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IMPROVING THE POTENTIAL YIELD OF SUGAR BEET CROP UNDER SALINITY STRESS

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ABSTRACT

The elevation of salt tolerance for sugar beet plants still presents problems. However, understanding the mechanisms of salt tolerance in plants is an essential for avoid or limit the risk of salt stress on the efficiency crop production. This work aimed to study the physiological and yield responses for transplanted (T) and direct seed sowing (DSS) as affected by increasing NaCl salinity up to half of the sea water level. Seeds and seven-week old plants were sown in pot experiments under five NaCl salinity levels (control, 50, 120, 190 and 260 mM). The results, under control treatment, indicated no significant differences between transplanted and direct seed sowing on root or sugar yields, but a marked reduction of water consumption (21%) was recorded by transplanting compared with direct seed sowing. However, adding the first NaCl level (50 mM) significantly reduced root and sugar yields around 63% and 58% respectively for direct seed sowing, while the reduction of yield recorded 13% and 10% for transplanted compared with the control respectively. Raising NaCl level to 120mM sharply reduced yields for direct seed sowing to loss around 97% and 95% from root and sugar yields respectively, meanwhile this reduction reached around 52% and 47% for transplanted, compared with control, respectively. Moreover, no further growing was detected for direct seed sowing under the NaCl salinity levels 190 and 260mM. Meanwhile the transplanted survived and grow till the half sea water salinity level (260mM). In spite of linearly decreases for K/Na ratio in leaf blade, petioles and roots due to salt treatments, but in case of transplanted at 50Mm NaCl level, K contents in all plant parts are still higher than Na contents (the value of ratio is over one). On the other hand, chlorophyll content per cm^2 for juvenile and adult leaves were increased by increasing NaCl but that because of reducing leaf area and increasing of leaf succulence.

INTRODUCTION

Sugar beet is one of the main sugar crops. In 1982, sugar beet has been introduced in Egypt as a new sugar crop and a second source for sugar after sugar cane. By the year 2003 it was cultivated in about 131,323 feddans as compared with 16,949 feddans in 1982. Bringing a new land under cultivation is a fundamental goal to minimize the gab between sugar production and consumption.

This work aimed to Improving the salt-tolerant of sugar beet as a strategy crop provides a sensible alternative to utilizing land and saline water which are unsuitable for other conventional crops. Millions of feddans are suffered from salinity problem; the majority of salt-affected soils are located in northern Nile delta along with the Mediterranean coast, Sahl El-Tina area located in northern Sinai which consists of 357,000 feddans and some oasis like Fayoum. However, except during the early stage, after crop establishment (25-35 days after sowing), the sugar beet crop is tolerant to salinity (Marschner, 1995). The mechanisms of sugar beet salt-tolerance are mainly depended on its ability to decrease root osmotic potential whereas, soluble sugars concentration in root play the main role for adjusting root osmotic potential (Lindhauer *et al* 1990; Marschner, 1995; Eisa *et al* 2001 and Eisa & Ali, 2005). World wide Sugar beet transplanted on paper pot is commonly used, for example to avoid root rot (Kajiyama and Tanaka, 2000), or for improving Biocontrol management (Grondona *et al* 2001) and for reducing fertilizer rates and increasing root yield (Gerne *et al* 2000).

MATERIAL AND METHODS

Two pot experiments were carried out during the two successive seasons 2001/2002 and 2002/2003 at the Faculty of Agriculture, Ain Shams University, Cairo, Egypt. A comparative study between normal sowing of sugar beet (direct seed sowing) and transplanting of adult plants cultivated in paper-pots as an alternative way to avoid the harmful effect of salt stress on sensitive growth stages (germination and early growth stages) of sugar beet was made. However, a germination test was initially done to determine the maximum NaCl salinity level for germination.

Germination test

Seeds of sugar beet (*Beta vulgaris* L. var. altissima Doell) genotype top were sterilized by immersing in 2% sodium hypochlorite for 7 min. and rinsing repeatedly with distilled water. Twenty five seeds were germinated in Petri dishes (9 cm) containing a filter papers (Whatman No.1), moistened with 3 ml of tap water or saline solution (17, 34, 51, 68, 85, 102, 119, 136 and 153 mM NaCl). Each treatment was performed five times. Dishes were incubated at 27°C±2 under dark condition, for two weeks after then the germination percentage and seedling fresh wight were estimated after two weeks (Table 2).

