasing significantly in either shoots or roots dry weights, roots were the most organs affected by such treatment as well as significant improvement in Chlorophyll a, b and carotenoids in leaves for both seasons after 60 days from sowing. Significant increases in yield and yield components, where the percentage of increase in grain yield reached to 75.68 and 77.33% for first and second seasons, respectively. Corresponding increases significantly in macronutrients (nitrogen, phosphorus and potassium) and micronutrients (zinc, manganese, ferrous and copper), crude protein, total carbohydrates in rice grain for two growing seasons. Significant increases were found in germination percentage, shoot length, radical length and electrical conductivity (EC) in grains of rice variety Giza 178 for both seasons. The obtained data declared that the superior interaction effect was recorded for grains soaked with humic acid + micronutrients added as foliar feeding at a rate of 0.3g/l for all studied characters for rice cultivar Giza 178 for two growing seasons.*

Key words: *Rice plant, Micronutrients (Zn+ Mn +Fe), Humic acid.*

INTRODUCTION

Rice (*Oriza sativa L.*) is considered one of the most important summer crops in Egypt. The productivity of rice is affected by many factors such as seeds germination, fertilization and quality of seedbed environment i-e soil moisture, salinity and temperature. Alam *et al.* (2001) noted that salinity

affects the growth of rice in varying degrees at all stages of its life cycle starting from germination up to maturity. Several studies indicated that rice is a salinity tolerant during germination and becomes very sensitive during early seedling stage. Thereafter, grains tolerance during vegetative growth but again becomes sensitive during pollination. However, it increasingly becomes more tolerant at maturity stage. Imtiaze *et al.* (2003) stated that lack of just one micronutrient can greatly reduce plant yield. Adequate nutrition of plant with micronutrients depends on many factors among them is the ability of soil to supply these nutrients, rate of nutrients absorption by the plant, distribution of nutrients to function sites and nutrients mobility within the plant. Gurmani *et al.* (2003) found that, the application of micronutrients to rice plants showed positive effect on yield and its components. Sairam and Tuagi (2004) suggested that foliar spraying with micronutrients especially Fe, Mn and Zn increased crops and mineral contents of many plants types under saline stress conditions.

Humic substances have many beneficial effects on soil and consequently on plant growth and are shown highly hormonal activity. These materials not only increased macronutrients contents and ion uptake but also enhanced micronutrients contents of the plant organs (Brunetti *et al.*, 2005). Salhyabama *et al.* (2004) reported that humic acid increased yield and N, P and K uptake in rice plant. Salib (2002) reported that combination of humic acid at rate of 50 mg/l and foliar application mixture of chelating micronutrients formed of Fe (6%), Mn (12%) and Zn (12%) at concentration of 1g/l significantly increased grain and straw yield, N, P, K concentration and uptake and protein % in grain and straw of wheat plant. Hakan (2008) found that soaking treatment of seed with potassium humate solution at rate of 0.01% for 24 hour before planting and subsequently spraying of the plant with a 0.005% solution significantly increases in various cereal crops yield.

The objective of this investigation are to 1) Study the response of Giza 178 rice variety to micronutrients mixture (Zn+ Mn+ Fe) either individually or in association with humic acid to show the best treatment that gives the highest effect on a) Shoot and roots dry weights. b) Chlorophyll a, b and carotenoids in leaves. c) Yield and its components. d) Macro- and micronutrients concentration in grains. e) Crude protein and total carbohydrates percentage in grains. f) Germination percent, shoot and radical lengths, seedling dry weights and electrical conductivity (EC) in grains and g) Chemical properties of the tested soil (2) Find an explanation for this response on the bases of test attributes.

MATERIALS AND METHODS

Two field experiments were conducted during two successive summer seasons 2009 and 2010 at Sahl EL-Hossinia, EL- Sharkia Governorate to study the efficiency of foliar spray mixture of Zn+ Mn +Fe at three rates (0.0, 0.3 and 0.6 g/l for each nutrient) either singly or in combination with humic

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acid on the yield of rice plant, (*Oriza sativa L.*) Giza 178 cultivar. Representative surface soil samples (0-30) were taken before the performance of the experiment and after harvesting, where some physical and chemical analysis were determined using the standard methods according to Black (1965) as shown in Table (1).

Table (1): Physical and chemical properties of the experimental soil.

Coarse sand (%)	Fine sand (%)	Silt (%)	Clay (%)	Texture	O.M (%)	CaCO ₃ (%)	pH (1:2.5) Soil suspension	EC (dS/m) (soil past)
2.74	48.39	13.20	35.67	Loamy	0.64	12.0	8.30	17.31
Soluble cation (mg/l)				Soluble anion (mg/l)				
Ca ⁺⁺	Mg ⁺⁺	Na ⁺	K ⁺	HCO ₃	Cl ⁻	SO ₄		
9.23	14.99	148	0.83	9.33	38.28	125.44		
Available macronutrients (mg/kg)				Available micronutrients (mg/kg)				
N	P	K	Fe	Mn	Zn	Cu		
42	5.75	196	1.79	1.77	0.86	0.083		

The rice grains were planted in rows on 5 and 10th of May for the first and second seasons, respectively, at a rate of 60 kg/ fed.. The experimental design used was a complete randomized with four replicates. The experimental units were 20 plots, where the area of each plot was m² (3x 3.5).

The experimental treatments were as the following

- a-Control (without micronutrients and humic acid).
- b-EDTA (1) [Zn+ Mn+ Fe at a rate of 0.3g/l for each element].
- c-EDTA (2) [Zn+ Mn+ Fe at a rate of 0.6 g/l for each nutrient].
- d-Humic acid + EDTA (1).
- e-Humic acid +EDTA (2).

