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RESPONSE OF SOME RICE VARIETIES TO IRRIGATION WITH BRACKISH WATER AND ORGANIC FARMING

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ABSTRACT: *Two experiments were carried out using lysimeter technique during the two summer seasons of 2005 and 2006 in the Rice Research and Training Center, Kafr El-Sheikh, to investigate the response of three rice varieties (IR29, Sakha 102 and Giza178) to the irrigation with brackish water at levels (0 , 4000 and 8000 ppm) and organic matter at 0.1gm/L. Results indicated that, The treated rice plants with brackish water at all levels significantly decreased plant height, No. of tillers, leaf area, shoot fresh and dry weights, No. of panicles/plant, No. of spikelets/panicle, No. of total grains/panicle, % fertility, straw yield, 1000-grains weight and harvest index, photosynthetic pigments, nucleic acids concentration and the total and relative water content, transpiration rate, the grain content of amylose and protein as well as the concentrations of N, P, K and Ca, while the heading date, No. of unfilled grains, proline, leaf water deficit, Na percentage and Na/K ratio were increased compared with control. Application the organic matter resulted in increasing all vegetative growth parameters under study, physiological and biochemical parameters as well as yield compared with control. while decreased No. of unfilled grains, LWD, proline concentration Na and Na/K ratio. Under salinity levels, the treated plants with organic matter improved all the previous characteristics compared with those grown under only brackish water and enhanced the growth and yield of all varieties and Giza 178 gave the highest increase in this respect. Plant genome study indicated that, there was no linkage between the two SSR markers (RM223 and RM315) linked to salinity and the salt tolerance in the varieties while, RM527 generated a clear level of polymorphism among the varieties but it wasn't linked to salinity tolerance. This means that, there is deference in molecular between the varieties under this study.*

Key Words *brackish water, organic matter, rice varieties, lysimeter, biochemical, Plant genome*

INTRODUCTION

Cereal crops are the most important sources of food as cereals; in particular rice (*Oryza sativa* L.) is the major food for more than one third of the world's population (Sedik *et al.*, 1998). It belongs to family Gramineae, its

crop plays a significant role in Egypt's strategy for sustaining the food self-sufficiency and for increasing the export. Further increase in rice production through increased yield per unit area is needed. This can be achieved through improving productivity of saline area which occupies about 25% from rice area in Egypt.

Salinity is one of the major abiotic stresses that adversely affect crop productivity and quality. About 20% of irrigated agricultural land is adversely affected by salinity (Flowers and Yeo, 1995). The problem of soil salinity is further increasing because of the use of poor quality water for irrigation and poor drainage. Adverse effects of salinity on plant growth may be due to ion cytotoxicity and osmotic stress. Ion cytotoxicity is caused by replacement of K^+ by Na^+ in biochemical reactions and conformational changes and loss of function of proteins as Na^+ and Cl ions penetrate the hydration shells and interfere with non covalent interactions between their amino acids. Metabolic imbalances caused by ionic toxicity, osmotic stress and nutritional deficiency under salinity may also lead to oxidative stress (Zhu. 2002). Salt stress is currently one of the major problems facing rice production worldwide. Improving salinity tolerance in rice could enhance productivity in salt affected areas and help in further expansion of rice production in salt affected areas that are currently not in use. Rice is rated as an especially salt-sensitive crop (Shannon *et al.*, 1998). The response of rice to salinity varies with growth stage. In the most commonly cultivated rice cultivars, young seedlings were very sensitive to salinity (Lutts *et al.*, 1995). Yield components related to final grain yield were also severely affected by salinity. It also delayed the emergence of panicle and flowering and decreased seed set through reducing pollen viability (Khatun and Flowers, 1995). In contrast, rice was more salt-tolerant at germination than at other stages.

Recent researches showed that organic matter can be used as a growth regulator to regulate hormone level, improve plant growth and enhance stress tolerance (Piccolo *et al*, 1992). Important soil constituent consisting of a range of organic components such as humic substances, organic acids of low and high molecular weight, carbohydrates, protein, peptides, amino acids, lipids, waxes, polycyclic aromatic hydrocarbons and lignin fragment (Stevenson and Ardakani, 1972). The most stable organic components in soils are humic substances: these can be divided into humic acids and fulvic acids (Stevenson, 1991). In this study, we used organic matter as a source of essential nutrients for plants as well as for the improvement of soil productivity as an effective agent for solving salinity problem.

The objective of this investigation was to study the morphological, physiological and biochemical characteristics and plant genome of rice plants grown under different salinity levels in response to organic farming condition (organic matter) with aim increase plant salinity tolerance and avoid plant damage.

MATERIALS AND METHODS

The present investigation was conducted using lysimeter technique (salinity controlled conditions). It is concrete beds filled with sand and gravel soil to 100 cm depth in three layers: 60 cm clay at surface, 20 cm sand at the middle and 20 cm gravel at the bottom (Fig. 1) at Rice Research and Training Center (RRTC), Kafr El-Sheikh, during the two summer seasons of 2005 and 2006 to study the effect of organic matter addition, on vegetative growth, yield, some physiological and biochemical characteristics as well as the plant genome on three rice varieties (*Oryza sativa* L.) namely: IR29, Sakha 102 and Giza178 (obtained from (RRTC)) grown under brackish water irrigation. The organic matter was in a powder shape and consists of humic acid 60%, fulvic acid 39% and urea 1%, obtained from Central Lab. of Organic Agriculture, Agricultural Research Center.

