

SALINITY EFFECT ON PLANT GROWTH, PSII PHOTOCHEMISTRY AND CHLOROPHYLL CONTENT IN SUGAR BEET (*BETA VULGARIS* L.) AND CABBAGE (*BRASSICA OLERACEA CAPITATA* L.)

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Abstract

Seedlings of sugar beet (*Beta vulgaris* L.) and cabbage (*Brassica oleracea capitata* L.) were grown in sand culture at salinities of 0 (control), 50, 100 and 150 mM NaCl to determine the effect of salt on growth, chlorophyll fluorescence and chlorophyll content. With increasing salt concentration, dry root and shoot weight, fresh leaf weight and leaf area decreased significantly whereas there were no changes in dry leaf weight and leaf water contents. Salinity induced no effects in both species on the maximal efficiency of PSII (F_v/F_m) photochemistry, efficiency of excitation energy capture by open PSII reaction centres, electron transport rate (ETR), photochemical quenching coefficient (q_p), non-photochemical quenching coefficient (q_n) and physiological state of the photosynthetic apparatus (F_o/F_m). However, leaf chlorophyll content increased significantly with increasing salt concentration both in cabbage and sugar beet.

Introduction

Photosystem II (PSII) is believed to play a key role in the response of photosynthesis to environmental perturbations (Baker, 1991). The effects of salinity stress on PSII have been studied extensively. However, the data on the effects of salinity stress on PSII photochemistry are conflicting. Some studies have shown that salt stress could inhibit PSII activity (Hasegawa *et al.*, 2000; Munns, 2002; Ashraf & Shahbaz, 2003; Ashraf, 2004) while others have indicated that salinity has no effect on this parameter (Robinson *et al.*, 1983; Morales *et al.*, 1992; Abadía *et al.*, 1999).

It has been reported that there were no changes in PSII photochemistry in salt-stressed cowpea, wheat, sorghum and barley when they were grown under relatively low light conditions (Morales *et al.*, 1992; Lu & Zhang 1998), suggesting that salt stress has no effects on PSII photochemistry and that it is the interaction between high light and other environmental stresses which result in damage to PSII (Larcher *et al.*, 1990; Masojidek *et al.*, 1991). Some studies have shown that salt stress led to no effects on PSII photochemistry if salt stress treatments were carried out under relatively low light, but predisposed to photoinhibition when salt-stressed plants were exposed to high light (Masojidek & Hall, 1992).

Leaf photosynthetic capacity depends on physiological characteristics such as chlorophyll contents, Rubisco activity and photosystem efficiency (Flore & Lakso, 1989; Bowes, 1991). It has been reported that chlorophyll content decreases in salt susceptible plants such as tomato (Lapina & Popov, 1970) and pea (Hamada & El-Enany, 1994), but that chlorophyll content increased in salt tolerant plants such as pearl millet (Reddy & Vora, 1986) and mustard (Singh *et al.*, 1990).

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In view of the contrasting reports mentioned earlier the objective of the present study was to evaluate the effect of salinity on growth, photosystem II efficiency and chlorophyll content in sugar beet (*Beta vulgaris* L.) and cabbage (*Brassica oleracea capitata* L.) and to assess whether these parameters bear a positive or negative relationship with the growth of the two crops under salt stress.

Materials and Methods

Plant material and salinity treatments: Seedlings of sugar beet (*Beta vulgaris* L.) and cabbage (*Brassica oleracea capitata* L.) were grown in plastic pots (16 cm diameter, 22 cm height) filled with sand and watered with full-strength Hoagland (Hoagland & Arnon, 1950) nutrient solution. After 3 weeks, the seedlings were subjected to salt treatment. Salt concentrations in Hoagland nutrient solution were 0 (control), 50, 100 and 150 mM NaCl. All measurements on the youngest and expanded leaves were made after 6 weeks, when plants had achieved a steady state. The average temperature for day/night was 25/15 °C and photoperiod for the day/night cycle 16/8 h.