All plots received basal doses of phosphorus fertilizer was applied at rate of 30 kg P₂ O₅/fed. as calcium super phosphate (15% P₂O₅) was applied during soil tillage before rice sowing, potassium fertilizer was used at rate of 48 kg K₂O/ fed. as potassium sulphate (48% K₂O), where its applied in two split equal doses, once before rice sowing and later after 35 days from sowing, nitrogen fertilizer at rate of 33.5 kg N/fed. as ammonium nitrate (33.5%), which applied at three times 21, 35 and 50 days from sowing.

The micronutrients mixture added as foliar spray method. Plants were sprayed after 45 and 70 days from sowing at a rate of 400 l/fed. in the form of EDTA (Ethyline diamine tetra acitic acid) compounds i.e. Zn-EDTA (14% Zn), Mn-EDTA (12% Mn) and Fe-EDTA (13% Fe).

Humic acid was added as soaking method. The chemical composition of the used humic acid was determined by using BaCl₂ precipitation methods as described by Fataftah *et al.* (2001) as shown in Table (2). Before start

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soaking, rice grains were surface sterilized in 5% sodium hypochlorite for 5 min. and rinsed well with double distilled water (Anonymous, 1985). Grains were soaked in one liter of 100 mg/l humic acid for 24 hour before planting. Other field practices were followed in the usual manner for rice cultivation. Plants were grown till maturity and sampled twice. The area of each sample was 1m². The first sample was taken after 60 days from sowing, where chlorophyll a, b and caroteinoids in leaves were recorded using spectrophotometer methods recommended by Metzner *et al.* (1965). Also, dry weights of shoots and roots were recorded. The second sample was taken at harvest. The harvest date was on 3 and 5 September for the first and second seasons, respectively. At harvest, agronomic trails were i.e. plant height (cm), number of panicles/m², panicle weight (g/plant), 1000-grains weights, straw and grains yield (ton/fed.).

Table (2): Main Characteristic of the used humic acid.

Compounds and units	Values	Properties unites	Values
Organic matter/total Solid (%)	42.81	Total nitrogen (N, %)*	7.55
Total humic acid/total solid(g/l)	175.11	C/N ratio	2.98
Organic carbon (%)	24.13	Total phosphorus (P ₂ O ₅ , %)*	5.14
pH (1:2)	8.10	Available potassium (K ₂ O, %)*	9.33
EC (dS/m)	5.8	Total calcium (Ca, %)**	0.09
		Total magnisum (Mg, %)*	0.07
		Total boron(B, mg/l)*	73.15
		Total manganese (Mn, mg/l)**	91.62
		Total zinc (Zn, mg/l)**	89.51
		Total iron (Fe, mg/l)**	895.14
		Total copper (Cu, mg/l)**	92.16

* Soluble in distilled water ** Digest by H₂SO₄.

Vigor tests:

Included seedling vigor assessments for shoot length (cm), radical length (cm), seedling dry weight as well as laboratory tests at Seed Technology Res. Dept. (ARC) for:

Germination percentage: seeds were incubated on moist filter paper at 30C for 8 days. Normal seedlings were counted according to international rules of I.S.T.A. (1993) and expressed as germination percentage. Ten normal seedlings from each replicate were taken to measure shoot and radical length (cm) according to Krishnasamy and Seshu (1990).

$$\frac{\text{Normal seedling}}{\text{Number of seed germination}} \times 100$$

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Electrical conductivity of grains weights ($\mu\text{S cm}^{-1} \text{g}^{-1}$): four samples of 50 grains from each experimental plot were weighted and placed into flask with 250 ml of distilled water and held at 25C for 25 hour (A.O.S.A. 1983). The electrical conductivity was measured using electrical conductivity meter. The mean values of the conductivity per gram of seed weight, expressed as $\mu\text{S.cm}^{-1}\text{g}^{-1}$, were calculated by the formula of

$$\frac{\text{Conductivity (us/ cm) for each flask}}{\text{Weight (g) of grain sample}}$$

For chemical determination, plants were fine powdered, wet digestion for dry material was carried out according to Chapman and Pratt (1961). Nitrogen percentage was determined in grains by micro kjeldahl method according to A.O.A.C. (1990). The content (%) of phosphorus, potassium, crude protein and total carbohydrate in grains were estimated according to A.O.A.C. (1990). The atomic absorption spectrophotometer was used to determine Zn, Mn, Fe and Cu in the same organ according to the method described by A.O.A.C. (1990). All data were subject to statistical analysis for comparing treatments means as described by Barbara and Brain (1994).

RESULTS AND DISCUSSION

Dry Weights of Shoots and Roots.

Data in Table (3) showed that, using any rate of micronutrients mixture fertilizer as foliar feeding significantly increased dry weights of shoots or roots after 60 days from sowing. Microelements mixture at 0.3 g/l for each element was the most effective one in this respect, where dry weights in shoots and roots reached 2.13 and 1.70 (g/ plant), respectively for the first season as well as 2.21 and 1.99 (g/plant), at a respective order for the second season, comparing to the control treatment. These findings coincide with Chen et al. (2009), they found that zinc application increased root length, root surface and root tips in rice plant. Aminder and Sadana (2010) reported that manganese addition caused higher root length, root surface area, shoot dry weights and root length density. Marschner, (1986) reported that the importance of iron as a component of chlorophyll, prosthetic group of heme- and nonheme proteins.

