Genetic variability for salinity water tolerance in tomato based on yield traits and related to molecular marker

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Abstract: Five tomato genotypes were sown in two successive seasons 2014/2015 and 2015/2016 at three saline water levels ($W1_{control}$ = 0.27ds/m, W2 = 5.53EC, W3=8.6EC).The main objective was to study the genetic variations for salinity tolerance in the local tomato genotypes based on yield traits and related to molecular markers. Highly significant differences were found between five tomato genotypes in both seasons across three level of salinity stress. Water salinity (W3) reduced the plant height (Ph), Number of branches (Nb), Number of clusters (Nc), Number of flowering / plant (Nfl), Number of fruits/plant (Nfr), Fruit set percentage (Fs%) ,Average weight /fruit (W/F) and yield/plant (Y/P) by 26.97% and 25.66%, 36.43% and 34.85 %, 35.52% and 35.35%, 29.42% and 29.44%, 47.o3% and 47.88%, 25.0% and 26.0 % , 37.29 and 38.78% , 47.83% and 47.95% relative to W1 at season 1 and season 2 for previous traits respectively .The loose percent / salinity unit in yield and different parameters were recorded. The highest mean performance for yield and weight fruits was displayed by line SV2 under water salinity stress, while for number of branches recorded by line SV1.The results of RAPD markers indicated that the fragment at molecular weight 527 bp with primer H19 was appeared only in line SV2 had highest mean of weight fruit and yield. As to markers with 1755, SV1 and 316 for primer Q 5and 300 bp for primer Q7 which had the highest number of branches. So,under upper Egypt conditions, these new breeding lines (SV1 and SV2) could be used to obtain high yielding tomato with water salinity tolerance.
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1. Introduction

Salinity stress is a major environmental constraint to irrigated agriculture in the arid and semiarid region. It is estimated between 30% and 40% of the world irrigated are affected by accumulated salts **(Flowers et al., 1986).** Tomato cultivars grow under specific and often extreme a biotic stress, such as salinity, drought and heat stress. This stress factor affected the plants during their life cycle from germination, vegetative growth until harvest. under such stress, plants are expose to many changes in their metabolisms and gene expression, which lead to a decrease in growth and increase in damage to the fruits. The irrigation water quality should be thus taken into account; excessive soil salinity reduces the yields of many crops. This may range from a slight loose to complete crop failure depending on the crop and the severity of the salinity problem. Therefore, breeding and understand of the nature and magnitude of the genetic variability play important role for developing tomato cultivars to be more tolerant to salinity. One way for increasing productivity in saline environment is to bread and selection crops more tolerant to salinity. However, success in breeding for salinity has been limited, therefore stress is controlled by many genes and selection is difficult. Agriculture productivity in arid and semi arid regions of the world is very low due to accumulation of salt in soil **(Ashraf and Sarwar 2002** and **Munns 2002**). Salinity is major factor limiting production in arid and semi arid region (**Bai et al., 2011)**. Now days, molecular markers for drought tolerance are essential and would be useful in screening different cultivars for their tolerance against salt stress. **Ehab et al., (2015)** used 16 inter-simple sequence repeat (TSSR) primers to study the genetic diversity among tomato cultivars. Also, their results were in good agreement with the results are regards selection induse and morphological characters.

RAPD (Randomly Amplified Polymorphism DNA) have recently shown excellent potentially to assist selection of quantitative trait loci associated with the salt tolerance, protein and enzyme **(Xue et al., 2008). Giora and Uri (2012)** reported that genotypic information is required in the form of markers for any quantitative trait loci or direct knowledge of the gens. Analysis of tomato cultivars grown under to different seasons in Egypt was analyzed by **Ahmed et al., (2009)**. Their results indicated that molecular diversity of cultivars were detected by using two molecular markers system of RAPD and TSSR. Developing some molecular

genetic markers associated with heat tolerance in tomato by using RAPD primers was developed by **(Kamel et al., (2010)**. They used bulk of the two extremely F_2 plants most tolerant and most sanative F2 group, the two contrasting parent and their F_1's to study the genetic variation for heat tolerance. Their results indicated tha RAPD markers with molecular size of 100 bp for primer A16 and 500 bp for primer Z13 were consider as reliable markers for heat tolerance as well susceptible genotype possessed eight RAPD markers 500 and 1500 bp for Co2, 1750 and 750for primer Co_3, 2400 bp for primer Co_5, 550 for Co_8, 400 bp for Co_{14}, and 650 pb for primer Co_{15}.The detection of RAPD markers on the genomic map of different field crops beneficial to improve breeding programs of these crops. It offers the simple and fasters method for detecting a great number in less period of time **(Edwards et al, 1992)**.Therefore, the present study aimed to study the performance and genetic variability for water salt tolerance among tomato genotypes based on yield traits and related molecular markers.

2. Materials and Methods

1-Field trails:

The present investigation was carried out at the experimental farm of Agricultural Research Center, Faculty of Agriculture, Qena, South Valley University, Egypt, which located at latitude 26°11' 25 N", and longitude 32° 44' 42" E, in hyper hot dry zoon around the tropic of cancer to study the genetic variability for salinity water tolerance in tomato based on yield traits and related to molecular marker. Three different saline irrigation water were used in this study; saline irrigation water with electrical conductivities (Ecw2 = 5.53 ds/m and Ecw3 = 8.6 ds/m) comparing with non-saline water with electrical conductivity (Ecw1 = 0.27 ds/m). Name, pedigree and source of the five tomato cultivars are presented in Table 1.