Measurements of chlorophyll fluorescence: Chlorophyll fluorescence was measured in attached dark adapted leaves with a portable Mini PAM fluorometer (PAM-2000, Walz, Germany). The data acquisition software (DA- 2000, Walz) was used to connect the fluorometer with a computer. The experimental protocol of Genty *et al.*, (1989) was basically used in this experiment. The minimal fluorescence level (F_o) in the dark-adapted state was measured by the measuring modulated light, which was sufficiently low ($<0.1 \mu\text{mol m}^{-2} \text{s}^{-1}$) not to induce any significant variable change in fluorescence. The maximal fluorescence level (F_m) was measured by a 0.8 s saturating pulse at $8000 \mu\text{mol (photon) m}^{-2} \text{s}^{-1}$. The measurements of F_o were recorded with the measuring beam set to a frequency of 0.6 kHz, whereas F_m measurements were performed with the measuring beam automatically switching to 20 kHz during the saturating flash. The leaves of both species were continuously illuminated with white actinic light at an intensity of $300 \mu\text{mol (photon) m}^{-2} \text{s}^{-1}$. The following calculations were made by using fluorescence parameters determined in leaves:

- (1) The maximal quantum yield of PSII photochemistry (F_v/F_m)
- (2) The photochemical quenching coefficient, $q_p = (F_m' - F) / (F_m' - F_o')$ (F_m' here represents the maximal fluorescence measured by a saturation pulse in given light state whereas F_m' and F_o' are the values measured after dark adaptation) and the non-photochemical quenching coefficient $q_N = 1 - (F_m' - F_o') / (F_m - F_o)$
- (3) The efficiency of excitation capture by open PSII (F_v'/F_m')
- (4) The physiological state of the photosynthetic apparatus (F_o/F_m) and
- (5) The electron transport rate (ETR) ($ETR = \text{yield} \times QFD \times 0.5 \times 0.84$) (the standard factor 0.84 corresponds to the fraction of incident light absorbed by leaf).

Measurements of leaf area and leaf water content: The leaf area of individual plant leaves were measured by using an Area meter (AM-200, ADC Bio-Scientific Ltd., England). After leaf area measurement, the fresh weight of leaves was determined. Root, shoot and leaves were oven dried at 80 °C for two days to determine the dry weight. The leaf water content was calculated as $(FW - DW) / DW \times 100$, where FW is leaf fresh weight and DW is leaf dry weight.

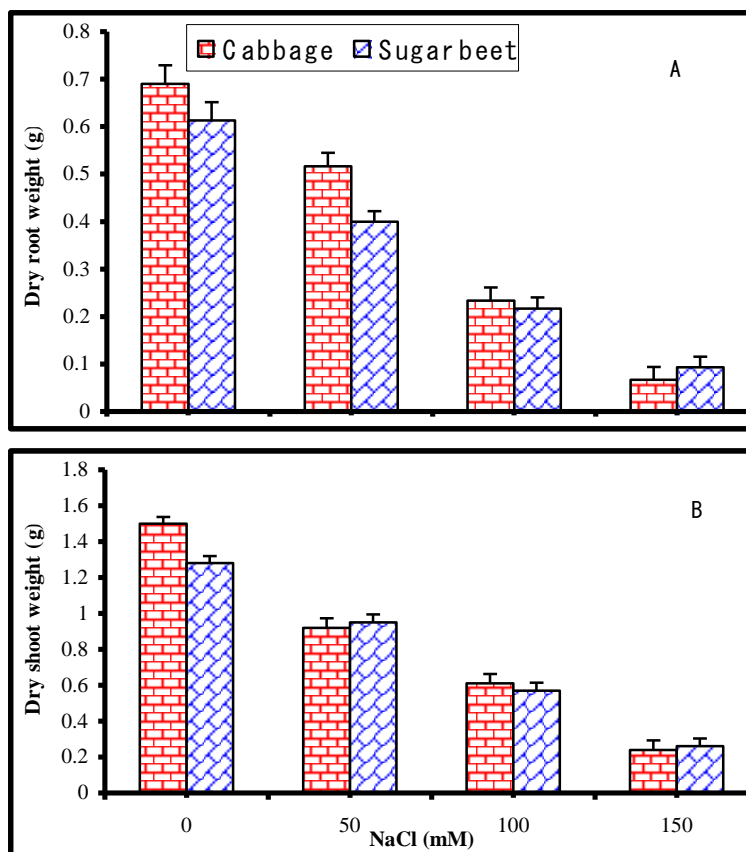


Fig. 1. Effect of salinity treatments on root dry weight (A) and shoot dry weight (B) of cabbage and sugar beet. Values are the means \pm SD of three replications. (SD: standard deviation)