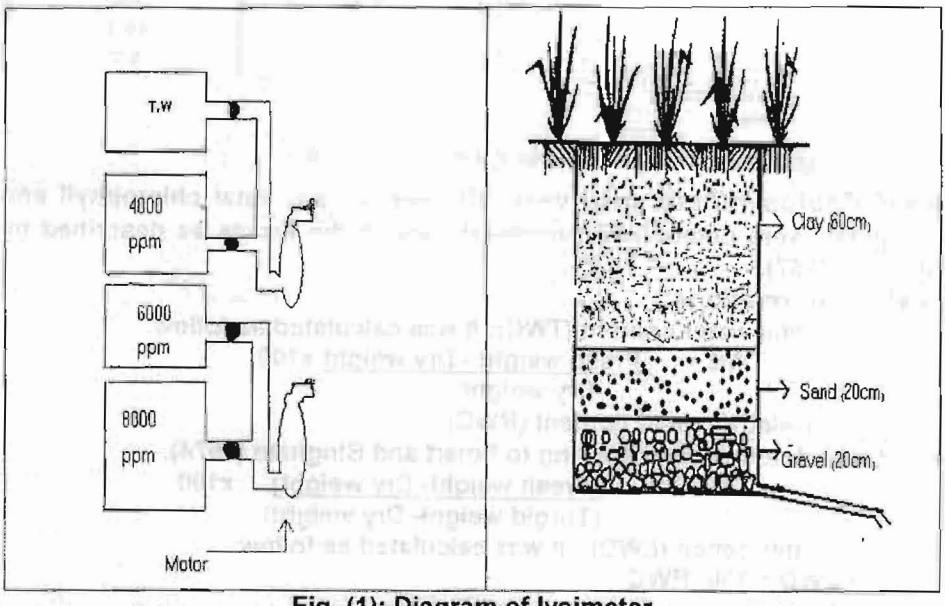

Fig. (1): Diagram of lysimeter

The physical and chemical analyses of experiment soil were presented in Table (1) according to the method described by Chapman and Pratt (1961). The plots were treated with brackish water at three levels; 0, 4000 and 8000 ppm by applying NaCl and $CaCl_2$ at the ratio of 2:1, respectively (El-Mowafy, 1994), beside to the control (Tap water) and organic matter at 0.1gm to every one liter brackish water after 15 days from transplanting untill harvest. The experiments were carried out using split-split plot design with three replicates.

One sample was taken at the heading stage from each treatment and the following parameters were recorded:-

1. Vegetative growth characteristics:

Plant height (cm), No. of tillers, Leaf area (cm^2), shoot fresh and dry weights (g).

Table (1): Chemical analysis of experiment soil Lysimeter (0-30 cm)

Chemical analysis	Values	Soluble ions (meq/L)	Values
pH	7.98	Ca^{++}	8.50
ECe (dS/m)	2.30	Mg^{++}	1.70
OM (%)	1.64	Na^{+}	12.60
		K^{+}	0.20
		CO	0.01
		HCO	4.01
		Cl	12.96
		SO_4	6.03

ECe = Electrical conductivity

OM = Organic matter

2. Physiological and biochemical characteristics:-

- *Leaf Photosynthetic pigments*: Chlorophyll a,b, total chlorophyll and carotenoids were determined calorimetrically in the leaves as described by Wettstein (1957).

-*Leaf Water relations*

Total water content (TWC): It was calculated as follow:

$$TWC = \frac{\text{Fresh weight - Dry weight}}{\text{Dry weight}} \times 100$$

Relative water content (RWC):

It was determined according to Smart and Bingham (1974).

$$RWC = \frac{\text{(Fresh weight- Dry weight)}}{\text{(Turgid weight- Dry weight)}} \times 100$$

Leaf water deficit (LWD):- It was calculated as follow:

$$LWD = 100 - RWC$$

Transpiration rate (TR, $mg/cm^2/h$): It was determined by weighting method according to Kreeb (1990).

- *Proline concentrationin shoot* (µmol/g F.W.): It was measured according to Bates *et al.* (1973).

- *Nucleic acids concentration* in shoot (µg/ml): They were estimated according to Charry (1973).

-*Mineral contents in shoot (%):* They were determined in the dry ashing plant material as follow: Nitrogen was determined by microkjeldahl according to the method described by A.O.A.C. (1985), phosphorus was determined by Ascorbic acid method using the Colorimetric method that described by Murphy and Riley (1962), Potassium, Sodium and Calcium were determined

by flame photometer as described by Chapman and Pratt (1961) and Na/K ratio.

3. Yield characteristics:

Heading date, number of panicles / plant, number of spikelets / panicle, number of total grains / panicle, (%) fertility, Straw yield (g), 1000-grains weight (g) and Harvest index (%).

Chemical components of grain (%): Amylose was estimated according to Juliano (1971) and crude protein of the tested samples was calculated by multiplying total nitrogen by the factor (5.95) as described by A.O.A.C. (1985).

N, P, K, Na and Ca in grains were analyzed using the same above mentioned methods.

4. Plant genome:

Microsatellite markers (SSR):

DNA isolation and quantification:

DNA of the tested genotypes was isolated using CTAB (Cetyl-tetramethyl ammonium bromide) method according to Murray and Thompson (1980).

4.1. SSR protocol:

Three simple sequence repeats (SSR) primers were used in this study; two of them were RM 223 and RM 315, which are linked to salt tolerance in rice. The other primer was RM 527 and this is unlinked to salinity. The primers sequences are:

Primers	Forward sequence	Reverse sequence
RM 223	GAGTGAGCTTGGGCTGAAAC	GAAGGCAAGTCTTGGCACTG
RM 315	GAGGTACTTCCTCCGTTTCAC	AGTCAGCTCACTGTGCAGTG
RM 527	GGCTCGATCTAGAAAATCCG	TTGCACAGGTTGCGATAGAG

PCR reactions were carried out in 10 μl volume containing:

Genomic DNA (15 ng/ μl)	1.00 μl
H_2O	4.74 μl
10 X PCR buffer(10 Mm Tris, pH 8, 50 mM KCl and 50 mM ammonium sulfate)	1.00 μl
$MgCl_2$ (25 mM)	0.80 μl
dNTPs (1mM)	0.40 μl
Taq DNA polymerase (5 U/ μl)	0.06 μl
Forward SSR primer (30 ng/ μl)	1.0 μl
Reverse SSR primer (30 ng/ μl)	1.0 μl
Total	10.0 μl

Using this profile: initial amplification at 94°C for 5 min, 35 cycles of amplification under the following parameters; template denaturation at 94°C for 1 min, primer annealing at 55°C for 1 min and primer extension at 72°C for 2 min. by the end of the 35th cycle, final extension at 72°C for 7 min was given, followed by storage at 4°C forever. PCR thermocycler machines from Biometra and Applied Bio systems were used.