The two sources of saline irrigation water (W2 and W3) were obtained from an existing local wells and non-saline water (W1) was gained from Nil river. The Chemical analyses of the three sources of irrigation water are shown in **Table 3**. Primer codes used in the experiment work in Table 4

Table 1: Name, Pedigree and source of the five (cultivar and selected lines) of tomato used in this study.

Code No.	Genotypes	pedigree	source
1	Edkawy	Local cultivar	*ARC
2	SV1	Selected lines by mass selection under south valley conditions.	**SV
3	SV2		
4	SV6		
5	SV7		

***ARC: Agricultural Research Center, Egypt.**
****SV: South Vally, Qena, Egypt.**

Some physical and chemical characteristics of soil samples and are shown in Tables 2.

Table 2: Some physical and chemical properties of representative soil samples.

Soil properties		Values
Texture analysis	Clay %	4.0
	Silt %	18.8
	Sand %	77.2
Texture grade		Loamy sand
Total $CaCO_3$ %		3.0
EC. ds/m (sat. paste)		5.4
pH (1:1 suspension)		8.3
Soluble cations		
Ca^{++} meq/100 g soil		10.0
Mg^{++} meq/100 g soil		21.53
Na^{+} meq/100 g soil		28.35
K^{+} meq/100 g soil		0.32
Soluble anions		
$CO_3^{=}$ + HCO_3^{-} meq /100 g soil		0.2
Cl^{-} meq/100 g soil		22.33
SO_4^{--} meq /100g soil		37.6

Table 3. Chemical analysis of the three sources of irrigation water.

Irrigation water sources	Characteristics								
	pH	(EC ds/m)	Cations and anions (mmol/L)						
			Ca^{++}	Mg^{++}	Na^{+}	K^{+}	$CO_3^{=}$ + HCO_3^{-}	Cl^{-}	SO_4^{--}
Nil water (W1)	**7.15**	**0.27**	**0.65**	**0.62**	**0.95**	**0.05**	**0.55**	**0.90**	**1.05**
Well 2 (W2)	**7.94**	**5.53**	**10.7**	**10.5**	**42.15**	**0.9**	**3.2**	**31.0**	**25.5**
Well 3 (W3)	**7.5**	**8.6**	**45**	**42**	**12.6**	**0.28**	**0.98**	**69.9**	**29.0**

Table 4 : Primer codes used in the experiment work .

No. primer	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11
cod	O8	H19	H7	Q15	M6	D11	M18	M8	Q11	Q5	Q7

Soil and water analysis:

Soil texture was determined using the pipette method **(Piper, 1950).** In soil and water total carbonates were determined using the calcimeter method **(Nelson, 1982)**. Soil pH was measured by using a pH-meter in 1:1 soil-water suspension. Total soluble salts in the soil paste extract was measured by the electrical conductivity and soluble cations and anions, were performed according to the methods as described by **Jackson, (1973).**

The five tomato genotypes were sown in two successive winter seasons 2014/2015 and 2015/2016, respectively. In both seasons, the five genotypes were subjected to the irrigation with the three levels of water salinity (W1, W2 and W3) starting with planting to harvest. The experiment was laid in a randomized Complete Block Design (RCBD) with three replicates. The length of row was 5 m and spaced at part 30 cm. Data were recorded from five plants of each replicates and every genotype and included the following characters:

1- Plant height in centimeter (Ph).
2- Number of branches / plant (Nb).
3- Number of clusters (Nc)
4- Number of flowering / plant (Nfl).
5- Number of fruits / plant (NFR).
6- Fruit set% (FS%).
7- Average weight/fruit (W/F).
8- Yield /plant in gram (Y/P).
9-Proline content in plant related to salinity tolerance.

Molecular markers:

I- Extraction and purification of DNA:

DNA was extracted from 0.2 g of random picked fresh young leaf tissue of planting using CTAB protocol **(Murray and Thomasen, 1980)**.

II- Primer and DNA markers used RAPD:

RAPD analysis was based on the polymerase chain reaction (PCR) amplification of random sites spread all over the genomic DNA. The protocol was performed by **William et al., (1990).** Eleven random oligonucleotide (RAPD) were tested in this study with five genotypes tomato to amplify the template DNA in Table 4.

Statistical analysis:

Data analyzed with analysis of variance (ANOVA) was calculated for environmental one factor following **Gomez and Gomez** (1984).

3. Results and Discussion

Mean performance of different tomato genotypes as affected by irrigation with different water salinity:

1- plant height (Ph):

The analysis of variance for all traits studied indicated that the differences between five tomato cultivars were highly significant in both seasons Table 5. This results are harmony with those reported by (**Rashwan 2015 and 2016)**. The mean values for plant height over genotypes ranged from 61.53 cm with saline water W3 (8.6 ds/m) to 84.3 cm with control (0.27 ds/m), Table 6. With regard to the average over five cultivars, the mean of plant height ranged from 51.55 to 85.22 cm in the first season. However, in the second season, the mean ranged from 58.6 cm with water W3 to 84.26 cm in control average over five genotypes.