Data in that table indicate that, grains soaked with humic acid at a rate of 100 ppm for 24 hour stimulate the action of micronutrients for producing the high significant increases in the parameters of dry matter accumulation with a pronouncing response especially with plants received humic acid plus Zn+ Mn+ Fe at a rate of 0.3g/l, where shoots and roots dry weights reached to 2.69 and 1.9 (g/plant), at a respective order for the first season and were 2.80 and 2.11 (g/plant), respectively for the second season. Likewise significant improvement in the whole plant dry weight by about 4.59 and 4.91 (g/plant). Hakan (2008) found that, humic acid have positive effect on branched roots and root hair development, so increased root length resulted in greater

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absorption of water and nutrients resulted in increased root area, permeability of root cells and biomass.

Table (3): Effect of micronutrients fertilizer and humic acid on dry weights of Shoots and roots of rice plant after 60 days from sowing (2009 and 2010).

Treatments	2009			2010		
	Dry weights (g /plant)			Dry weights (g/plant)		
	Shoots	Roots	Whole plant	Shoots	Roots	Whole plant
Control	0.52	0.36	0.88	0.54	0.40	0.94
EDTA (1)	2.13	1.70	3.83	2.21	1.99	4.20
EDTA (2)	1.59	1.16	2.75	1.65	1.19	2.84
Humic acid+ (1)	2.69	1.90	4.59	2.80	2.11	4.91
Humic acid+ (2)	1.88	1.36	3.24	1.95	1.51	3.46
L.S.D.* 5%	0.01	0.02	0.03	0.02	0.03	0.04
1%	0.02	0.03	0.05	0.04	0.04	0.06
C.V. **	10.29	11.52	9.21	11.30	11.50	9.20

*: Treatments.

** : Coefficient of variation

EDTA (1): Zn +Mn +Fe at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.

Chlorophyll Contents

The results in Table (4) show that, foliar application with micronutrients at a rate of 0.3 g/l was the best treatment, where the increases of chlorophyll a, b and carotenoids were significant, comparing to the control treatment. Alam (2006) reported that, foliar supply of micronutrients can results in increasing the photosynthetic efficiency and it is possible to modify the physiology of leaf.

Table (4): Effect of micronutrients and humic acid on chlorophyll content in Leaves of rice plant after 60 days from sowing (2009 and 2010).

Treatments	2009				2010			
	Chl. a (ug/ml)	Chl b (ug/ml)	Chl a+b (ug/ml)	Carotenoids (ug/ml)	Chl a (ug/ml)	Chl b (ug/ml)	Chl a+b (ug/ml)	Carotenoids (ug/ml)
Control	2.38	1.32	3.70	2.13	2.84	1.52	4.36	2.44
EDTA (1)	5.18	1.96	7.14	3.01	5.40	2.04	7.44	3.14
EDTA (2)	3.40	1.54	4.94	2.26	4.14	1.66	5.80	2.71
Humic acid+(1)	8.42	3.46	11.88	4.46	9.86	4.04	13.90	5.04
Humic acid+(2)	3.70	1.56	5.26	2.24	4.82	1.74	6.56	2.90
L.S.D. * 5%	0.10	0.01	0.07	0.03	0.04	0.03	0.16	0.02
1%	0.15	0.02	0.10	0.04	0.06	0.04	0.23	0.04
C.V. **	7.16	8.28	7.56	8.54	7.41	9.05	9.09	8.40

*: Treatments.

** : Coefficient of variation

EDTA (1): Zn +Mn +Fe at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.

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Moreover, exogenous application of humic acid as soaking method in combination with micronutrients mixture, achieved high significant increases in chlorophyll content. The combined treatment includes of humic acid plus 0.3 g/l of Zn +Mn +Fe was the best promotive effect on this respect. Since chlorophyll a, b and carotenoids amounted to 8.42, 3.46 and 4.46, respectively for the first season. Corresponding significant increases were 9.86, 4.04 and 5.04, respectively for the second season. Santiago *et al.* (2010) reported that humic acid increased chlorophyll content.

Yield and Yield Components

Data in Table (5) show that, yield and yield components were increased significantly with the foliar feeding with Zn +Mn +Fe, particularly at a rate of 0.3 g/l. This was true for both seasons. Since plant height (cm) and number of panical/m² reached to 79.41 and 295, respectively for the first season and 82.16 and 305, respectively for the second season. Prior treatment significantly increased panicle length (cm) and panicle weights (g) by about 19 and 2.56 at a respective order for the first season. While data in the second season, were 22 and 2.60, respectively. These results indicated the response of rice plant to the application of micronutrients and the soil under study was deficient in available Zn, Mn and Fe. The positive effect of Zinc might be due to its function as catalyst or stimulant in most of the physiological and metabolic process and metal activator of enzymes, resulting in growth and development, which ultimately gave higher yield and yield components (Singh *et al.*, 2004). The significant favorable effect of manganese may be due to the physiological role of this nutrient on enzymes synthesize and function on plant growth as well as yield and its components (EL-Hadidy *et al.*, 2000). The promotive action of ferrous may be due that ferrous involved in photosynthesis and mitochondrial respiration. It is a constituent of the enzyme catalase and peroxidase and help in the utilization of nitrogen and increased sink size (grain/ panicle) (Sarang and Sharma, 2004). These results are in agreement with those of Gurmani *et al.*, (2003) and Shaban *et al.*, (2010) on rice plants.