The obtained data in the second season were in line with the findings at the first one, so data of the first season were presented.

RESULTS AND DISCUSSION

1. Vegetative growth characteristics:

Data recorded in Table (2) show that, there was a remarkable gradual decrease in plant height, No. of tillers, leaf area, fresh and dry weights with increasing the salt concentration.

Table (2): Effect of brackish water, organic matter, varieties and their interactions on vegetative growth characteristics of rice during 2005 season.

Treatments \ Characteristics			Plant height (cm)	No. of tillers	Leaf area (cm^2)/plant	Shoot fresh weight (g)/plant	Shoot dry weight (g)/plant
Salinity (ppm)		CONTROL	90.44	19.67	438.69	78.28	19.23
		4000	83.83	16.83	376.15	70.48	16.64
		8000	69.17	7.22	279.47	46.64	11.17
Organic matter		-OM	77.89	13.93	348.59	60.98	14.66
		+OM	84.41	15.22	380.94	69.28	16.71
Varieties		IR29	78.17	10.94	299.16	46.25	13.72
		Sak.102	84.61	13.44	354.84	61.22	14.97
		Giza178	80.67	19.33	440.31	87.92	18.36
-OM	CONTROL	IR29	87.00	14.00	355.80	59.64	15.65
		Sak.102	90.33	17.33	435.19	67.98	17.46
		Giza178	88.33	24.67	478.57	92.52	20.41
	4000	IR29	80.33	11.33	326.84	48.10	13.47
		Sak.102	85.00	16.00	356.74	63.72	15.73
		Giza178	81.00	21.00	429.20	88.91	18.63
	8000	IR29	61.67	4.00	202.67	23.65	8.10
		Sak.102	64.67	6.00	251.29	39.52	9.39
		Giza178	62.67	11.00	330.96	64.75	13.06
+OM	CONTROL	IR29	88.33	17.33	382.39	65.11	19.43
		Sak.102	98.00	18.67	464.32	77.61	20.08
		Giza178	90.67	26.00	515.87	106.80	22.33
	4000	IR29	81.33	14.33	318.32	56.71	15.93
		Sak.102	90.33	16.00	376.25	68.31	16.63
		Giza178	85.00	22.33	479.45	97.11	19.46
	8000	IR29	70.33	4.67	208.91	24.31	9.71
		Sak.102	79.33	6.67	275.14	50.20	10.54
		Giza178	76.33	11.00	407.82	77.40	16.24
$LSD_{0.05}$	Salinity		1.652	0.630	0.006	0.014	0.015
	OM		0.883	0.488	0.006	0.009	0.008
	Var.		0.835	0.778	0.006	0.014	0.011
	OM X Var.		1.215	0.978	0.009	0.018	0.014
	Sal. X Var.		N.S.	1.186	0.001	0.022	0.019
	OM X Sal.		1.709	0.744	0.011	0.016	0.015
	OM X Sal. X Var.		N.S.	1.680	0.016	0.031	0.025

The maximum reduction was obtained by 8000 ppm, as compared with the plants under control condition, the best results was obtained by Giza 178. The reductions in growth of rice plants under the salt stress conditions are probably attributed to increasing the osmotic pressure of the soil solution to a point which retarded the intake of water (Mengel and Kirkby, 1987), resulting in water stress in the plant and decreasing cell division, cell enlargement and the intensity of photosynthesis (Nieman, 1965) and the decline in the nucleic acids content (Sheoran and Grag, 1978). Similar results were obtained by Demiraal and Turkan (2005) and Djanaguiraman *et al.* (2006) on rice. Addition of organic matter significantly increased all previous characteristics if compared with untreated one. The increase under organic matter treatment may be due to its promotive effect on cell division and cell elongation, stimulation and balancing cells, creating optimum growth (Poapst & Schnitzer, 1971). These results were in harmony with those obtained by Adani *et al.* (1998) on tomatoes and Karr (2001) on many plants.

2. Yield characteristics

Data in Table (3) illustrate that, salinity at all levels delayed heading and decreased No. of panicles/plant , No. of spikelets/ panicle, No. of total grains/ panicle, (%) fertility, straw yield (g), total biomass (g), 1000-grains weight (g) and harvest index (%) compard with control. The best results was obtained by Giza 178, while IR29 gave the worst one. This reduction was increased with increasing salinity levels, the highest reduction was obtained by 8000 ppm, this reduction might result from the loss of photosynthetic capacity due to the effects of salinity on leaf development or longevity effects on panicle development, reduced production of assimilates, ability to utilize photosynthates for growth, and/or an increased utilization of photosynthates in respiration (Wignarajah, 1990). These results are in line with those obtained by Yousaf *et al.* (2004) and Natarajan *et al.* (2005a) on rice. Under salt stress conditions, heading date was earlier in plants treated with organic matter than untreated and No. of panicles/piant, No. of spikelets/ panicle, No. of total grains/ panicle, (%) fertility, straw yield (g), total biomass (g), 1000-grains weight (g) and harvest index (%) significantly increased in plants treated with organic matter, while No. of unfilled grains/ panicle significantly decreased compared with control. These results are probably attributed to vital activity of cells, changing the pattern of the metabolism of carbohydrates, resulting in an accumulation of soluble sugars which increase the pressure of osmosis inside the cell wall (Kononova, 1966). These results were previously observed by Sangakkara *et al.* (2005) and Nozoe *et al.* (2006) on rice.

Table (3): Effect of brackish water, organic matter, varieties and their interactions on yield characteristics of rice during 2005 season.