2- Number of branches (Nb):

In the first season, the mean values ranged from 8.6 to 13.53 indicating that wide range over their salinity stress, but in the second seasons ranged from 8.6 to 13.40. Meanwhile, average over salinity stress water, the genotype Edkawy gave the highest number of branches followed by genotype SV_2 and SV_2 in both seasons.

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Table (5): Analysis of variance for all studied traits of tomato genotypes in two seasons.

Season 1 (2104 / 2015)													
Items	d.f	plant hight			Number of branches			Number of clusters			Number of flowers		
		Ecw1	Ecw2	Ecw3	Ecw1	Ecw2	Ecw3	Ecw1	Ecw2	Ecw3	Ecw1	Ecw2	Ecw3
Rep.	2	0.067	0.200	0.467	1.867	0.267	0.200	0.200	1.400	0.467	1.667	5.067	6.200
G	4	694.9**	707.4**	401.767**	2.1**	1.667**	4.733**	12.233**	15.67**	11.5**	126.667**	93.567**	87.90**
Error	8	7.65	6.2	6.717	0.45	0.517	0.783	1.283	0.567	0.55	2.417	1.317	1.20
		Number of fruits			Number of fruit set %			Average fruit weight			Fruit yield		
Rep.	2	1.800	12.800	1.400	7.533	10.48	3.034	0.867	2.867	2,867	10166.667	13046.667	1860.00
G	4	11.100*	15.60**	77.10**	74.963**	38.61**	315.379**	225.833**	258.767**	323.167**	280250.00**	229166.667**	95026.667**
Error	8	2.300	0.300	0.900	5.019	1.345	4.840	3.783	1.876	2.867	1625.00	1421.667	1367.667
Season 1 (2105 / 2016)													
Items	d.f	plant hight			Number of branches			Number of clusters			Number of flowers		
		Ecw1	Ecw2	Ecw3	Ecw1	Ecw2	Ecw3	Ecw1	Ecw2	Ecw3	Ecw1	Ecw2	Ecw3
Rep.	2	5.267	7.267	4,200	0.001	0.867	1.667	0.867	1.867	0.600	0.600	6.200	0.601
G	4	616.900**	530.40**	385.567**	1.567**	2.600	5.333**	8.933**	11.433**	11.233	125.400**	89.100**	51.60**
Error	8	3.350	1.850	2.617	0.417	0.95	0.583	0.783	0.283	0.433	1.850	1.2	3.35
		Number of fruits			Number of fruit set %			Average fruit weight			Fruit yield		
Rep.	2	5.4	0.600	1.400	7.463	1.050	4.734	0.001	1.267	0.867	6846.667	5446.66	6.667
G	4	21.900**	30.00**	83.100**	117.087	89.369**	426.905**	146.10**	236.567**	357.167**	332233.33**	199900.00**	89556.667
Error	8	2.15	3.35	2.40	3.091	4.632	8.240	3.50	1.517	1.617	1831.333	530.00	481.66

Table (6): Mean performance for all studied traits of five tomato genotypes as affected by irrigation with different water salinity.

Tomato genotypes	Season 1 (2014/2015)			Mean of season 1	Season 2 (2015/2016)			Mean of season 2	Means of two seasons
	Ecw1 (0.27 ds/m)	Ecw2 (5.53 ds/m)	Ecw3 (8.6 ds/m)		Ecw1 (0.27 ds/m)	Ecw2 (5.53 ds/m)	Ecw3 (8.6 ds/m)		
	plant high (cm)								
Edkawy	59.66	50.00	45.00	51.55	60.00	52.66	43.00	51.89	51.72
SV1	90.00	80.00	65.00	78.33	87.66	82.00	61.00	76.89	77.61
SV2	94.66	88.00	73.00	85.22	97.00	85.00	71.33	84.44	84.83
SV6	80.00	70.00	54.66	68.22	84.66	74.00	51.66	70.11	69.16
SV7	97.00	85.00	70.00	84.00	92.00	82.66	66.00	80.22	82.11
Average	84.26	74.60	61.53	73.47	84.26	75.26	58.60	72.71	73.09
L.S.D. 0.05	7.35	6.55	6.81		5.90	3.58	5.62		
C.V %	3.28	3.34	4.21		2.17	1.81	2.87		
	Number of branches/ plant								
Edkawy	15.00	12.66	10.66	12.77	14.33	12.33	11.00	12.55	12.66
SV1	13.00	11.00	7.33	10.44	12.66	10.66	7.00	10.11	10.28
SV2	13.33	11.66	8.66	11.22	13.66	12.33	8.66	11.55	11.38
SV6	13.00	10.66	8.00	10.55	12.66	10.33	8.33	10.44	10.50
SV7	13.33	12.00	8.33	11.22	13.66	11.66	8.00	11.11	11.16
Average	13.53	11.60	8.60	11.24	13.39	11.46	8.60	11.15	11.20
L.S.D. 0.05	1.61	1.88	2.32		1.70	2.49	2.01		
C.V %	4.96	6.16	10.29		2.05	8.50	8.75		
	Number of clusters								
Edkawy	24.00	18.33	15.33	19.22	22.00	18.00	15.00	18.33	18.78
SV1	27.66	23.66	18.66	23.33	26.66	22.66	18.33	22.55	22.94
SV2	23.66	18.66	13.66	18.66	24.00	18.33	13.33	18.55	18.61
SV6	22.33	19.66	14.33	18.77	23.33	19.33	14.66	19.11	18.94
SV7	25.33	21.66	16.33	21.11	24.66	21.00	16.66	20.77	20.94
Average	24.60	20.39	15.66	20.22	24.13	19.86	15.60	19.86	20.04
L.S.D. 0.05	2.97	1.96	1.93		2.33	1.4	1.74		
C.V %	4.61	3.69	4.73		3.67	2.68	4.22		
	Number of flowers / plant								
Edkawy	68.00	64.00	52.00	61.33	66.00	63.00	51.00	60.00	60.67
SV1	82.00	77.00	57.00	72.00	80.00	75.00	55.00	70.00	71.00
SV2	69.33	65.00	45.00	59.78	72.00	67.00	47.00	62.00	60.89
SV6	80.00	74.00	54.00	69.33	82.00	76.33	56.00	71.44	70.39
SV7	79.00	72.00	59.00	66.00	77.00	70.00	57.00	64.67	65.33
Average	75.67	70.40	51.40	65.69	75.40	70.27	51.20	65.62	65.66
L.S.D. 0.05	4.05	2.80	2.88		3.58	2.88	4.81		
C.V %	2.05	1.26	2.07		1.8	1.56	3.44		