Leaf chlorophyll measurement: Chlorophyll content meter (CCM-200, Opti-Science, USA) was used to measure leaf chlorophyll content. The chlorophyll content was measured 8 times from leaf tip to the leaf base and then averaged for analysis.

Statistical analysis: Analysis of variance was performed by using MS-Excel Statistical software. Mean values for growth, photosynthesis parameters, chlorophyll content, leaf area and leaf water content were compared by LSD using Tukey's *t* test (Li, 1964).

Results

Salinity effect on plant growth: The growth of sugar beet and cabbage was significantly decreased by salt treatment. Root and shoot dry weight decreased significantly with the increase in salt concentration up to 150 mM NaCl. (Fig. 1), and shoot dry weight was reduced more than root dry weight. Dry weights of root and shoot were more reduced in cabbage than in sugar beet at 150 mM NaCl (Fig. 1).

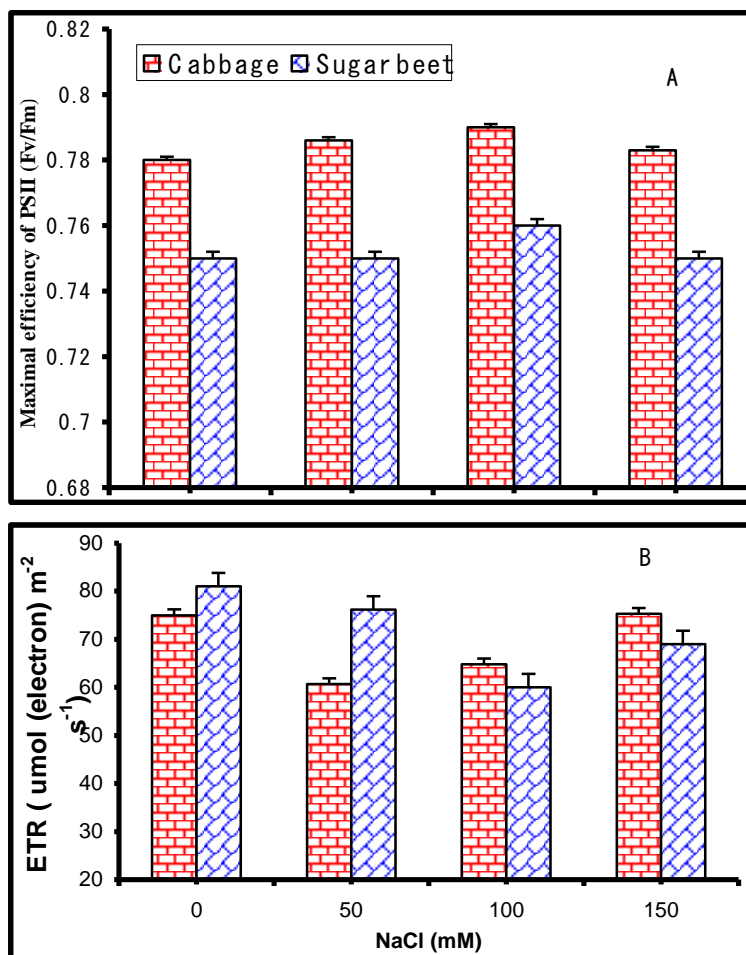


Fig. 2. Effect of salinity treatments on maximal quantum yield of PSII (F_v/F_m) (A) and electron transport rate (ETR) (B) in sugar beet and cabbage. Values are the means \pm SD of three replications.