Treatments \ Characteristics		Heading date	No. of panicles/ plant	No. of spikelets/ panicle	No. of total grains /panicle	(%) Fertility	Straw yield (g)/plant	1000-grains weight (g)	Harvest Index (%)
Salinity (ppm)	CONTROL	86.33	14.22	9.17	135.3	87.43	7.97	20.74	47.69
	4000	89.83	7.72	7.89	115.2	77.49	6.82	17.97	48.24
	8000	95.11	4.22	5.28	71.7	47.01	3.16	13.41	32.25
Organic matter	-OM	88.96	7.96	7.07	105.4	67.18	5.68	16.37	42.26
	+OM	91.89	9.48	7.81	109.4	74.10	6.29	18.37	43.19
Varieties	IR29	86.11	7.56	6.17	119.3	59.43	5.12	14.48	42.19
	Sak.102	91.28	8.83	7.56	97.0	64.21	5.56	16.82	42.97
	Giza178	93.89	9.78	8.61	105.9	88.29	7.28	20.81	43.03
-OM CONTRO	IR29	80.33	11.33	7.00	144.0	78.27	6.81	17.90	48.60
	Sak.102	89.33	14.67	9.67	112.3	82.20	6.99	18.27	46.87
	Giza178	85.67	13.33	10.33	150.3	98.20	9.63	24.50	46.07
-OM 4000	IR29	83.33	5.33	6.33	141.0	58.57	6.17	17.23	47.30
	Sak.102	90.00	7.00	8.00	99.0	72.40	6.31	16.50	47.10
	Giza178	92.33	8.67	8.33	98.0	95.57	7.26	18.37	48.60
-OM 8000	IR29	88.33	2.67	3.33	76.0	36.40	2.03	9.27	29.40
	Sak.102	95.67	3.67	4.87	72.0	23.17	2.47	11.27	32.43
	Giza178	95.87	5.00	6.00	55.87	59.87	3.43	14.03	34.00
+OM CONTRO	IR29	84.67	14.67	8.33	139.0	78.17	6.94	17.37	47.40
	Sak.102	87.67	15.33	9.00	111.3	88.63	7.02	20.73	46.60
	Giza178	92.33	16.00	10.67	155.0	99.10	10.45	25.70	48.63
+OM 4000	IR29	87.33	8.00	7.00	141.7	62.10	6.42	13.33	48.63
	Sak.102	89.00	8.00	8.33	103.7	79.40	6.61	18.67	49.73
	Giza178	96.33	9.33	9.33	107.7	98.90	8.19	23.70	48.10
+OM 8000	IR29	90.67	3.33	5.00	74.3	43.07	2.36	11.80	31.80
	Sak.102	96.67	4.33	5.67	83.7	39.47	3.96	15.50	31.50
	Giza178	102.33	6.33	7.00	66.7	80.10	4.71	18.57	34.37
LSD 0.05	Salinity	1.893	0.951	0.556	2.762	1.587	0.038	0.374	0.259
	OM	0.785	0.444	N.S.	1.219	1.179	0.032	0.247	0.134
	Var.	1.253	0.761	0.678	1.753	0.923	0.031	0.176	0.225
	OM X Var.	1.574	N.S.	N.S.	2.246	1.469	0.045	0.296	N.S.
	Sal. X Var.	N.S.	N.S.	N.S.	3.279	1.809	0.053	0.396	0.371
	OM X Sal.	N.S.	N.S.	N.S.	N.S.	1.838	0.047	0.412	N.S.
	OM X Sal. X Var.	N.S.	N.S.	N.S.	N.S.	2.482	0.075	0.516	0.502

4.3. Physiological and biochemical compositions of shoot:

4.3.1. Photosynthetic pigments:

Data recorded in Table (4) show that, brackish water at all levels significantly decreased leaf pigments concentration, (chl.a, chl.b, total chl. and carotenoids), the most harmful effect was obtained by IR29. This decrease tended to increase with increasing brackish water levels.

Table (4): Effect of brackish water, organic matter, varieties and their interactions on physiological characteristics of rice at heading stage during 2005 season.

Treatments \ Characteristics		Chloro-phyll (a)	Chloro-phyll (b)	Total chl.	Caroten-oids	Total water content	Relative water content (%)	Leaf water deficit (%)	Transpiration rate (mg/cm²/h)
Salinity (ppm)	CONTROL	2.79	1.72	4.51	0.859	67.46	76.21	23.79	52.88
	4000	2.37	1.32	3.69	0.698	64.86	72.83	27.17	41.44
	8000	2.04	1.17	3.22	0.629	59.52	67.93	30.70	36.67
Organic matter	-OM	2.23	1.29	3.52	0.669	62.32	70.99	29.16	40.87
	+OM	2.58	1.51	4.09	0.788	65.57	73.66	25.28	46.46
Varieties	IR29	0.96	0.58	1.54	0.356	54.63	52.59	47.41	11.45
	Sak.102	1.70	0.97	2.67	0.478	60.78	72.13	27.87	27.72
	Giza178	4.55	2.65	7.19	1.352	76.42	92.24	8.39	91.82
-OM CONTROL	IR29	1.07	0.64	1.71	0.394	58.75	57.73	42.27	13.30
	Sak.102	1.82	1.04	2.86	0.498	62.51	73.93	26.07	29.25
	Giza178	5.06	3.21	8.27	1.458	78.49	95.73	4.27	105.46
-OM 4000	IR29	0.85	0.49	1.34	0.356	57.57	50.77	49.23	10.77
	Sak.102	1.60	0.92	2.52	0.468	58.08	70.43	29.57	26.47
	Giza178	3.92	2.27	6.19	1.148	75.81	91.73	8.27	80.39
-OM 8000	IR29	0.79	0.45	1.24	0.217	38.09	37.07	62.93	6.22
	Sak.102	1.49	0.78	2.27	0.435	58.90	69.33	30.67	25.91
	Giza178	3.46	1.82	5.27	1.025	72.72	92.17	9.20	70.07
+OM CONTROL	IR29	1.15	0.74	1.89	0.396	60.67	59.57	40.43	15.35
	Sak.102	1.86	1.06	2.92	0.499	83.56	74.00	26.00	29.31
	Giza178	5.80	3.61	9.41	1.907	80.79	96.27	3.73	124.61
+OM 4000	IR29	1.02	0.62	1.65	0.393	60.22	56.43	43.57	12.59
	Sak.102	1.78	1.03	2.82	0.496	60.55	72.93	27.07	28.84
	Giza178	5.04	2.57	7.61	1.342	76.94	94.70	5.30	89.57
+OM 8000	IR29	0.88	0.55	1.43	0.379	52.50	54.00	48.00	10.48
	Sak.102	1.67	0.99	2.66	0.455	61.11	72.17	27.83	26.53
	Giza178	3.98	2.46	6.43	1.229	73.79	82.87	7.57	80.81
LSD 0.05	Salinity	0.006	0.039	0.044	0.004	1.55	N.S.	1.467	0.742
	OM	0.003	0.023	0.032	0.002	0.92	4.591	1.207	0.270
	Var.	0.004	0.030	0.030	0.001	1.62	N.S.	3.498	0.771
	OM X Var.	0.006	0.039	0.044	0.001	N.S.	N.S.	3.705	0.818
	Sal. X Var.	0.008	0.051	0.057	0.001	2.59	N.S.	N.S.	1.203
	OM X Sal.	0.006	0.042	0.049	0.004	N.S.	N.S.	1.848	0.732
	OM X Sal. X Var.	0.009	0.089	0.078	0.002	N.S.	N.S.	5.482	1.302