Table (6) continuous:

Tomato genotypes	Season 1			Mean of season 1	Season 2			Mean of season 2	Means of two seasons
	Ecw1 (0.27 ds/m)	(Ecw2 (5.53 ds/m)	Ecw3 (8.6 ds/m)		Ecw1 (0.27 ds/m)	(Ecw2 (5.53 ds/m)	Ecw3 (8.6 ds/m)		
Number of fruits / plant									
Edkawy	43.00	36.00	28.00	35.67	41.00	36.00	27.00	34.67	35.17
SV1	47.00	42.00	27.00	38.67	45.00	40.00	25.00	36.67	37.67
SV2	44.00	40.00	21.00	35.00	46.00	42.00	23.00	37.00	36.00
SV6	42.00	38.00	18.00	32.67	40.00	36.00	16.33	30.78	31.72
SV7	45.00	38.00	20.00	34.33	43.00	36.00	18.00	32.33	33.33
Average	44.20	38.80	22.80	35.27	43.00	38.00	21.87	34.29	34.78
L.S.D. 0.05	3.98	1.44	2.49		3.68	4.81	4.07		
C.V %	3.46	1.40	4.09		3.44	4.82	2.98		
Number of fruit set %/ plant									
Edkawy	63.23	56.25	53.84	57.77	62.12	57.14	52.94	57.40	57.59
SV1	57.31	54.54	47.36	53.07	56.25	53.33	45.45	51.68	52.37
SV2	63.46	61.53	46.66	57.22	63.88	62.68	48.93	58.50	57.86
SV6	52.50	51.35	33.33	45.73	48.78	47.16	29.16	41.70	43.71
SV7	56.96	52.77	40.80	50.18	55.84	51.42	38.29	48.52	49.35
Average	57.85	56.13	44.40	52.79	57.37	54.35	42.95	51.56	52.18
L.S.D. 0.05	1.82	3.04	5.79		4.62	5.66	7.55		
C.V %	3.85	2.07	5.02		3.09	3.96	6.79		
Average fruit weight									
Edkawy	120.00	115.00	90.00	108.33	118.00	113.00	87.33	106.11	107.22
SV1	137.66	132.33	86.00	118.66	134.00	130.00	83.66	115.89	117.28
SV2	124.66	120.00	75.00	106.55	126.00	122.00	77.00	108.33	107.44
SV6	119.00	110.00	70.00	99.67	121.00	113.00	67.33	100.44	100.06
SV7	115.33	108.66	65.33	96.44	117.00	108.00	61.33	95.44	95.94
	123.33	117.20	77.27	105.93	123.20	117.20	75.33	105.24	105.59
L.S.D. 0.05	5.11	3.59	4.45		4.92	3.24	3.38		
C.V %	1.58	1.16	2.19		1.52	1.05	1.69		
Fruit yield / plant									
Edkawy	1650.00	1400.00	1000.00	1350.00	1600.00	1416.66	1033.33	1350.00	1350.00
SV1	2450.00	2100.00	1373.33	1974.44	2466.60	2050.00	1390.00	1968.87	1971.66
SV2	2300.00	2033.33	1200.00	1844.44	2283.30	1993.33	1193.33	1823.32	1833.88
SV6	2083.30	1750.00	916.66	1583.32	2060.00	1700.00	953.33	1571.11	1577.22
SV7	2233.30	1800.00	1100.00	1711.10	2273.30	1856.66	1043.33	1724.43	1717.77
Average	2143.32	1816.67	1118.00	1692.66	2136.64	1803.33	1122.66	1687.54	1690.10
L.S.D.0.05	106.14	99.28	30.29		121.71	60.62	59.79		
C.V %	1.88	2.08	3.32		1.99	1.27	1.95		

3- Number of cluster (NC):

The analysis of variance as shown in Table 5 revealed that the differences among five tomato genotypes were highly significant over three level of water salinity. The mean performance of number of cluster ranged from 15.66 to 24.6 over the five cultivars indicating the wide range manifested. In the second season, the range was15.6 to 24.13 over the genotypes. The highest mean value were found for genotype SV_2 and SV_7 in both seasons.