Pot experiments

Seeds of sugar beet, variety top, were sown on September 10^{th} and 15^{th} for the first and second season respectively. Two sowing methods (transplanting using paper pot and seed direct sowing) were tested under five salinity irrigation levels namely; control, 50, 120, 190 and 260 mM NaCl. The pot experiments were set in a completely randomized design with eight replications for each treatment.

As for the paper-pot sowing, the paper-pot plate (Figure 1) was initially stretched and fixed on a bottom drainage foam plate. The paper pot holes were filled with washed sand soil, and then three seeds per hole were sown and irrigated with tap water for the first two weeks. Seedlings were thinned to one per hole after two weeks from sowing then irrigated with nutrient solution (Arnon and Hoagland, 1940). After seven weeks from sowing date, each individual paper-pot sack content one plant, was separated and transplanted into 35 cm diameter pot (bottom drainage pots), filled with sandy soil. Immediately after transplanting the pots were irrigated with salt treatments till the end of the experiments (22 weeks). The characters of seven weeks old plants are presented in Table (1).

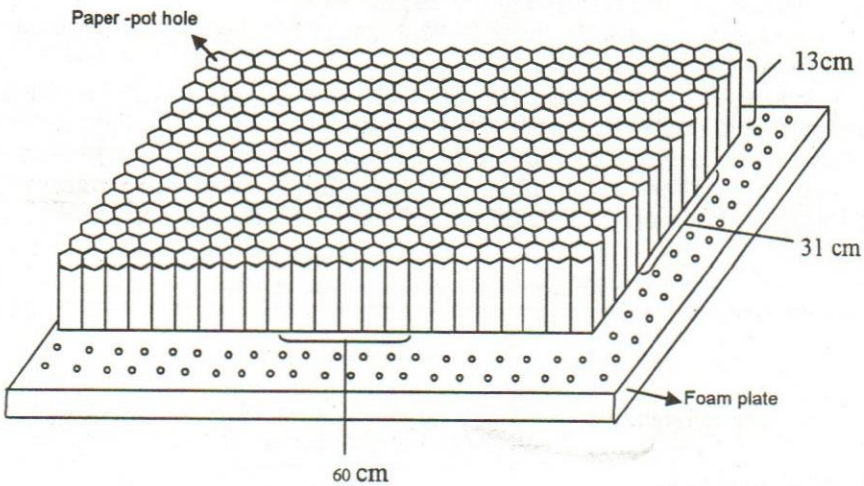

Fig. (1). Diagrammatic sketch of the paper-pot

Table (1): The characters of seven-week old plants before transplanting

Plant height (cm)	15-17
Shoot fresh weight (g)	14-17
Root fresh weight (g)	1.3-1.7
Root diameter (mm)	0.80-0.90
Number of full expanded leaves	Five
Total soluble carbohydrates in root	30-34%
Total soluble carbohydrates in shoots*	0.9-1.1%

* Total soluble carbohydrates were estimated according the method described in A.O.A.C.

As for direct seed sowing, three seeds were sown per pot (35 cm diameter with bottom drainage). The pots were filled with washed sandy soil. The pots irrigated with tap water for the first two weeks then seedlings were thinned to one per pot. Salt treatments were added to water irrigation after two weeks from sowing date till the end of the experiments (22 weeks).

Basic nutrient solution after Arnon and Hoagland (1940) was used after second week to irrigate the cultivated plants till the 19^{th} week from sowing date for all pot experiments.

Sampling:

Three plants were randomly taken from each treatment at the 12^{th} week from sowing date (after five weeks from transplanting date). Fresh weight of roots, total leaf area per plant as well as juvenile leaf area (the youngest full emergent leaf from the top) and adult leaf area (the oldest healthy green leaf) were estimated.

Leaf succulence was calculated using the following equation:

$$\frac{\textit{Leaf mass (g fresh weight)}}{\textit{leaf area (cm}^2\textit{)}} X\ 100$$

Potassium and sodium contents in leaf blades, petioles and roots were determined using flame photometer Petracourt PFPI.