It can be visualized from the results in Table (5) that, the high significant response in yield and its components were obtained from soaking grains with humic acid in the presence of foliar spray with microelements for two growing seasons. The highest significant effect induced under humic acid + Zn +Mn +Fe at rate of 0.3 g/l. Since the increase in 1000- grains weight raised to 33 and 35(g) for the first and second seasons, respectively. These could be explained according to the findings of Cheng *et al.* (1998) who stated that humic acid increased the ability rate of leaves for photosynthesis process, increased the grain filling intensity, enhanced the drought resistance of plants and increased its thousand grains weights. Such treatment induced the highest significant improvement in grains and straw yield by about 2.71 and 3.85 ton/ fed., at a respective order for the first

season as well as 2.76 and 3.95 ton/fed. at the order state for the second season. The increase in rice grains and straw yield under such treatment probably resulted from increasing macro- and micronutrients availability needed for plant growth through matter decomposition (Ali, Laila and ELbording, 2009). Similar results were found by Bhattacharya *et al.*, (2003) and Verlinden *et al.*, (2009).

Table (5): Effect of micronutrients and humic acid on yield and yield components of rice plant (2009 and 2010).

Growth seasons	Treatments	Plant heights (cm)	No of Panicles /m ²	Panicles Length (cm)	Panicle weights (g/plant)	1000-weights (g)	Grains Yield (ton/fed.)	Straw Yield (ton/fed.)
2009	Control	71.23	247	15	2.3	26	1.48	2.1
	EDTA (1)	79.41	295	19	2.56	29	2.60	3.70
	EDTA (2)	72.44	262	16	2.36	27	1.67	2.42
	Humic acid +(1)	82.91	310	20	2.61	33	2.71	3.85
	Humi cacid +(2)	75.11	275	18	2.41	28	1.96	2.65
	L.S.D. * 5%	0.33	1.03	2.45	0.16	1.13	0.16	0.19
	1%	0.48	1.50	3.57	0.23	1.67	0.24	0.28
C.V **	11.02	10.18	11.41	12.43	10.92	11.44	10.25	
2010	Control	73.51	253	17	2.34	27	1.50	2.15
	EDTA (1)	82.16	305	22	2.60	31	2.66	3.80
	EDTA (2)	73.81	270	18	2.38	28	1.70	2.55
	Humic acid+ (1)	86.55	322	26	2.64	35	2.76	3.95
	Humic acid+ (2)	77.21	280	20	2.43	30	1.98	2.70
L.S.D. *	5%	0.40	3.37	5.61	0.11	0.84	0.3	0.21
	1%	0.59	4.90	8.16	0.17	1.23	0.44	0.31
C.V. **	11.22	10.56	12.55	10.22	11.49	10.35	11.44	

*: Treatments.

** : Coefficient of variation

EDTA (1): Zn+ Mn +Fe at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.

Mineral Concentrations

Macronutrients and Sodium Concentrations

The data in Table (6) show that, high salt (Na uptake) competes with the uptake of other nutrient ions especially K leading to K deficiency. Dealing with micronutrients mixture at a rate of 0.3 g/l as foliar nutrition led to significant decrease in Na concentration followed by 0.6 g/l of these nutrients in rice grains under saline conditions. Since the percentage of decrease reached to 57.6 and 60.0% for the first and second seasons, respectively, as compared with the control treatment. Such treatment increased significantly macronutrients concentrations in grains for both seasons. The nitrogen concentration rose from 1.22 to 1.56% for the first season and from 1.25 to

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1.58% for the second season at control and Zn +Mn +Fe at a rate of 0.3 g/l, at a respective order. In this respect EL-Fouly and Fawzi (1996) noted that micronutrients application led to encourage the growth of roots, which in turn to higher content of N,P and K and finally being reflected to increased the yield. These results are in accordance with those obtained by Shaban *et al.* (2010).

Furthermore, the results indicated high significant effect on those concentrations for plant received humic acid as soaking treatment plus foliar fertilizer with micronutrients mixture (Table 6). The combined treatment of humic acid + 0.3 g/l Zn +Mn +Fe was more significant effective on macronutrients concentration than other treatments. The concentrations (%) of phosphorus and potassium amounted to 0.396 and 0.74, respectively for the first season and 0.401 and 0.76 at order of state for the second season. Such treatment decreased significantly sodium concentration by about 49.15 and 51.67% for first and second seasons, respectively, as comparing with control. In this connection, Chen and Aviad, (1990) reported that, humic acid enhanced cell permeability, which in turn made for rapid entry of mineral into roots - cell and so resulted in higher uptake of nutrients. Jackson (1993) found that, humic acid can get into cellular nutrients stream and make the cellular membrane more permeable allowing the improvement of nutrients flow and cell division. These results are consistent with the results published by Salib (2002) and Salhyabama *et al.* (2004).

Table (6): Effect of micronutrients fertilizer and humic acid on macronutrients and sodium concentrations in grains of rice plant (2009 and 2010).

Treatments	2009				2010			
	N%	P%	K%	Na%	N%	P%	K%	Na%
Control	1.22	0.203	0.32	0.59	1.25	0.209	0.34	0.60
EDTA (1)	1.56	0.320	0.67	0.34	1.58	0.336	0.69	0.36
EDTA (2)	1.37	0.244	0.40	0.55	1.39	0.261	0.42	0.56
Humic acid+(1)	1.70	0.396	0.74	0.29	1.75	0.401	0.76	0.31
Humic acid+(2)	1.40	0.295	0.50	0.52	1.42	0.302	0.53	0.55
L.S.D. * 5%	0.02	0.08	0.03	0.02	0.03	0.10	0.11	0.04
1%	0.04	0.12	0.05	0.03	0.04	0.16	0.19	0.06
C.V. **	7.62	8.16	9.45	7.95	8.91	8.20	7.98	8.01

*: Treatments.

** : Coefficient of variation

EDTA (1): Zn +Mn +FE at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.