4- Number of flower/ plant (NFL)

In the first season, the mean value ranged from 51.40 to 75.66 over the five genotype, but the mean values of five genotypes ranged from 61.33 for

Edkawy genotype to 72.o for SV_1. As to the second season, the mean values ranged from 51.20 to 75.4 over the five tomato genotype, meanwhile ranged from 60.0 for Edkawy genotype to to 71.4 for genotype SV_1.

5- Number of fruit/ plant (NFR) :

Data in Table 6 shows the mean values of number of fruit ranged from 22.8 in case of application W3 water (8.6ds/m) to 44.2 in control over the genotypes in the first season.

Regards to the second season, the mean ranged from 21.87 to 43.0 averaged over five tomato genotypes.

6- Fruit set (FS%):

The analyses of variance for all studied traits revealed that the differences were highly significant among five tomato genotypes , Table 5 showed that the mean values for set fruit over the five tomato genotypes ranged from 44.4% to 57.85% in the first season. In contrast, the mean values ranged from4 2.95 to 57.37 in the second season. Average over five tomato cultivars, the mean of fruit set ranged from 45.73% to 57.77 % in the first season and from 41.7 to 57.4 in the second season.

7- Weight fruit (WF):

The mean values of weight fruits are shown in Table 6.The mean average from77.27 g to 123.33 g in the first season and from75.33 to 123.2 in the second season over the five tomato genotypes. The mean of five tomato genotypes ranged from 95.44 to 115.89 in the second season.

8- Yield /plant (YP)

The analyses of variance were highly significant among five tomato cultivars in both season, Table 5. The mean of yield ranged from 1118 g / plant to 2143.33 over genotype in the first season, while in the second season ranged from 1122.66 to 2136.40 g/plant might be due to environmental change which represent by different levels of salinity Table 6. As to the five tomato cultivars, the mean range from 1350 to 1974.4 in first season, while in the second season ranged from 1350.0 to 1968.8, table 6. **Ahmed (2001)** stated that Edkawy cv. was the most superior in all studied vegetative growth, fruit quality, yield and yield components compared with other cultivars under soil salinity.

Reduction and looseing for all studied traitsas affected by water salinity:

The results in table 7 for the two growing seasons showed reduction in plant height, number of branches, number of clusters, number of flowers, number of fruits, number of fruit set %, average fruit weight and fruit yield as results of increasing salinity which accumulated after irrigation with saline water. Table7. At first season 2014/2015, comparing the growth parameters of five tomato cultivars grown under different water salinity. Data of the first season showed that all growth parameters and yield of different tomato cultivars irrigated with both of saline water (EC_{w2} = 5.5 dS/m) and (EC_{w3} = 8.6 dS/m) found to be less significantly in all parameters and yield comparing the with non saline water (EC_{w1} = 0.0.27 dS/m). The results also showed that the reduction in all parameters were higher in case of irrigation with ECw3 = 8.6 dS/m than EC_{w2} = 5.5 dS/m. The most pronounced reductions were recorded with number of fruits / plant and fruits yield /plant. For plant fruits number, the reduction reached to 16.28%, 10.64%, 9.09% , 9.52 % and 6.98% when saline water with EC_{w2} = 5.5 dS/m was used for irrigation tomato genotypes Edkawy cv., SV1, SV2, SV6, andSV7, respectively, while the reduction reached to 34.88 %, 42.55%, 52.27%, 57.14% and 55.56 % after irrigation with EC_{w3} = 8.6 dS/m for the previous cultivars respectively. With regard to the effect of irrigation with these water on the fruit yield / plant, the reduction reached to 15.15%, 14.29%, 11,59%, 16.00% and 19.40% when saline water with (EC_{w2} = 5.53 dS/m) was used for irrigation tomato cultivars Edkawy cv., SV1, SV2, SV6, and SV7respectively, while the reduction in fruit yield /plant when application saline water with (EC_{w3} = 8.6 dS/m) was higher were the reduction reached to 39.39%, 43.95 %, 47,83%, 56.0% and 50.75% for the previous cultivars respectively. The results showed similar trend for the second season 2015/2016 as it previously found in the case of the first season. These results agree with a long number of researchers, where they pointed out that the increase in the salinity of water irrigation led to a decrease of the yield plant and its components directily or indirectily **(Mahmoud et al.1986 ; Ashraf and Meneilly 1988 ;Cerda and Martineze 1988; Hashim et al. 1988; Hamed et al.1988; Abdel-Noure 1989; Caro et al.1991; Al-Rawahy et al.1992; Johanson et al. 1992 ; Sary and omar 1993; Alarcon et al.1994; Ahmed 1996; Mahmoud 1996;Van 1996; Hassan 1999; Amico et al.2003; Maggio et al. 2004; Hajer et al. 2006 ; Wan 2007; Abo- baker 2009; Boamah et al.2011 and Elameen 2013)**

The parameters and yield loose / unit increase in water salinity were calculated as shown in **Table 8** showed that loosed per 1 unit increase in water salinity were happened in all growth parameters and fruits yield of the two growing season .