Chlorophyll content in juvenile leaf and adult leaf was determined according (Arnon, 1949).

Harvesting took place 22 weeks after sowing date for both seasons. Five plants from each treatment were taken to determine the fresh root yield and sucrose percentage in root. Root sucrose percentage was measured according to Shaffer and Hartmann (1921). Sugar yield and water use efficiency per plant were calculated according to the following equations:

$$\textit{Sugar yield/ plant (SY)} = \frac{\textit{root sucrose percentage x root fresh weight}}{100} = \textit{g sugar/ plant}$$

$$\textit{Water use efficiency} = \frac{\textit{sugar yield per plant}}{\textit{Received water per plant}} = \textit{g sugar / liter}$$

Statistical analysis

Statistical analysis was performed using Statistical Analysis System (SAS). Means of the two seasons were compared using the least significant difference, which developed by Duncan (1955) at 5% level of probability.

RESULTS

Germination test:

Table (2) showed that germination percentage of sugar beet seeds reached the maximum in tap water (control) and the first NaCl level (17mM) treatments. Meanwhile, raising NaCl levels decreased the rate of germination to record 40% and 4% at 68 mM and 136 mM NaCl respectively.

Table (2): Effect of NaCl salinity levels on germination percentage and seedling fresh weight (FW) after two weeks

Salinity levels (Mm)	Germination percentage (%)	Seedling FW (g)
Control	80	0.10
17	80	0.11
34	72	0.06
51	60	0.05
68	40	0.04
85	24	0.04
102	12	*
119	8	*
136	4	*
153	--	*

-- Seed fell to germinate

* Germinated seed had failed to develop into seedling

However, the seed fell to germinate at153 mM NaCl during the first two weeks. On the other hand, the seedling fresh weight obviously reduced at 34 mM NaCl but, starting from 102mM NaCl, the seeds had failed to develop into seedling (Table 2).

Growth parameters and chemical analysis:

As for some growth parameters at twelve weeks from sowing, data in Table (3) show an interesting results, whereas the root fresh weight (RFW) for direct seed sowing (DSS) significantly reduced by adding the first NaCl level (50mM) to be 55.6% less than control treatment. Meanwhile, no significant reduction in root fresh weight was recorded at the same NaCl level (50mM) when sugar beet was transplanted (T). However, further increasing on NaCl level to 120mM caused significant decrease of root fresh weight for transplanting. It is clear from the results to mention that the RFW of transplanting at 50 mM NaCl level was two times more than that of RFW recorded by direct seed sowing at the same NaCl level (50mM). Moreover, the RFW value of transplanting recorded at 120 mM was significant more than that recorded at 50mM salinity level for direct seed sowing. The results also indicated that, the RFW for transplanting at 120 mM NaCl level was around 13 times more than the RFWvalues recorded by direct seed sowing at the same NaCl level (120 mM). No further growing was recorded for DSS at 190mM or 260 mM NaCl salinity levels. In case of transplanting the plant survived and grown till the 260 mM NaCl salinity (half of the sea water salinity level) and recorded a root yield higher around four times than that of DSS at 120 mM NaCl salinity level (Table 3).

Table (3): Effect of NaCl salinity levels on root fresh weight (RFW) and leaf area (LA) of sugar beet plants at twelve weeks from sowing date.

Growth parameters	RFW g/plant		LA cm^3/plant	
Sowing method / NaCl levels (mM)	Direct seed sowing (DSS)	Transplanting (T)	Direct seed sowing (DSS)	Transplanting (T)
Control	81^a	84^a	1580^b	1630^a
50	36^c	79a	950^e	1490^c
120	3.4^f	44b	219^h	1240^d
190	-	26^d	-	777^f
260	-	13^e	-	414^g

Mean of each parameter with the same letter are not significant at 5% level

As for leaf area, the results show a significant decrease by increasing NaCl levels for both sowing methods (Table 3). However, the values of leaf area gave similar trend as root fresh weight, whereas the transplanting method was higher than direct seed sowing under all salinity treatments. The same results were obtained for juvenile and adult leaf area (Fig 2). Concerning the leaf succulence the result presented in Table (4) show gradually increases of leaf succulence by adding the first NaCl level (50 mM) for transplanted ranged between 7% and 5% for juvenile and adult leaf respectively. However, increasing NaCl salinity levels up to 260 mM increased leaf succulence to reach 35% and 39% for juvenile and adult leaves as a compared with their control respectively. On the other hand, leaf succulence for DSS was more affected by increasing salinity levels whereas, succulence's for juvenile and adult leaves increased around 15% and 25% more than their controls at 50 mM NaCl level and reached 52% and 69% succulence's for juvenile and adult leaves respectively at 120 Mm NaCl salinity level more than their controls.