Micronutrients concentrations

Data in Table (7) show that, the increase mixture of micronutrients consist of Zn +Mn +Fe rate from 0.0 to 0.6 g/l for each element yielded in high significant increases in microelements concentrations (mg/kg) in grains. This

was true for both seasons. Where zinc and copper concentrations amounted to 19 and 15, respectively, for the first season. While in the second season data were 21 and 16, respectively. It was found from these results, that rice plant need to micronutrients to accelerate the metabolic processes during the reproductive stage which run from panicle imitation to flowering and ripening phase and from flowering to maturity (Shaban et al., 2010). These results coincide with those obtained by Mohammed (2008) who found that, the micronutrients enriched with NPK fertilizer increases the concentrations of micronutrients in rice grains.

Furthermore, the combination of humic acid with micronutrients mixture at different rates produced more significant increases in the concentrations for two growing seasons (Table 7). The highest significant effect was recorded under humic acid plus Zn +Mn +Fe at a rate of 0.6 g/l treatment. The increase in manganese and ferrous concentrations (mg/kg) rose to 54 and 452 at order of state for the first season as well as 56 and 455, respectively, for the second season. In this connection, David et al. (1994) reported that, humic acid promote seed germination, increase nutrients uptake and stimulate plant growth by its effect on ion transfer at the root level, by activating the oxidation reduction state of the plant growth medium and so increased absorption of micronutrients. Similar results were obtained by Bohme and Thilua (1997); they reported that humic acid enhanced micro-nutrients of the plant organs.

Table (7): Effect of micronutrients fertilizer and humic acid on micronutrients concentrations (mg/kg) in grains of rice plant (2009 and 2010).

Treatments	2009				2010			
	Zn	Cu	Mn	Fe	Zn	Cu	Mn	Fe
Control	12	11	38	271	13	12	39	273
EDTA (1)	18	13	44	371	20	14	46	373
EDTA (2)	19	15	45	381	21	16	47	384
Humic acid (1)	30	16	53	441	32	17	55	443
Humic acid (2)	31	17	54	452	33	18	56	455
L.S.D. & 5%	1.69	1.84	2.66	3.16	2.06	2.51	1.68	4.46
1%	2.45	2.75	3.87	4.67	3.00	3.89	2.45	6.49
C.V. **	7.62	7.96	8.53	7.28	8.95	7.60	9.96	7.61

*: Treatments.

** : Coefficient of variation

EDTA (1): Zn+ Mn +Fe at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.

Crude protein and total carbohydrates percent

The results in Table (8) reveal that, spraying rice plants two times with Zn +Mn +Fe at a rate of 0.3 g/l significantly increased the crude protein in grains by about 9.75 and 9.88 % for the first and second seasons followed by 0.6 g/l

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of those nutrients. Similar trend was found for total carbohydrate percentage in grains. The increase in carbohydrate percentage may have been due to the effect of those nutrients on chlorophyll concentrations, activation of carboxylation and dehydrogenase enzymes of CO₂- fixation (Kumar and Prasad, 1986). However, the increase in crude protein percent was ascribed to effect on RNA synthesis which in turn plays an important role in protein biosynthesis, activity of nitrate reductase in leaves (Kvyatkovski, 1988).

Table (8): Effect of micronutrients fertilizer and humic acid on crude protein and total carbohydrates percent in grains of rice plant (2009 and 2010).

Treatments	2009		2010	
	Crude protein percent	Total carbohydrates percent	Crude protein percent	Total carbohydrates percent
Control	7.62	75.3	7.81	75.6
EDTA (1)	9.75	80.3	9.88	80.8
EDTA (2)	8.56	77.3	8.69	77.5
Humic acid + (1)	10.62	81.2	10.74	81.7
Humic acid + (2)	8.75	78.3	8.88	78.6
L.S.D. *				
5%	0.11	0.17	0.33	0.12
1%	0.16	0.24	0.48	0.18
C.V. **	7.64	8.11	7.92	8.10

*: Treatments.

**: Coefficient of variation

EDTA (1): Zn +Mn +Fe at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.

Moreover, the application of humic acid as soaking treatment in presence of Zn +Mn +Fe mixture, more significant improvement on studied parameters were obtained. The combined treatment of humic acid + those nutrients at a rate of 0.3 g/l produced the highly significant effect on studied characters. Since total carbohydrates percent reached to 81.2 and 81.7 for first and second seasons, respectively. This may be due explained the role of humic acid in increasing the concentration of messenger ribonucleic acid, increases in enzymes synthesis and an increase in the crude protein and carbohydrates as found by Atak *et al.* (2005). Similar results were obtained by Ali, Laila and Elbording (2009).

Seedling vigour test.

Data recorded in Table (9) emphasized that foliar addition with micronutrients exhibited significant effect on germination percentage, speed of germination, shoot and radical length, seedling dry weight and electrical conductivity (EC) for the two growing seasons, surpassed the control

treatment. Foliar supply 0.3 g/l with those nutrients was the best effective treatment on the above mentioned characters. Since the increase in germination percentage and shoot length reached to 90 % and 6.0 (cm), in the order stated for the first season and 90% and 6.2 (cm), respectively for the second season. The improvement in those parameters may be due to the importance of micronutrients in enhancing accumulation of assimilate in the grain (during grain filling stage) and produced more viable and vigorous seedling, because they supplying the young seedling to developed a large root system (Marschner, 1995).

Table (9): Effect of micronutrients fertilizer and humic acid on seedling vigor test in rice plant (2009 and 2010).