According to the results of fruit yield / plant as indicator of tomato genotype performance, data showed that the lower loose in yield / EC_w were recorded in case of SV1 and SV2 lines, where these looses when with irrigation with w2 water reached to (2.72 % and 3.21%) and for SV1 line in the first and second season respectively; and reached to (5.28% and 5.24%) with application W3 water with the same

arrangement respectively. In case of SV2 line when with irrigated with w2 water these looses in yield reached to (2.20 % and 2.41%) for SV1 for first and second season respectively; and reached to (5.74% and 5.73%) with application W3 water with the same arrangement respectively.

Table 7: Reduction percentage of tomato growth parameters of five tomato cultivars grown as affected by different water salinity comparing with non saline water.

Tomato genotypes	Season 1 (2014/2015)		Season 2 (2015/2016)	
	Irrigation water 2 (Ecw = 5.53 ds/m)	Irrigation water 3 (Ecw = 8.6 ds/m)	Irrigation water 2 (Ecw = 5.53 ds/m)	Irrigation water 3 (Ecw = 8.6 ds/m)
	Reduction % in plant hight			
Edkawy	16.19	24.57	12.23	28.33
SV1	11.11	27.78	6.46	30.41
SV2	7.04	22.88	12.37	26.46
SV6	12.50	31.68	12.59	38.98
SV7	12.37	27.84	10.15	28.26
	Reduction % in number of branches/ plant			
Edkawy	15.60	28.93	13.96	23.24
SV1	15.38	43.62	15.80	44.71
SV2	12.53	35.03	9.74	36.60
SV6	18.00	38.46	18.40	34.20
SV7	9.98	37.51	14.64	41.43
	Reduction % in number of clusters			
Edkawy	23.63	36.13	18.18	31.82
SV1	14.46	32.54	15.00	31.25
SV2	21.13	42.27	23.63	44.46
SV6	11.96	35.83	17.15	37.16
SV7	14.49	35.53	14.84	32.44
Reduction % in number of flowers / plant				
Edkawy	5.88	23.53	4.55	22.73
SV1	6.10	30.49	6.25	31.25
SV2	6.25	35.09	6.94	34.72
SV6	7.50	32.50	6.91	31.71
SV7	8.86	37.97	9.09	38.96
Reduction % in number of fruits / plant				
Edkawy	16.28	34.88	12.20	34.15
SV1	10.64	42.55	11.11	44.44
SV2	9.09	52.27	8.70	50.00
SV6	9.52	57.14	10.00	59.18
SV7	15.56	55.56	16.28	58.14
Reduction % in number of fruit set %/ plant				
Edkawy	11.04	14.85	8.02	14.78
SV1	4.83	17.36	5.19	19.20
SV2	3.04	26.47	1.88	23.40
SV6	2.19	36.51	3.32	40.22
SV7	7.36	22.68	7.92	31.43
Reduction % in average fruit weight				
Edkawy	4.17	25.00	4.24	25.99
SV1	3.87	37.53	2.99	37.57
SV2	3.74	39.84	3.17	38.89
SV6	7.56	41.18	6.61	44.36
SV7	5.78	43.35	7.69	47.58
Reduction % in fruit yield / plant				
Edkawy	15.15	39.39	11.46	35.42
SV1	14.29	43.95	16.89	43.65
SV2	11.59	47.83	12.70	47.74
SV6	16.00	56.00	17.48	53.72
SV7	19.40	50.75	18.33	54.11

Table 8: loose percentage / ECw unite in tomato parameters under irrigation with saline water (w2 and w3) comparing with non saline water (w1) at two season.

Tomato cultivars	Season 1		Season 2	
	Irrigation water 2 (Ecw = 5.53 ds/m)	Irrigation water 3 (Ecw = 8.6 ds/m)	Irrigation water 2 (Ecw = 5.53ds/m)	Irrigation water 3 (Ecw = 8.6 ds/m)
	Loose % / ECw unite in plant hight			
Edkawy	**3.08**	2.95	2.33	3.40
SV1	**2.11**	3.33	1.23	3.65
SV2	1.34	2.75	2.35	3.18
SV6	2.38	3.80	2.39	4.68
SV7	2.35	3.34	1.93	3.39
	Loose % / ECw unite in number of branches/ plant			
Edkawy	2.97	3.47	2.65	2.79
SV1	2.92	5.24	3.00	5.37
SV2	2.38	4.21	1.85	4.39
SV6	3.42	4.62	3.50	4.11
SV7	1.90	4.50	2.78	4.97
	Loose % / ECw unite in number of clusters/plant			
Edkawy	4.49	4.34	3.46	3.82
SV1	2.75	3.91	2.85	3.75
SV2	4.02	5.07	4.49	5.34
SV6	2.27	4.30	3.26	4.46
SV7	2.75	4.27	2.82	3.89
loose % / ECw unite in number of flowers / plant				
Edkawy	1.12	2.82	0.86	2.73
SV1	1.16	3.66	1.19	3.75
SV2	1.19	4.21	1.32	4.17
SV6	1.43	3.90	1.31	3.81
SV7	1.68	4.56	1.73	4.68
Loose % / ECw unite in number of fruits / plant				
Edkawy	3.09	4.19	2.32	4.10
SV1	2.02	5.11	2.11	5.34
SV2	1.73	6.28	1.65	6.00
SV6	1.81	6.86	1.90	7.10
SV7	2.96	6.67	3.09	6.98
Loose % / ECw unite in number of fruit set %/ plant				
Edkawy	2.10	1.78	1.52	1.77
SV1	0.92	2.08	0.99	2.30
SV2	0.58	3.18	0.36	2.81
SV6	0.42	4.38	0.63	4.83
SV7	1.40	2.72	1.50	3.77
Loose % / ECw unite in average fruit weight				
Edkawy	0.79	3.00	0.81	3.12
SV1	0.74	4.51	0.57	4.51
SV2	0.71	4.78	0.60	4.67
SV6	0.71	4.94	1.26	5.32
SV7	1.10	5.20	1.46	5.71
Loose % / ECw unite in fruit yield / plant				
Edkawy	2.88	4.73	2.18	4.25
SV1	2.72	5.28	3.21	5.24
SV2	2.20	5.74	2.41	5.73
SV6	3.04	6.72	3.32	6.45
SV7	3.69	6.09	3.48	6.50