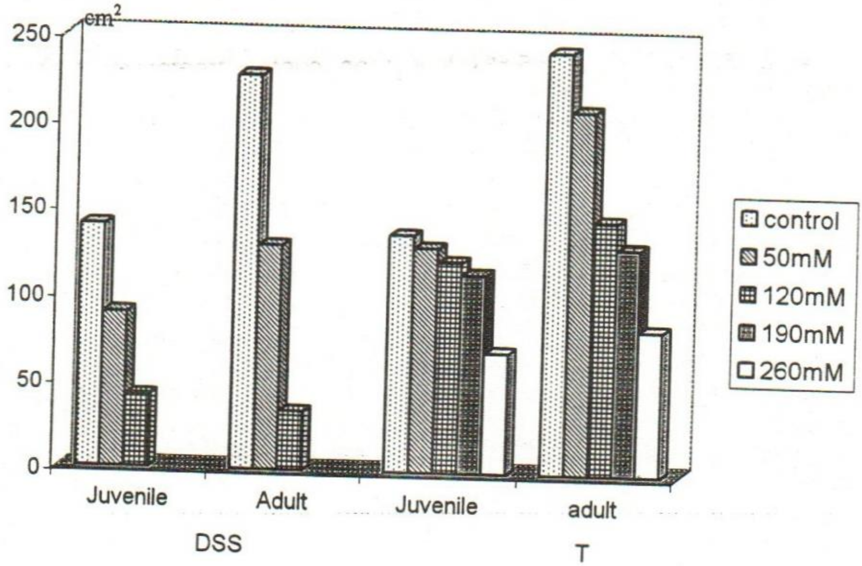

Fig.(2): Effect of NaCl salinity levels (mM) on juvenile and adult leaf area of sugar beet plants at twelve weeks after sowing.

Table (4): Effect of NaCl salinity levels on leaves succulence

Treatments NaCl levels (mM)	Direct seed sowing (DSS)		Transplanting (T)	
	Juvenile	Adult	Juvenile	Adult
Control	2.7	3.9	2.8	4.1
50	3.1	4.9	3.0	4.3
120	4.1	6.6	3.5	5.2
190	-	-	3.4	5.5
260	-	-	3.8	5.7

Chlorophyll content per unit for juvenile and adult leaves is illustrated in Fig. (3). The results clearly show an increase of chlorophyll content for DSS by increasing NaCl level to reach the maximum value at 120mM NaCl for both juvenile and adult leaf. This increase was also detected for (T) until 120 mM NaCl for juvenile leaves and 50 mM for adult leaf, thereafter was decreased by increasing NaCl levels.

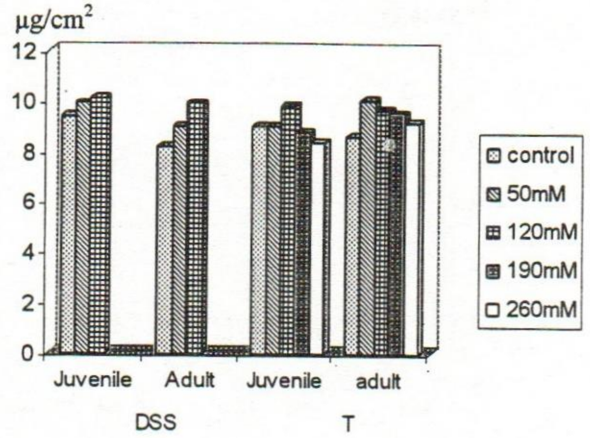

Fig. (3): Effect of NaCl levels on chlorophyll content in juvenile and adult leaves of sugar beet at twelve week from sowing date.