Growing seasons	Treatments	Germination %	Shoot length (cm)	Radical length (cm)	Dry weights of seeds (g)	EC
2009	Control	78	5.0	3.3	0.59	87.1
	EDTA (1)	90	6.0	4.2	0.71	91.4
	EDTA (2)	82	5.0	3.0	0.67	95.05
	Humic acid+(1)	94	6.8	3.8	0.77	82.81
	Humic acid+(2)	82	5.5	3.2	0.65	91.8
	L.S.D. * 5%	0.84	0.15	0.2	0.03	0.08
	1%	1.23	0.21	0.3	0.04	0.11
C.V. **	10.52	11.37	12.22	11.98	10.05	
2010	Control	80	5.2	3.5	0.60	89.7
	EDTA (1)	90	6.2	4.8	0.73	92.6
	EDTA (2)	84	5.3	3.2	0.67	96.7
	Humic acid + (1)	96	7.1	4.0	0.8	88.6
	Humic acid + (2)	84	5.6	3.8	0.69	92.16
	L.S.D. * 5%	1.85	0.16	0.23	0.05	0.06
	1%	2.69	0.25	0.34	0.07	0.09
C.V. **	12.33	11.52	13.17	10.92	12.04	

*: Treatments.

**: Coefficient of variation

EDTA (1): Zn +Mn +Fe at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.

It could be noted that, application of humic acid as soaking treatment increased the efficiency of micronutrients and thereby the most promotive response of studied parameters for the two seasons under study. The highly significant increases were only appreciable at humic acid plus foliar addition with mixture contained Zn +Mn +Fe at a rate of 0.3 g/l for each element. Where radical length (cm) and seedling dry weights being 3.8 and 0.77 at a respective order for the first season. Also, the results were 4.0 and 0.8 (cm),

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respectively for the second season. Such treatment decreased significantly EC (82.8 and 88.60 for the first and second seasons, respectively, as compared with that of the control (87.10 and 89.70 for first and second seasons at a respective order). These results explained that the application of humic acid may increase the water absorption and germination rates in rice (Micheal, 2001). These results are in agreement with those of Ali, Laila and ELbording (2009); they reported that, soaking significantly improved germination and the highest performance in all the parameters. They also showed that optimal level of soaking is thought to have enhanced effect on germination and growth probably due to hydrolysis of complexes into simple sugars that are readily utilized in the synthesis of auxins and proteins. The auxins produced to help to soften cell walls to facilitate growth and the proteins readily utilized in the production of new tissues.

CONCLUSION

The main chemical properties of the used soil were salinity, poor in the nutrients-bearing micronutrients and organic acid which are a storehouse for the essential plants nutrition. Micronutrients have been found to a profound effect on not only the biological activity and soil structure but also on the plant life. This due to their stimulate the biosynthesis of endogenous auxin, increase the photosynthetic activity and improve the nutrients concentrations and uptake, thereby increasing rice growth and productivity. We can not neglected the importance of humic acid which enriched the activity of micronutrients, where the efficiency of combination more than using microelements individually, especially for such poor soil to obtain better growth accompanied by higher yield which is the main point for our research.

REFERENCES

- Alam, S.M., R. Ansari, S.M. Mugtaba and Sh. Aisha (2001). Saline lands and rice. N I A Tantojam. Pakistan. Apri. 23-29.
- Alam, S.M. (2006). Foliar spray for plant growth. Dawan internet edition. Zilhaj, 03: 1427.
- Ali, Laila, K.M. and M.M. ELbording (2009). Response of wheat plants to potassium humate application. J. of Applied Sci. Res. 5 (9): 1202-1209.
- Aminder, K. and U.S. Sadana (2010). Nitrogen source and manganese application effects on manganese dynamic in the rhizosphere of wheat cultivars grown on manganese-deficient soil. J.of Plant Nutri. 33 (6), 832-845.
- Anonymous, A.M. (1985). International rules for seed testing. Seed Sci Tech., 13: 299-320.
- A.O.A.C. (1990). Association of Official Methods of Analytical Chemists, Official Methods of Analysis 5th edn. Washington D.C., USA.