The most reduction was obtained by 8000 ppm compared with control. This decrease may be due to the inhibitory effect of chloride on the activity of Fe-containing enzymes; cytochrome oxidase which in turn may decrease the rate of chlorophyll biosynthesis (Fouda, 1999), high rate of chlorophyll

degradation (Sharma and Gupta, 1986) and the high activity of chlorophyllase (Reddy and Vora, 1986). These results are in agreement with those obtained by Demiral and Turkan (2005) and Djanaguiraman *et al.* (2006) on rice. Application of organic matter significantly increased leaf pigments concentration, (Chl.a, Chl.b, total chl. and carotenoids) under stress conditions compared with control. This effect may be due to stimulating metabolism (Rashid, 1985), relieving oxygen deficiency and increasing the vital activity of cells, which aids chlorophyll synthesis. Similar conclusion was obtained by Levinsky (2001) and Oliver *et al.* (2007) on tomatoes.

4.3.2. Water relations:

Data in Table (4) show that, brackish water at all levels significantly decreased total water content, relative water content and transpiration rate, while increased leaf water deficit compared with control. This decrease tended to increase with increasing brackish water levels, the maximum reduction was noticed by 8000 ppm. These results may be attributed to the accumulation of toxic ions (Na and Cl) (Hasegawa *et al.*, 2000), reducing leaf expansion and stomatal closure leading to a reduction in intracellular CO_2 partial pressure or non-stomatal factors (Bethke and Drew, 1992). These results are in line with those obtained by Makihara *et al.* (2001) and Arunroj *et al.* (2004) on rice. Using organic matter significantly increased total water content, relative water content and transpiration rate, while decreased leaf water deficit in the leaves of plants irrigated with saline water compared with control. This improvement may be due to that, low-molecular-weight humic substances, such as fulvic acid enhanced ion transport, which may regulate transpiration rate and reduce water loss (Schnitzer and Khan, 1972) and enhanced plant circulatory systems, promoted optimum plant respiration and transportation systems (Rashid, 1985). These results are similar to those obtained by Sangakkara *et al.* (2005) on mungbean.

4.3.3. Proline concentration:

Results recorded in Table (5) show that, brackish water at all levels significantly increased proline concentration with increasing salinity levels in both seasons if compared with control. 8000 ppm gave the highest value. These results may be due to the accumulating of osmolytes that do not perturb enzyme functions so as to maintain continuous water absorption at the low soil water potential (Robinson and Jones, 1986) and via preserving osmotic balance and stabilizing the quaternary structure of complex proteins, membranes and many functional units like oxygen evolving PS-II complex (Rajasekaran *et al.*, 1997). These results are in accordance with those found by Demiral and Turkan (2005) and Djanaguiraman *et al.* (2006) on rice. Organic matter significantly decreased proline concentration in leaves of rice plants grown under salt stress conditions compared with untreated plants. These results are in agreement with those obtained by Oliver *et al.* (2007) on tomatoes.

Table (5): Effect of brackish water, organic matter, varieties and their interactions on shoot biochemical components of rice at heading stage during 2005 season.

Characteristics / Treatments			Proline	Nucleic acids (µg/ml)	N (%)	P (%)	K (%)	Na (%)	Ca (%)	Na/K ratio
Salinity (ppm)	CONTROL		0.635	0.014	2.68	0.251	2.82	0.610	0.854	0.223
	4000		0.819	0.009	2.41	0.219	2.36	0.674	0.717	0.292
	8000		1.863	0.007	1.89	0.173	1.62	0.849	0.526	0.558
Organic matter	-OM		1.209	0.009	2.00	0.199	2.16	0.755	0.635	0.403
	+OM		1.002	0.011	2.65	0.229	2.38	0.668	0.763	0.313
Varieties	IR29		1.229	0.008	2.07	0.177	2.02	0.825	0.562	0.465
	Sak.102		1.154	0.008	2.25	0.207	2.18	0.744	0.652	0.371
	Giza178		0.933	0.014	2.65	0.259	2.59	0.566	0.882	0.237
-OM	CONTROL	IR29	0.668	0.009	2.15	0.195	2.46	0.782	0.620	0.318
		Sak.102	0.666	0.013	2.26	0.217	2.54	0.674	0.747	0.266
		Giza178	0.559	0.017	2.64	0.275	3.06	0.426	1.073	0.139
	4000	IR29	0.918	0.008	1.87	0.164	2.14	0.851	0.460	0.397
		Sak.102	0.914	0.005	2.03	0.198	2.20	0.772	0.520	0.350
		Giza178	0.786	0.012	2.35	0.253	2.50	0.563	0.890	0.225
	8000	IR29	2.527	0.005	1.39	0.119	1.13	1.028	0.336	0.909
		Sak.102	2.117	0.003	1.51	0.162	1.48	0.879	0.432	0.595
		Giza178	1.687	0.011	1.83	0.213	1.92	0.819	0.635	0.426
+OM	CONTROL	IR29	0.666	0.012	2.75	0.226	2.71	0.687	0.816	0.254
		Sak.102	0.661	0.013	2.96	0.263	2.93	0.637	0.854	0.217
		Giza178	0.551	0.019	3.34	0.328	3.23	0.456	1.012	0.141
	4000	IR29	0.812	0.008	2.43	0.205	2.19	0.714	0.725	0.325
		Sak.102	0.789	0.007	2.64	0.224	2.33	0.666	0.785	0.286
		Giza178	0.698	0.014	3.12	0.273	2.81	0.479	0.920	0.170
	8000	IR29	1.788	0.007	1.85	0.151	1.50	0.885	0.415	0.589
		Sak.102	1.777	0.005	2.12	0.175	1.63	0.835	0.577	0.511
		Giza178	1.279	0.012	2.64	0.215	2.04	0.653	0.763	0.320
$LSD_{0.05}$	Salinity		0.007	0.004	0.011	0.005	0.036	0.001	0.008	0.005
	OM		0.004	0.002	0.003	0.003	0.024	0.002	0.008	0.003
	Var.		0.005	0.002	0.009	0.004	0.021	0.004	0.009	0.002
	OM X Var.		0.006	N.S.	0.011	N.S.	N.S.	0.005	0.013	0.004
	Sal. X Var.		0.008	0.005	0.015	N.S.	0.041	0.006	0.015	0.005
	OM X Sal.		0.008	N.S.	0.011	0.005	N.S.	0.003	0.011	0.005
	OM X Sal. X Var.		0.012	0.007	0.019	0.009	0.055	0.008	0.022	0.007