As for K/Na ratio in sugar beet leaf blades, petioles and roots (Fig. 4), it is quite evident that at non NaCl salinity treatment the highest K: Na ratio was observed in root followed by petioles, but K/Na ratio in leaf blades was the lowest and recorded 25% of that obtained in root. This fact was true for both DSS and T. However, adding the first NaCl level (50mM) caused a linearly decrease of K/ Na ratio for all plant parts but further increasing of NaCl levels to 120mM had slow gradual decrease. The aforementioned results were recorded either by direct seed sowing or transplanting. On the other hand, the K/Na ratio for transplanting recorded always a value over one at 50mM NaCl in all plant parts.

Root and sugar yield at harvest stage:

According the sugar beet root yield at harvest stage presented in Table (5), it is clear that no significant different was obtained for root yield between direct seed sowing and transplanting in the control treatment. However, root yield was significantly decreased at 50mM NaCl. This decrease of root yield at the first NaCl level (50mM), compared with the control of DSS, reached around 13% for transplanting but recorded 63%for direct seed sowing. However, the harmful effect of NaCl salinity on root yield was more aggressive by increasing NaCl levels for direct seed sowing than for transplanting. Whereas, the root yield for direct seed sowing decreased around 29 times, at 120mM, NaCl less than control, meanwhile, the root yield for transplanting decreased around two times at the same salinity level as compared with DSS control. As for root sugar yield illustrated in Fig. (5), the results gave almost the same trend as root yield.

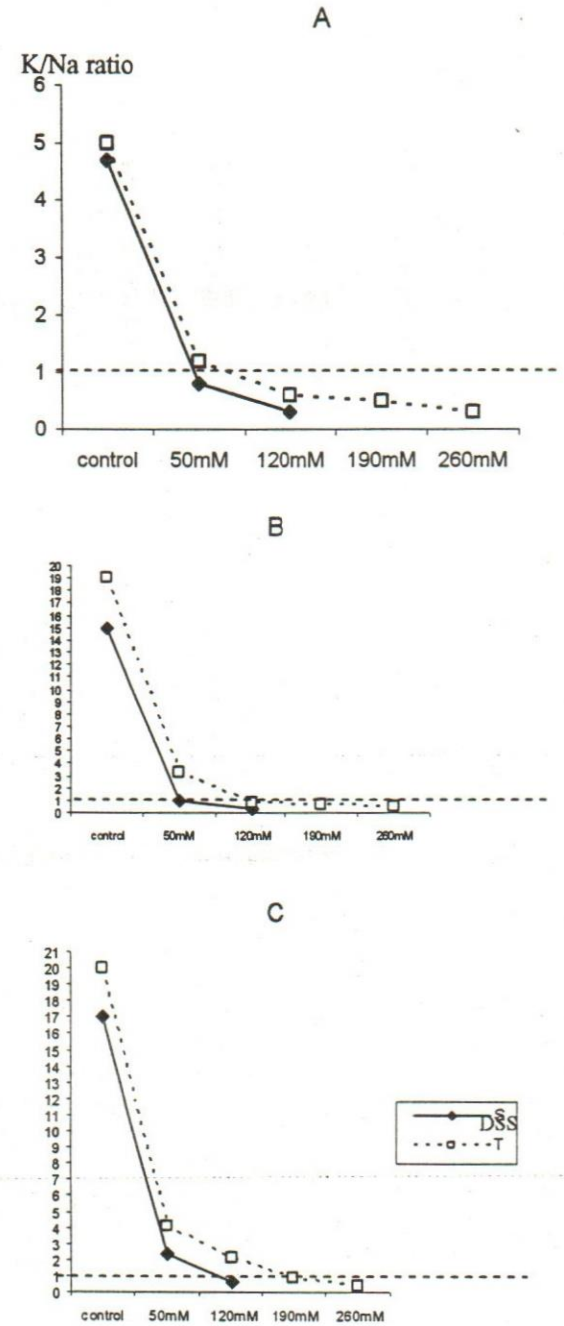

Fig. (4): Effect of NaCl salinity levels on K/Na ratio of sugar beet plant blades (a), petioles (b) and root (c), at twelve weeks from sowing date.

Table (5): Effect of NaCl salinity levels on sugar beet root yield per plant at harvest stage.