- A.O.S.A. (1983). Association of Official Seed Analysis Vigor Testing Hand Book East Lasting 88 P (contribution, 32).
- Atak, M.K., K.M. Khawar, C.Y. Cifici and S. Ozean (2005). Effect of pre-sowing treatment with zinc and foliar spray of humic acids on yield of common bean (*Phaseolus Vulgaris L.*). Int. J.of Agric. and Biology 7: 875-878.
- Barbara, F.R. and L.J. Brain (1994). Minitab Hand Book. Duxbury press. An Imprint of Wad Sworth Publish. Comp. Belonont California.
- Bhattacharya, S.P., S. Sarkar, A.J. Karmakar, P.S. Bera and L. Mondal (2003). Effect of humic acid (Earth) on the growth and yield of transplanted summer rice. Environment and Ecology 21 (3): 680-633.
- Black, C.A. (1965). Methods of Soil Analysis. Amer. Society of Agronomy Madison. Wascomsin USA.
- Bohme, M. and H. Thilua (1997). Influence of mineral and organic treatments in the rhizosphere on the growth of tomato plants Acta Hortic. 450: 161-168.
- Brunetti, G., C. Plaza and N. Seneri (2005). Oliva pomace amendment in Mediterranean conditions. Effect on soil and humic acid properties and wheat (*Triticum turgidum L.*) yield. J. Agric. Food Chem., 53 (17): 6730-6737.
- Chapman, H.O. and P.E. Pratt (1961). Methods of Analysis of Soil and Plants and Water. Univ. of California Agric. Sci. Preiced Publication 4034, P 50.
- Chen, Y. and T. Aviad (1990). Effect of humic substances on plant 161-186. In p. Maccarthy, C.E.; R.L. Clapp and P.R. Bloom (Eds). Humic substances in soil and crop sciences. Soil Sci Am. Madison, USA.
- Chen, W.R., Z.L. He, X.E. Yang and Y. Feng (2009). Zinc efficiency in correlation with root morphology, ultra structure and Ant oxidative enzymes in rice. J. Plant Nutri Vol. 32: 287-305.
- Cheng, F.J., D.Q. Yang and Q.S. Wu (1998). Physiological effect of humic acid on drought resistance of wheat. Chinese J. of Applied Ecology. 4-6: 363-367.
- David, P.P., P.V. Nelson and D.C. Sanders (1994). Humic acid improves growth of tomato seedling in solution culture. J. Plant. Nutr. 17: 173-184.
- EL-Fouly, M. and A.A. Fawzi (1996). Fertilizer Use in the Near East Agriculture Over View. Regional Workshop on Fertilizer Use for Sustainable Agriculture. Amman. Jordan 4-6.
- EL- Hadidy, E.M., A.A. Taha, M.M. EL-Zeky and M.H. EL-Kholy (2000). Soil salinity and fertilization influence on Fe and Mn uptake by soil seed rape. J. Agric. Sci. Mansoura Univ., 25 (2): 1101-1108.
- Fataftah, A.K., D.S. Watia, B.C. Ains and S.I. Katab (2001). Comparative evolution of known liquid humic analysis method. Arctoch Inc. Chantilly, V.A. 20165. U.S.A.
- Gurmani, A.R., M. Qasim Khan, A. Bakhsh and A.H. Gurmani (2003). Effect of various micronutrients (Zn, Cu, Fe, Mn) on the yield and field components of Paddy. Sarhad J. Agric. 19 (2): 221-224.

Effect of grains soaking with humic acid and micronutrients.....

- Hakan, U. (2008). Tarla Bitkileri Tarfinda Humik Asit Uygulamasi. KSU Fen ve Mühendislik Dergisi, 11 (2). pp 119-127.
- Imtiaz, B.J., K.H. Alloway, S.H. Shah, M.Y. Siddiqui, M.A. Memon and P. Khan (2003). Zinc nutrition of wheat. II- Interaction of zinc with other trace elements. Asian J. of Plant Science. @ (2): 156-160.
- I.S.T.A. (1993). International Rules for Seed Testing Association. Seed Science and Technology 13: 307-355.
- Jackson, W.R. (1993). Organic Soil Conditioning (Humic Fulvic and Microbial. Balance) isbn # O- 9635741-0-8. X.F. Ali, Laila and Elbording (2009).
- Krishnasamy, V. and D.V. Seshu (1990). Phosphine Funigation influence on rice seed germination and vigor. Crop Sci., 30: 28-85.
- Kumar, V. and U.K. Prasad (1986). Response of wheat to different rates of zinc and nitrogen under varying levels of soil moisture. Indian. J. Agron. 31 (4): 405-406.
- Kvyatkovski, A.F. (1988). The effect of trace elements on nitrate reduction activity and chlorophyll content in maize leaves under irrigation. Fiziol Biochem. Kult. Past. 20 (1): 39-42.
- Marschner, H. (1986). Function of Mineral Nutrition : Micronutrients. In Brace Jovanovich Publisher. Part 1, pp. 269.
- Marschner, H. (1995). Mineral Nutrition of Higher Plants. Academic Press. New York.
- Metzner, H., H. Rau and H. Senger (1965). Untersuchungen zur synchronisierbarkeit einzelner pigmenten angel. Mutanten Von Chlorella. Planta 65: 186.
- Micheal, K. (2001). Oxidized Lignites and Extracts from Oxidized Lignites in Agriculture. Soil Biol.& Biochem., 32: 415-419.
- Mohammed, J.M. (2008). The effect of micronutrients in ensuring efficiency use of macronutrients. Turk. J. Agric. for Tarbiat. Madares Univ., (32): 215-220.
- Sairam, P.K. and A. Tuagi (2004). Physiological and molecular biology of saline stress tolerance in plants. Current Sci. *6 (3): 407-421.
- Salhyabama, K., G. Selvakumari, R. Naleson and P. Singaram (2004). Humic acid and fertilizer effect on nutrition of rice on Aefisol and Inceptisol. Madras Agric. J. 91 (1-3): 100-108.
- Salib, M.M. (2002). The integrated effect of humic acid and micronutrients in combination with effective micro-organisms on wheat and peanut grown on sandy soils. Zagazig J. Agric. Res., Vol.29 No. (6): 2033-2050.
- Santiago, A., A. Exposito; J.M. Quintero; E. Garmano and A. Delhado (2010). Adverse effect of humic acid from different origin on lupin as related to iron source. J. of Plant Nutri. Vol. 33 Issue 2: 143-156.
- Sarang, S.R. and H.C. Sharma (2004). Influence of mode of iron application and growth regulators on development, yield attributes and yield of upland rice (*Oryza sativa*). Indian J. of Agric Sci. 74 (12): 637-642.

- Shaban, Kh.A., Manal A. Attia and Awatef A. Mahmoud (2010). Response of rice plant grown on newly reclaimed saline soil to mixture of chelated Fe, Mn and Zn applied by different methods and rates. *J. of Soil Sci. and Agric. Engin.* 1 (2): 123-134.
- Singh, V., S.P. Rajvir and K.L. Totawat (2004). Effect of phosphorus and zinc nutrition of wheat (*Triticum aestivum*) in soil of sub-humid southern plains of Rajasthan. *Ind. J. of Agron.* 49 (1): 46-48.
- Verlinden, G., B. Pycke, J. Mertens and J. Bries (2009). Application of humic substances results in consistent increases in crop yield and nutrients uptake. *J. of Plant Nutri.* 32 (9), 1407- 1426.