4.3.4. Nucleic acids concentration:

Results presented in Table (5) show that, brackish water at all levels significantly decreased nucleic acids concentration in shoot of rice plants in both seasons. This decrease tended to increase with increasing levels of brackish water. The most reduction was found under 8000 ppm. The reduction due to salinity was attributed to impair synthesis and/or enhancement DNase activity and leakage of divalent cations that normally stabilize ribosomes against endogenous nucleases as suggested by (Sheoran and Garg, 1978). These results are in line with those obtained by Mittal and Dubey (1990) on rice. Addition of organic matter significantly increased nucleic acids concentration in shoot of rice plants irrigated with brackish water compared with those untreated. This increase may be attributed to intensifying the metabolism of RNA, definitely increasing DNA contents in cells and also increasing and enhancing the rate of RNA synthesis (Khristeva, 1968). Similar results were recorded Levinsky (2001) on cotton.

4.3.5. Minerals concentration:

Data in Table (5) show that, brackish water at all levels significantly decreased shoot nitrogen, phosphorus, potassium and calcium percentages. The most decrease was pronounced especially at treatment 8000 ppm, while increased sodium percentage and Na/K ratio compared with control in the first season, second season showed the same trend. The deleterious effect of brackish water on nutrients uptake could be due to the competition and resultant selective uptake between potassium and sodium which caused an increase in the uptake of sodium at the cost of potassium and increasing concentration of sodium in the root medium which ultimately resulted in the increase uptake of sodium by plant (Aslam and Muhammed, 1972). Similar results were reported by Hussain *et al.* (2003) and Arunroj *et al.* (2004) on rice. Under salt stress conditions, application organic matter significantly increased shoot nitrogen, phosphorus, potassium and calcium percentages, while decreased sodium percentage and Na/K ratio compared with control in the first season. An explanation for this stimulative effect was that, organic matter enhances the availability of nutrients and makes them more readily absorbable, allows minerals to regenerate and prolongs the residence time of essential nutrients, prepares nutrients to react with cells and allows nutrients to inter-react with one another, breaking them down into the simplest ionic forms chelated by the fulvic acid electrolyte (Christman and Gjessing, 1983). These results are in accordance with those recorded by Sahrawat (2005) and Nozoe *et al.* (2006) on rice.

4.4. Chemical components of grain:

4.4.1. Amylose and Protein concetrations:

Data in Table (6) show that, brackish water at all levels significantly decreased amylose and protein percentages in grain, compared with control.

The most decrease was obtained in 8000 ppm of salinity levels at the first season, the same trend was noticed at the second one. The hazard effect of brackish water may be due to a reduction of protein synthesis or an acceleration of their degradation and/or an inhibition of amino acids incorporation into proteins (Fouda, 1999). These results confirmed with those obtained by Khan and Zaibunnisa (2003) and Acharya *et al.* (2008) on rice. Organic matter significantly increased amylose and protein in grain of the stressed plants compared with control. The second season was in the same line with the first one. These findings are in line with those obtained by. Levinsky (2001) on potatoes and tomatoes.

4.4.2. Mineral concentrations:

Data in Table (6) show that, brackish water at all levels significantly decreased grain nitrogen, phosphorus, potassium and calcium percentages. The most decrease was pronounced especially at treatment 8000 ppm, while increased sodium percentage compared with control in the first season, second season showed the same trend. The deleterious effect of brackish water may be due to a reduction of protein synthesis or an acceleration of their degradation and/or an inhibition of amino acids incorporation into proteins (Fouda, 1999). These results confirmed with those obtained by Mohiuddin *et al.* (1997) on rice. Organic matter significantly increased grain nitrogen, phosphorus, potassium and calcium percentages, while decreased sodium percentage of the stressed plants compared with control in the first season. Similar trend was found in the second one.

4.5. Plant genome (Molecular Analysis of Genetic Diversity of the Tested Varieties):

A total of three SSR markers *i.e.* RM223, RM315 and RM527 were used in this study, two of them are linked to salinity tolerance (RM223 and RM315), while RM527 was used randomly. A total of five alleles were detected among the seven genotypes. The number of alleles per locus ranged from one to three, with an average of 1.7 alleles per locus. There was no linkage among the SSR markers used and the salt tolerance in the varieties under this study. The two linked SSR markers to salinity (RM223 and RM315) didn't show any polymorphism among the studied varieties Table (7) and fig.(2 and 3). This may be because that, salinity tolerance is a quantitative trait controlled by a lot number of genes. The used markers aren't linked to the salt tolerance genes found in the studied genotypes. On the other hand, RM527 generated a clear level of polymorphism among the varieties fig. (4) but it wasn't linked to salinity tolerance.

Table (6): Effect of brackish water, organic matter, varieties and their interactions on grain biochemical components of rice at heading stage during 2005 season.