Growth parameters	RFW g/plant	
Sowing method	Direct seed sowing (DSS)	Transplanting (T)
NaCl levels (mM)		
Control	487^{A}	510^{A}
50	180^{D}	424^{B}
120	17^{G}	232^{C}
190	_	129^{E}
260	_	60^{F}

Mean of each parameter with the same letter are not significant at 5% level.

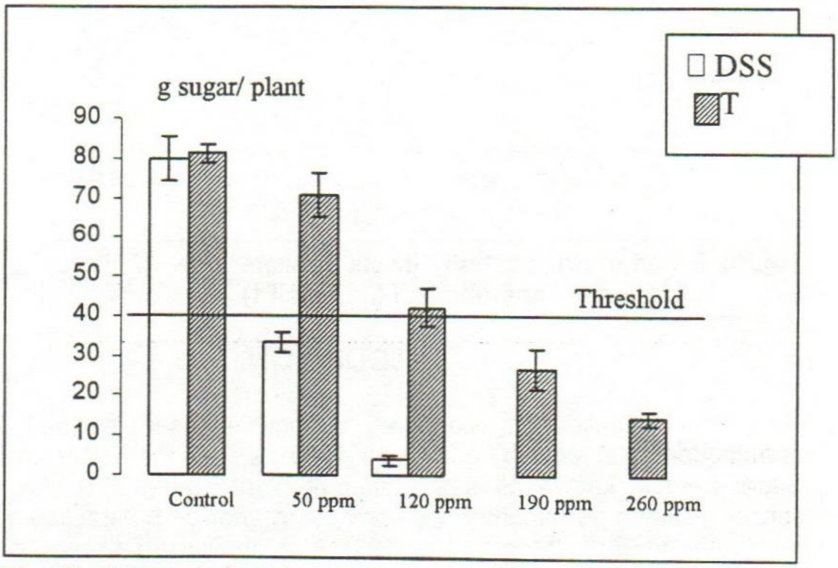

Fig.(5). Effect of NaCl levels on sugar yield per plant at harvest.

The water use efficiency is most important point in this work. Data presented in Fig. (6) Show that the sowing of sugar beet by transplanting obviously increased the efficiency of water use to produce sugar yield as compared with direct seed sowing under all treatments. It is clear from the results under control treatment; transplanting using paper-pot consumed around 51 liter water/plant (after transplanted until the harvest), while direct seed sowing consumed around 65 liter/plant water. Consequently, the water use efficiency (WUF) was obviously higher for transplanting than that of direct seed sowing. Whereas, one liter of water produced 1.6 g sugar for transplanted and 1.2 g sugar for DSS. The above mentioned results mean that reducing of water consumption reached around 21% when the sugar beet was transplanted as compared with direct seed sowing under control treatment. Concerning NaCl salinity, it is generally evident that, water consumption was decreased by increasing NaCl levels. Therefore, the water use efficiency decreases more sharply for direct seed sowing than for transplanting.

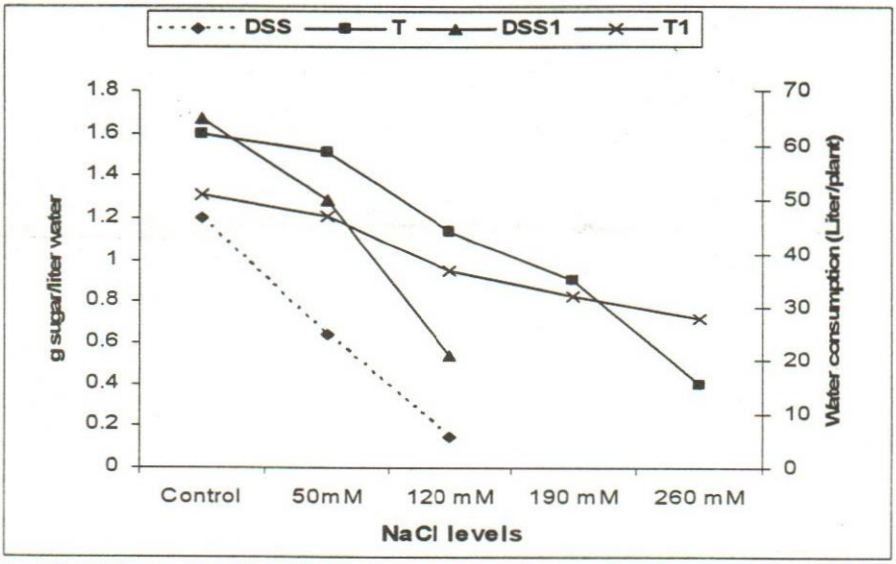

Fig.(6). Effect of NaCl salinity levels on water use efficiency (S and T) or on water consumption (S1 and T1)