This indicated that SV1 and SV2 lines had more ability to water salinity tolerance comparing to SV6 and SV7 which recorded higher loosing in yield reached to (3,04 % and 3.32 %) when SV6 irrigated with water W2 for first and second season respectively, and (3.69% and 3.48%) for application W2 water with SV7 line at first and second season respectively.

Also, the same trend was found with irrigation with W3 water where the two lines SV6 line and SV7 recorded higher loosing in yield reached to (6.72% and 6.45%) for SV6 line and (6.09 % and 6.5%) for SV7 line at first and second season respectively.

Pasternak et al., 1979; Cuartero and **Soria, 1997** reported that small increase in the salinity of irrigation water is expected to produce yield losses. In general, the number of fruits / plant, average fruit weight and fruit yield / plant had the higher loose per 1 unit increase in water salinity than the rest parameters. This may be due to a possibility that plants grown under saline condition utilize energy for osmotic adjustment process at the expense of growth and the most important factor which is the high soil water potential, hence the water flow from soil to plant is very much limited under saline conditions **(Ragab et al., 2008).**

Also, **Maas and Grattan (1999);** provide a guidelines for estimated yield of vegetable with long-terms use of different irrigations qualities, the guidelines indicated that 100% tomato yield can achieve with irrigation with ECw = 1.7 ds/m and decrease in yield reached to 10%, 25% and 50% when water salinity reached to 2.3 ds/m, 3.4 ds/m and 5 ds/m, respectively.

Irrigation water quality can affect soil fertility and efficiency of the irrigation system as well as crop productivity and soil physical situation **(Ayers and Westcot 1985 and Al-Omran *et al.*, 2010)**. According to **Olympios *et al.* (2003)**, increasing EC of irrigation water from 1.5 to 3.2 dS/ m did not affect the vegetative growth, but the yield was 45% less. **Zein *et al.* (2003)** found that wheat grain and straw yields as well as plant height, spike length, and 1000 grain weight were significantly affected by increasing irrigation water salinity.

Similar results were reported by **Al -Harbi *et al.* (2009)**. They mentioned that, irrigation with saline water having EC 4.7 dS/ m significantly reduced the total fruits yield by 24.3%. **Maggio *et al.* (2007)** reported that there was an approximately 6% reduction in plant dry mass per one dS/ m increase until approximately 9 dS m-1, whereas, only 1.4% decrease in yield per dS/ m after 9 dS/ m. **Al-Omran *et al.* (2012)** concluded that the adverse effect of irrigation with saline water on total dry biomass and total fresh tomato fruit yield were the reduction in WUE and TYWUE. **CAST (1988)** reported that at a given salinity level of applied water, corn yield decreases as salinity levels increase.

II- Random amplified polymorphic DNA (RAPD):

RAPD marker can be efficiently used to study the genetic diversity under salinity water stressed and find out genetic relationship among the cultivars, which is an essential component in germplasm characterization and conservation RAPD based DNA fingerprints of chose tomato cultivars. In this study, four primers gave high polymorphism, namely H7, Q5, Q7 and H19 with 100%, 100, 50% and 20% respectively, Table 9.

Table 9: Polymorphism percentage genrrated by eleven primers in four tomato cultivars.

Primer	Monopheric band	Unique	Nonunique	Total bands	Polymorphic %
O 8	2	0	0	2	-
H 19	4	0	1	5	20
H 7	0	0	3	3	100
Q 15	3	1	0	4	-
M 6	4	0	0	4	-
D 11	7	0	0	7	-
M 18	4	0	0	4	-
M 8	4	0	0	4	-
Q 11	2	0	0	2	-
Q 5	-	0	3	3	100
Q7	2	0	1	4	50

The primer Q5 generate three fragments ranged from 316 bp to 1755 which displayed in only line SV1 which disapperead in other cultivars as well as Q7 primers generated one fragment with 300 bp in line SV1, but not manifested in other cultivars. **Ehab et al., (2015)** revealed that the data of molecular markers were in good agreement with selection indices the four RAP marker of primer Q5 and Q7

could be considered reliable markers for salinity stress. Semilar results were obtained by **Kamel et al., (2010)** who found two RAPD markers with molecular size of 100 bP for primer A16 and 500 6P for primer Z13 and one ISSR marker with 650 bP were considered as reliable marker for heat tolerance. The two DNA 6 and at molecular weight 527 for H19 primer and 1000 bP for primer Q15 were unique to line SV1 and line SV7.