Salinity effect on chlorophyll fluorescence: The changes in PSII photochemistry were first studied in dark adapted leaves by using the Mini PAM fluorometer. Fig. 2A shows that no changes occurred in the maximal quantum yield of PSII (F_v/F_m) measured in the dark adapted leaves between control plants and salt-stressed plants in both species.

The results shown above indicated that salinity had no effects on the primary photochemistry of PSII in the dark adapted leaves. It was investigated further whether salinity induced any significant change in light adapted leaves. The changes in fluorescence characteristics in light adapted leaves were observed by use of pulse modulated fluorescence. Salinity had no significant effects on the efficiency of excitation energy capture by open PSII reaction centres (F_v'/F_m'), the photochemical quenching coefficient (q_p) and non-photochemical quenching coefficient (q_N) as well as physiological state of the photosynthetic apparatus (F_o/F_m) (Figs. 3 and 4). No significant changes in ETR were observed in cabbage and sugar beet with salinity.

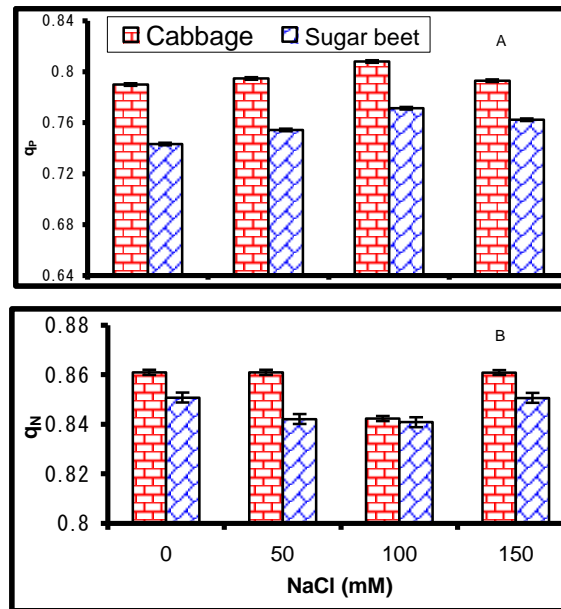


Fig. 3. Effect of salinity treatments on photochemical quenching coefficient (q_p) (A) and non-photochemical quenching coefficient (q_n) (B) in sugar beet and cabbage. Values are the means \pm SD of three replications.

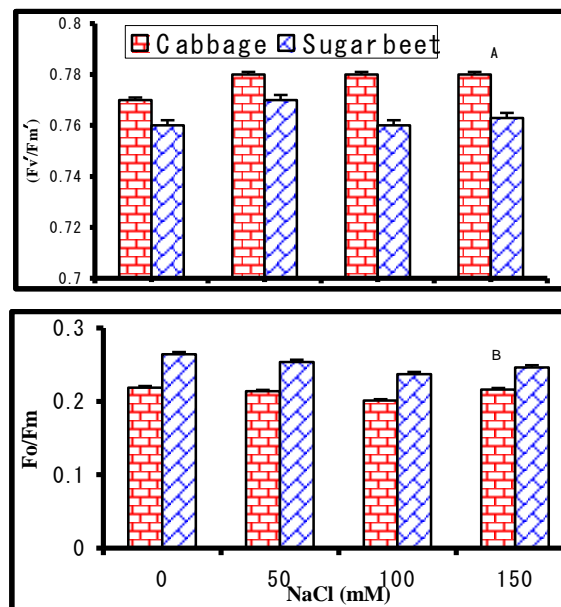


Fig. 4. Effect of salinity treatments on efficiency of excitation capture by open PSII reaction (F_v'/F_m') (A) and physiological state of the photosynthetic apparatus (F_o/F_m) (B) in sugar beet and cabbage. Values are the means \pm SD of three replications.

Table 1. Effect of salinity treatments on leaf area, leaf water content and chlorophyll content (SPAD value) of cabbage and sugar beet.