Treatments \ Characteristics			Amylose	Protein	N(%)	P (%)	K (%)	Na (%)	Ca (%)
Salinity (ppm)	CONTROL		16.86	18.79	3.16	0.454	3.37	0.263	0.268
	4000		14.33	15.11	2.54	0.397	2.90	0.418	0.241
	8000		10.76	5.65	0.95	0.296	2.22	0.519	0.189
Organic matter	-OM		13.41	11.93	2.01	0.376	2.74	0.412	0.200
	+OM		14.56	14.44	2.43	0.389	2.92	0.388	0.265
Varieties	IR29		12.78	11.75	1.98	0.318	2.49	0.471	0.207
	Sak.102		14.73	12.75	2.14	0.365	2.69	0.407	0.225
	Giza178		14.44	15.05	2.53	0.463	3.30	0.323	0.265
-OM	CONTROL	IR29	15.80	16.60	2.79	0.384	2.95	0.291	0.215
		Sak.102	17.50	17.43	2.93	0.432	3.13	0.273	0.226
		Giza178	17.00	18.45	3.10	0.528	3.79	0.214	0.264
	4000	IR29	15.21	11.13	1.87	0.328	2.46	0.518	0.187
		Sak.102	14.23	12.44	2.09	0.368	2.69	0.456	0.203
		Giza178	12.74	16.07	2.70	0.472	3.34	0.348	0.235
	8000	IR29	8.18	3.69	0.62	0.228	1.86	0.677	0.139
		Sak.102	10.28	4.64	0.78	0.273	1.97	0.525	0.151
		Giza178	9.74	6.96	1.17	0.367	2.47	0.409	0.183
+OM	CONTROL	IR29	15.33	18.68	3.14	0.390	3.11	0.304	0.275
		Sak.102	17.71	18.86	3.17	0.446	3.33	0.271	0.296
		Giza178	17.83	22.73	3.82	0.543	3.91	0.226	0.334
	4000	IR29	11.77	15.65	2.63	0.341	2.63	0.428	0.243
		Sak.102	15.60	16.96	2.85	0.384	2.82	0.412	0.264
		Giza178	16.44	18.45	3.10	0.486	3.46	0.343	0.312
	8000	IR29	10.41	4.76	0.80	0.237	1.96	0.605	0.185
		Sak.102	13.07	6.19	1.04	0.286	2.20	0.506	0.212
		Giza178	12.89	7.68	1.29	0.384	2.83	0.397	0.264
$LSD_{0.05}$	Salinity		0.116	0.053	0.009	0.002	0.022	0.001	0.002
	OM		0.143	0.086	0.014	0.001	0.012	0.001	0.001
	Var.		0.149	0.078	0.013	0.001	0.015	0.001	0.001
	OM X Var.		0.209	N.S.	N.S.	0.002	0.020	N.S.	0.001
	Sal. X Var.		0.227	0.116	0.019	0.003	0.028	0.002	0.002
	OM X Sal.		0.186	0.108	0.018	N.S.	0.023	0.002	N.S.
	OM X Sal. X Var.		0.339	0.181	0.030	N.S.	0.037	0.003	0.003

Table (7): The presence (+) and absence (-) matrix for SSR amplified fragments for the seven studied varieties:-

Markers	Varieties / No. of alleles	IR29	SK101	SK102	SK104	G177	G178	G182
RM223	1	+	+	+	+	+	+	+
RM315	1	+	+	+	+	+	+	+
RM527	1	-	+	-	+	-	+	+
	2	+	-	-	-	+	-	-
	3	-	-	+	-	-	-	-

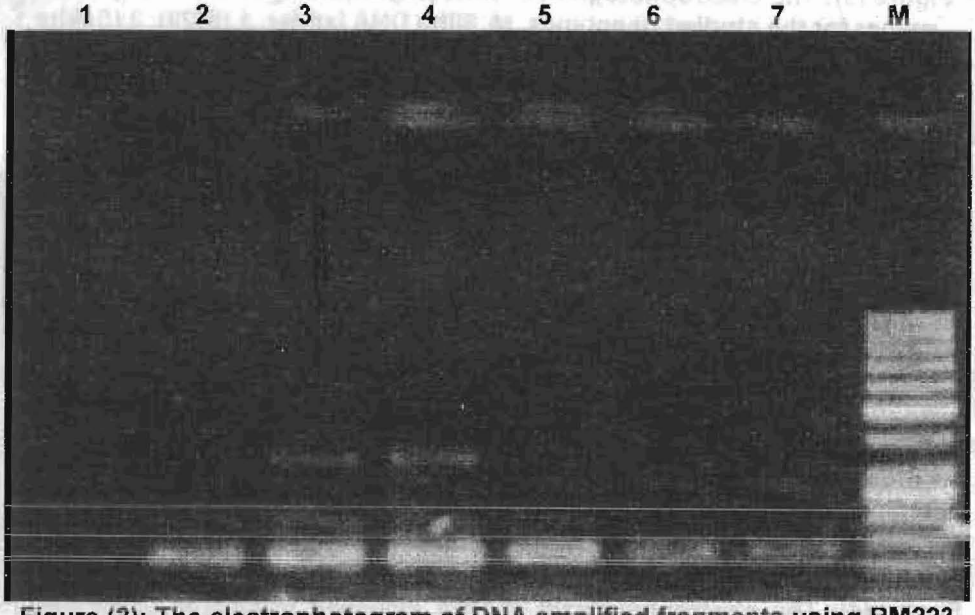

Figure (2): The electrophotogram of DNA amplified fragments using RM223 primer for the studied genotypes. M, 50bp DNA ladder, 1 (IR29), 2 (Sakha 101), 3 (Sakha 102), 4 (Sakha 104), 5 (Giza177), 6 (Giza178) and 7 (Giza182).