These results reflect that two primers were able to distinguish the tomato cultivars. Unique bands have also been reported by **Teshale et al., (2003).**

RAPD marker related to agro-morphological:

The result in Table10 and Fig 1. Showed that the fragment at molecular weight 527 bP for primer H19 was appeared only in line SV2 which had the highest mean value of weight for fruit and yield. The markers with molecular 1755, 452 and 316 bP for primer Q5 and 300 bP for primer Q7 were appeared only in line SV1 which had highest mean number of branches.

Table 10: Survey of the eleven primers fragments with five tomato cultivars.

Primer	Ms bP	Line SV1	Line SV2	Line SV6	Line SV7
O 8	440	1	1	1	1
H 19	527	0	1	0	0
H 7	338	1	0	1	1
Q 15	1000	1	1	1	0
M 6	355	1	1	1	1
D 11	1	1	1	1	1
M 18	421	1	1	1	1
M 8	119	1	1	1	1
Q 11	352	1	1	1	1
Q 5	1775	1	0	0	0
-	542	1	0	0	0
-	316	1	0	0	0
Q7	300	1	0	0	1

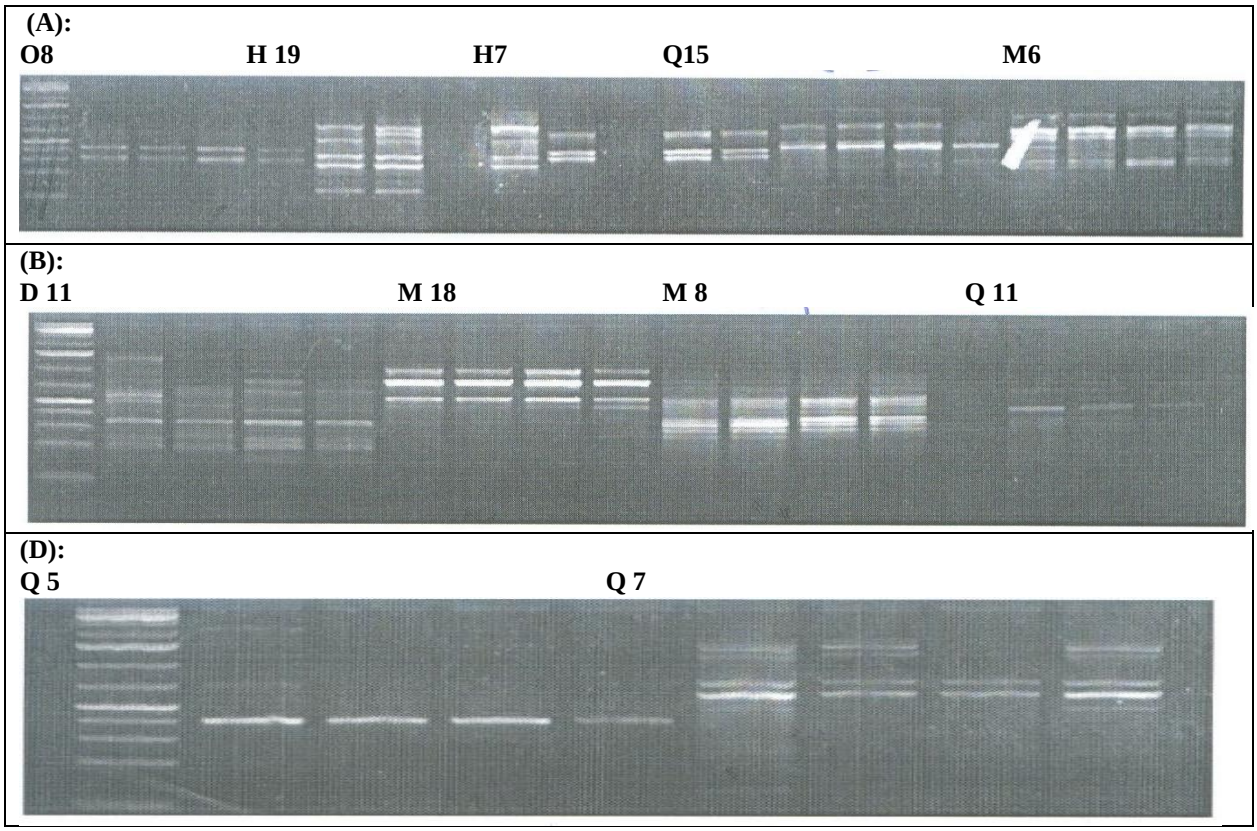

Figure 1: RAPD PCR fragments (A) of primers 08,H19,H7,Q15,M6,(B) D11,M18,M8,Q11, (C),Q5 and Q7.

Conclusion

In most environmental conditions in which it is cultivated, the tomato begins to loose yield when irrigated with water whose EC is above 2-3 dS/ m when compared to fresh water irrigation. With regards to the results of the looses in yield / EC_w increasing unit for different tomato genotypes under investigation, and in accordance with the positive results of primer polymorphism based on RAPD markers, results showed that line SV1 and SV2 more tolerant to salinity up to 8.6 ds/m. So, breeding of tomato cultivars tolerant to moderate-high salinity will occur after pyramiding in a single genotype several characteristics that each alone could not confer a significant increase in the tolerance. Under Upper Egypt conditions, these new breeding lines (SV1 and SV2) could be used to obtain high yielding tomato with salinity tolerance.

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