DISCUSSION

This investigation monitored changes caused by salt stress on germination rate, growth, and sugar yield as well the water consumption. Sugar beet is known as a salt tolerant plant during most life cycle but sensitive during germination and early stage after crop establishment (25-35 days after sowing), (Marschner, 1995). Also, the results show a decrease of seedling fresh weight and germination percentage by increasing salinity levels. Such conclusion has reviewed by Zapata *et al* (2004). The salt tolerance mechanisms of sugar beet plants have been studied by many investigators (Marshner, 1995; Koyro & Huchzermeyer, 1997; Ali *et al* 2000; Eisa *et al* 2001 and Eisa & Ali, 2005). Sugar beet has an includer mechanism to achieve salt tolerant, whereas the plant has the ability to uptake the salt water and immediately transplanted into shoot. This mechanism facilitates osmotic adjustment but can lead to toxicity and/ or nutritional imbalance. However, the main salt tolerance mechanism for sugar beet is based on its ability to regulate root osmotic potential by accumulation of the soluble sugars in root (Eisa and Ali 2005). So, the rate of soluble sugars accumulated in root play the main role to decrease root osmotic potential against the low osmotic potential in root medium under salinity condition. Therefore, the sugar beet is sensitive during its germination and early growth stage, where there is no or/not enough photosynthesis area to build sugar are created. However, that not the case in late growth stage, when the plant has enough leaves to build sugar and transport into root consequently decreased its osmotic potential.

Accordingly, transplanting the adult plant may be the way to avoid the harmful effect of salinity on sensitive growth stage of sugar beet which finally reflects on yield. In sugar beet as well as in other crops, agronomic characteristics such as yield and also some chemical analysis are most commonly used criteria measuring salt tolerance. This is probably due to their ease of measurement and the fact that yield under saline conditions, is what ultimately matters and more relevant criteria for improving salt tolerance in crop.

The results of growth characters as affected by NaCl salinity at twelve weeks show a great decreases of root fresh weight and leaf area for direct seed sowing, as affect by increasing NaCl levels but that was not the case for transplanting. Whereas, the RFW for transplanting was not significantly affected by adding the first NaCl level (50 mM), meanwhile, it was significantly decreased for direct seed sowing to loss around 63% as a compared to its control. This response can be explained by the role of a total soluble sugar accumulated in root in case of transplanting consequently that lead to uptake salt water which content a high level of Na. However, the uptake Na rapidly translocated into the shoot and there occurred a replacement of K^+ by Na^+ in various metabolic functions. The accumulation of Na in leaves parallel with decreasing K content, may give us an important guide for the reflection of salt stress on yield. Here it might be suggested that, the value of leaves K/Na ratio over one reflected unsaturated leaves from Na and this was concomitant with neglect harmful effect on plant due to toxic or imbalance effects. On the other words, if the leaves K/Na value was over one, that might be an indication for an active includer mechanism and the leaves in this case could uptake more Na. However, increasing the level of NaCl salinity in root medium caused a decrease of leaves K/Na value lower than one and that was although associated with a drastic decline of root and sugar yield. Interesting however, this above indication was only true for K/Na values in leaves, but not for root.

This may be due to the way of transport of Na^+ and K^+ through the vascular system. Since Na^+ translocate to the leaves is to some extent related to transpiration but a considerable net efflux of K^+ takes place in phloem to uploading it into growth sinks (root and young leaves). Therefore, the values of K/Na ratio were higher in root than that of leaves. Similar results were reported by Gorham *et al* (1985); Marschner (1995); Daoud *et al* (2003) and Messedi *et al* (2003).