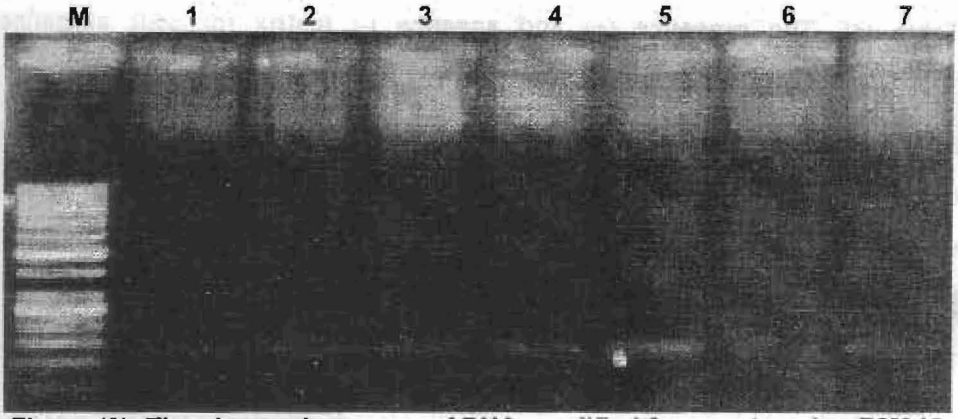

Figure (3): The electrophotogram of DNA amplified fragments using RM315 primer for the studied genotypes. M, 50bp DNA ladder, 1 (IR29), 2 (Sakha 101), 3 (Sakha 102), 4 (Sakha 104), 5 (Giza177), 6 (Giza178) and 7 (Giza182).

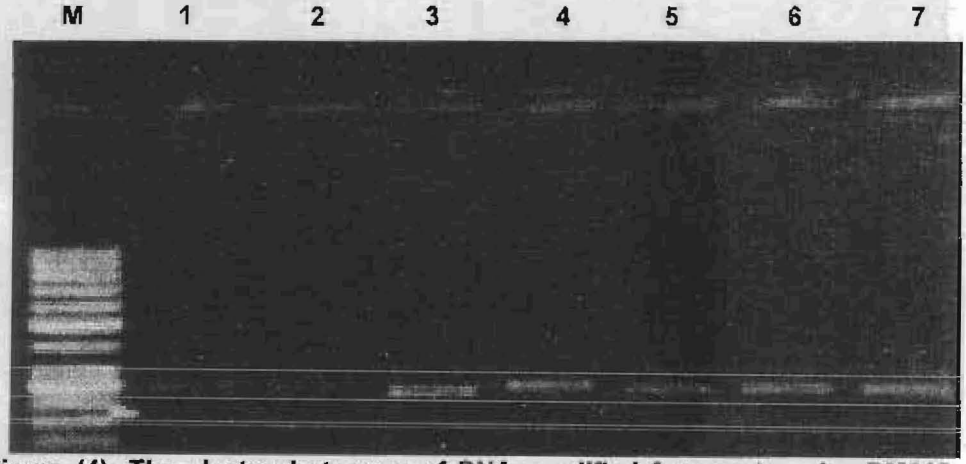

Figure (4): The electrophotogram of DNA amplified fragments using RM315 primer for the studied genotypes. M, 50bp DNA ladder, 1 (IR29), 2 (Sakha 101), 3 (Sakha 102), 4 (Sakha 104), 5 (Giza177), 6 (Giza178) and 7 (Giza182).

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استجابة بعض أصناف الأرز للرى بالماء المالح والزراعة العضوية

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الملخص العربي:

أجريت هذه التجربة باستخدام تقنية الليزيميتر بمركز البحوث والتدريب فى الأرز (كفر الشيخ) فى الموسم الصيفى لعامى ٢٠٠٥، ٢٠٠٦ وكان الهدف الرئيسى منها هو دراسة مدى تأثير المادة العضوية على الصفات المورفولوجية و الفسيولوجية والبيوكيميائية و الجينوم النباتى لثلاثة أصناف من الأرز هى جيزة ١٧٨ وسخا ١٠٤ وIR29 تم ريها بماء يحتوي عى خليط من أملاح كلوريد الصوديوم والكالسيوم(١:٢) بتركيزات ٠ و ٤٠٠٠ و ٨٠٠٠ جزء فى المليون ومن أهم النتائج التي توصلت اليها الدراسة:-

أن ري النباتات بالماء المالح بتركيزات ٤٠٠٠ – ٨٠٠٠ جزء في المليون أدى الى نقص معنوى فى طول النبات وعدد الخلفات والمساحة الورقية والوزن الطازج والجاف للمجموع الخضري وعدد السنابل/نبات – عدد السنيبلات /سنبلة – عدد الحبوب الكلية/سنبلة – % للخصوبة – وزن القش/نبات – وزن ١٠٠٠ حبة – دليل الحصاد بينما أدت الي تأخير التزهيربالمقارنة بالكنترول وكذلك أدت الي نقص معنوى في محتوي الاوراق من الصبغات ونسبة الماء الكلي – الماء النسبي – معدل النتح – الأحماض النووية في الأجزاء الخضرية ونسبة الأميلوز – البروتين – النيتروجين – الفسفور – البوتاسيوم – الكالسيوم في كلا من الأجزاء الخضرية والحبة بينما أدت الي زيادة عدد الحبوب الفارغة ونقص الماء الورقي – البرولين ونسبة الصوديوم – الصوديوم/البوتاسيوم. جيزة ١٧٨ هو أفضل الأصناف.

أدت المعاملة بالمادة العضوية الي زيادة معنوية في جميع صفات النمو الخضري والمحصولي والخصائص البيوكيميائية لكلا من المجموع الخضري والحبوب بالنسبة لجميع

الاصناف تحت الدراسة مع قلة في عدد الحبوب الفارغة - نقص الماء الـورقي - البـرونين. هذه الزيادة تتوقف علي مدي استجابة كل صنف لهذه المعاملة

النباتات النامية تحت ظـروف الملوحـة المـذكورة تحـسنت صـفاتها المورفولوجيـة. الفسيولوجية والبيوكيميائية عند معاملتها بالمادة العضوية وذلك عند مقارنتها بالنباتات النامية تحت ظروف الملوحة فقط ويتوقف ذلك علي الصنف ومدى حـساسيته للنمـو تحـت ظـروف الملوحة. وبذلك يمكن استخدامها لتقليل الأثر الضار للملوحة.

لم يكن هناك صلة بين الماركرين المرتبطين بالملوحة المستخدمين وصفة تحمل الملوحـة في الاصناف المستخدمة بينما.الماركر غير المرتبط بالملوحة اعطي تباين واضح بين الأصناف المستخدمة مع انه ليس مرتبط بالملوحة وذلك يدل علي أن هناك اختلافات علي مستوي الجزئ بين الأصناف تحت الدراسة.