تأثير نقع الحبوب في حامض الهيوميك مع الرش بالعناصر الصغرى على جودة وإنتاجية الأرز تحت ظروف الأراضي الملحية

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الملخص العربي

اجريت تجربتان حقليتان خلال الموسم الصيفي ٢٠٠٩ - ٢٠١٠ في منطقة سهل الحسينية بمحافظة الشرقية لدراسة مدي استجابة نبات الأرز صنف جيزة ١٧٨ لثلاث مستويات من مخلوط العناصر الصغرى المكون من زنك + منجنيز + حديد في صورة مخلبية (صفر، ٣ ، ٦ ، جرام/ لتر). وتم استخدام هذه العناصر في محاليل الرش إما في صورة منفردة أو متحدين مع حامض الهيوميك بطريق النقع لحبوب نبات الأرز بمعدل ١٠٠ مليجرام في اللتر مدة ٢٤ ساعة قبل الزراعة. تم رش النباتات مرتين الرشة الأولى بعد ٤٥ يوم والثانية بعد ٧٠ يوم من تاريخ الزراعة وذلك بمعدل ٤٠٠ لتر/ فدان. زنك مخلبي ١٤% زنك و منجنيز مخلبي ١٢% منجنيز وحديد مخلبي ١٣% حديد. أكدت النتائج المتحصل عليها فاعلية الرش بمخلوط العناصر الصغرى بمعدل ٣، جرام/ لتر لكل عنصر وتفوقه علي نظيرة ٦، جرام / لتر من كل عنصر في إحداث إستجابة معنوية في الأوزان الجافة للمجموع الخضري والجذور و كانت الجذور هي أكثر أعضاء النبات تأثراً بهذة المعاملة وأيضاً زيادة معنوية لمحتوي الأوراق من الكلوروفيل أ وب والكاروتينات وذلك بعد ٦٠ يوم من تاريخ الزراعة في موسمي الزراعة. كما وجد زيادة إحصائية لمحصول الأرز ومفرداته حيث بلغت الزيادة النسبية لمحصول الحبوب 75.68 و 77.33 % في كل من الموسم الأول والثاني للزراعة. وأيضاً زيادة إحصائية لمحتوي الحبوب من العناصر الكبرى (نيتروجين - فوسفور - بوتاسيوم) والعناصر الصغرى (حديد - زنك - منجنيز - نحاس). والبروتين الخام والكربوهيدرات الكلية في حبوب الأرز في كل من موسمي الزراعة. كما إرتفعت نسبة الإنبات إحصائياً بالإضافة إلي طول الريشة والجذير

والوزن الجاف للبادرة والتوصيل الكهربائي لحبوب نبات الأرز صنف جيزة ١٧٨ في كل من موسمي الزراعة.

وسجلت النتائج أيضا أن إتحاد حامض الهيوميك مع مخلوط العناصر الصغرى بمعدل ٣، جرام/ لتر كان الأكثر تأثيرا في إظهار النتائج المرجوة علي كل التقديرات السابقة التي أجريت في هذه الدراسة لصنف جيزة ١٧٨ في موسمي الزراعة.

Table (10): Effect of micronutrients fertilizer and humic acid on EC, pH, macronutrient and micronutrients availability in experimental soil (2009 and 2010).

Treatments	2009								
	EC	pH	Available macronutrients (mg/kg)			Available micronutrients (mg/kg)			
			N	P	K	Zn	Mn	Fe	Cu
Control	15.29	8.24	59	5.98	200	0.92	2.63	2.96	0.089
EDTA (1)	14.31	8.16	74	6.41	208	1.01	3.14	3.28	0.095
EDTA (2)	14.89	8.22	66	6.13	203	1.04	3.41	3.36	0.094
Humic acid+(1)	13.59	8.14	81	6.55	210	1.05	3.49	3.54	0.095
Humic acid+(2)	14.74	8.18	71	6.21	206	1.09	3.62	3.66	0.098
L.S.D. * 5%	0.014	0.016	1.79	0.017	2.06	0.05	0.017	0.15	0.002
1%	0.02	0.024	2.60	0.024	3.00	0.07	0.025	0.2	0.003
	2010								
C.V. **	8.16	7.11	8.42	9.53	8.23	9.26	9.17	7.66	9.16
Control	13.42	8.86	63	6.05	206	0.99	2.71	3.04	0.093
EDTA (1)	11.24	8.06	79	7.49	216	1.05	3.34	3.48	0.12
EDTA (2)	12.04	8.13	70	7.18	211	1.10	3.51	3.42	0.10
Humic acid +(1)	10.48	8.04	85	7.62	219	1.09	3.81	3.81	0.11
Humic acid +(2)	11.89	8.11	77	7.29	214	1.13	3.92	3.90	0.12
L.S.D. * 5%	0.09	0.017	1.98	0.002	1.68	0.06	0.018	0.02	0.017
1%	0.13	0.025	2.87	0.003	2.45	0.08	0.026	0.029	0.024
C.V. **	8.33	9.11	7.91	9.41	8.63	9.57	7.69	8.27	8.52

*: Treatments.

** : Coefficient of variation

EDTA (1): Zn +Mn +Fe at a rate of 0.3 g/l for each nutrient.

EDTA (2): Zn +Mn +Fe at a rate of 0.6 g/l for each nutrient.