The injury effect of salinity levels on sugar beet root and sugar yield has been reported by several investigators (Katerji *et al* 2003; Ebrahim, 2005; Almodares & Sharif, 2005 and Shereif *et al* 2005). However, the reasons of decreasing sugar beet yield under considerable salinity levels may be due to osmotic stress reducing leaf area and decreasing chlorophyll contents. The results indicated also decreases of leaf area consonant with increases of leaf succulence and increasing of chlorophyll content due to increasing NaCl levels. This reduction of leaf area is among the mechanisms salt includer species use in order to minimize the evaporating surface (Daoud *et al* 2003 and Koyro & Huchzermeyer, 2004) and consequently increase their water content leading to succulence which is a way to dilute the Na concentration in leaf tissues. However, the increases of chlorophyll content in leaf may be due to the reduction of leaf area and increasing leaf succulence not due to stimulation of chlorophyll formation by NaCl salinity.

Finally, it could be concluded that at 50mM NaCl the paper-pot transplanting has no osmotic stress but the losses of yield may be due to a nutrient imbalance in leaves by uptake the saline water. Raising NaCl levels up to 120 mM lead to osmotic and imbalance nutrition

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تحسين محصول بنجر السكر تحت ظروف الاجهاد الملحى

سيد سعيد شعبان عيسى
قسم النبات الزراعى – كلية الزراعة – جامعة عين شمس

أقيمت تجربة أصص خلال موسمي ٢٠٠١/٢٠٠٢ و٢٠٠٢/٢٠٠٣ لدراسة مقارنة إستجابة نباتات بنجر السكر المنزرعة مباشرة بالبذرة مع تلك المشتولة باستخدام الأصص الورقية عند عمر ٧ أسابيع في تربة رملية وتروى بخمس مستويات من الملوحة بحد أقصى نصف مستوى ماء البحر (كنترول ٥٠، ١٢٠، ١٩٠، ٢٦٠ مليمولر من كلوريد الصوديوم).

أضيفت الملوحة بعد أسبوعين في حالة زراعة المباشرة للبذرة ومباشرة بعد نقل الشتلة للأصص في حالة الشتل.

أظهرت النتائج التأثر الشديد لمحصولي الجذور والسكرعند المستوى الأول للملوحة (٥٠ مليمولر) في حالة الزراعة المباشرة بالبذرة حيث انخفض محصول الجذور بمقدار ٦٣% ومحصول السكر بمقدار ٥٨% مقارنة مع الكنترول بينما سجل هذا الانخفاض ١٣% في محصول الجذور و١٠% في محصول السكر عند الشتل مقارنة مع الكنترول.

ارتفاع مستوى الملوحة في ماء الرى أدى إلى الانخفاض الشديد في محصولي الجذور والسكر في حالة الزراعة المباشرة بالبذرة حيث سجل الفقد في محصول الجذور ٩٦% وفي محصول السكر ٩٥% مقارنة بالكنترول. وفشلت تماماً النباتات المنزرعة بالبذرة في النمو تحت مستوى ملوحة مقداره ١٩٠ مليمولر. بينما استمرت النباتات في النمو حتى ٢٦٠ مليمولر في حالة الشتل.

بالرغم من عدم وجود فرق معنوي تحت ظروف الكنترول (بدون ملوحة) في محصول الجذور والسكر بين الزراعة بالبذرة أو الشتل إلا أن كفاءة استهلاك الماء في حالة الشتل كانت أفضل حيث انخفض استهلاك الماء حوالي ٢١% أقل من الزراعة بالبذرة مباشرة.

دراسة العلاقات الفسيولوجية أظهرت انخفاض حاد في نسبة البوتاسيوم إلى الصوديوم في أنسجة نصل الورقة والعنق وكذلك الجذور تحت معاملات الملوحة، إلا أنه في حالة الشتل وعند المستوى الأول من الملوحة (٥٠ مليمولر) ظل محتوى البوتاسيوم أعلى من محتوى الصوديوم في كل الأجزاء النباتية السابقة (قيمة النسبة أعلى من الواحد الصحيح).

ازداد محتوى الكلوروفيل لكل ١ سم^{2} من الأوراق الحديثة الناضجة إلا أن هذه الزيادة راجعة إلى الانخفاض الشديد في مساحة الأوراق وزيادة العصرية للأوراق متأثرة بالملوحة.