Genotypes	NaCl (mM) (%)	Leaf area (cm ²)	Leaf water content	Chlorophyll Content (SPAD Value)
Cabbage	0	125 a	90 a	12.55 a
	50	123 ab	91 a	23.32 b
	100	77 b	91 a	24.12 c
	150	35 c	93 a	33.48 d
Sugar beet	0	98 a	88 a	37.11 a
	50	90 b	91 a	37.73 a
	100	70 c	89 a	39.28 b
	150	62 d	85 a	40.73 bc

Mean values with different letters within each of the growth parameters are significantly different ($p < 0.05$).

Salinity effect on leaf area, leaf water content and leaf chlorophyll content: Significant decreases in number of leaves (data not shown) and leaf area were observed in both plant species. Maximum leaf area reduction was observed in cabbage (Table 1). However, salinity had non-significant effects on leaf water contents. A significant increase in leaf chlorophyll content was observed in both plant species and the maximum increase was measured in cabbage as compared to that in sugar beet (Table 1).

Discussion

It has been reported that salt stress alone had no effect on PSII photochemistry at relatively low light but induced photodamage to PSII when salt-stressed leaves was exposed to high light (Robinson *et al.*, 1983; Morales *et al.*, 1992; Abadía *et al.*, 1999). The lack of changes in the maximal efficiency of PSII photochemistry (F_v/F_m) in control and salt-stressed plants suggests that salt stress had no effects on PSII primary photochemistry in cabbage and sugar beet. Although there was a slight variation in F_v/F_m in control and salt-stressed plants, no significant differences in this parameter were observed between control and salt-stressed plants (Fig. 2A). The results show that there were no large differences in F_v'/F_m' , ETR , q_P , q_N as well as F_o/F_m between control and salt-stressed plants both in sugar beet and cabbage (Fig. 3 and 4). It seems that the effects of salt stress on PSII photochemistry in plants depend on light intensity. It has been shown that the PSII photochemistry of barley did not change significantly in response to salt stress when grown under low light (Morales *et al.*, 1992). On the other side, some studies have showed that salinity in the presence of high light induced significant changes in PSII photochemistry in sorghum (Masojidek *et al.*, 1991). Similar results were found in *Phaseolus vulgaris* (Brugnoli & Lauteri, 1991) and spinach leaves (Downton *et al.*, 1985) in response to salinity.

Salt stress induces a significant decrease in dry weights of root and shoot and leaf area, but it had no significant effect on leaf water contents (Fig. 1 and Table 1). Plant growth is ultimately reduced by salinity stress, although plant species differ in their tolerance to salinity (Munns & Termaat, 1986). Reduction of plant growth under saline conditions is a common phenomenon (Ashraf & Harris, 2004) but such reduction occurs differently in different plant organs. For example, in the present experiment, shoot dry weight was reduced more than root dry weight by salt stress. Similar kind of results were earlier reported by Jamil *et al.*, (2005). They observed that salt stress inhibited the growth of shoot more than root in *Brassica* species. Salt stress reduces leaf growth rate by

shortening the length of the leaf elongating zone and decreasing the growth intensity in its central and distal portions (Bernstein *et al.*, 1993). Greenway & Munns (1980) reported that the effect of salinity on leaf area was greater than on dry weight, as salt accumulation in the shoot occurs via transpiration stream, which is highest in old leaves killing them. These results are not in agreement with those of Meena *et al.*, (2003) who found that leaf water content decreased significantly with increasing salinity.

An increase in chlorophyll content (Table 1) has been thought to be due to the accumulation of NaCl in the chloroplast (Kirst, 1989). Misra *et al.*, (1997) also concluded that salt stress induced an increase in the chlorophyll content, which could be due to an increase in the number of chloroplasts in stressed leaves. It has been reported that chlorophyll content decreases in salt susceptible plants such as tomato (Lapina & Popov, 1970) and pea (Hamada & El-Enany, 1994), but increases in salt tolerant plants such as pearl millet (Reddy & Vora, 1986) and mustard (Singh *et al.*, 1990).

Our results indicate that cabbage and sugar beet showed tolerance of PSII to high salinity stress and also increased chlorophyll contents, even when treated with salinity as high as 150 mM NaCl. It is suggested that tolerance of PSII to high salinity stress and low light stress can be viewed as an important strategy for cabbage and sugar beet to grow in very high saline soils.

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