Research Article

Effect of salt stress on some morphological and biochemical characteristics of sunflower (Samsung-600)

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Abstract

Salinity in the growth medium causes significant reduction in plant growth parameters like leaf area, leaf length, root and shoot dry weight etc. The present study was conducted to assess the effect of salt stress on morphological and biochemical characteristics of sunflower. The hybrid (Samsung-600) seeds were grown under normal and saline conditions. The plants were treated with different doses of sodium chloride. Salt stress significantly reduced plant height, number of leaves, fresh and dry plant weight. Salinity increased the moisture in seeds. The moisture percentage increased with increase in NaCl treatment. Salt stress increased the ash contents in the seeds. Similarly, salinity stress considerably increased the accumulation of proteins contents and fats in the seeds but caused reduction in crude fibers percentage. Carbohydrates also showed a decreased tendency with increase in NaCl concentration.

Key words: Morphological; Biochemical; Characteristics; Salt stress; Sunflower

Introduction

Sunflower makes up the genus *Helianthus*. In Greek *Helios* means Sun and *annus* means flower. The genus, which contains about 67 species, is thought to be native to the Americas (North, South) originally, and was domesticated around 1000 BC. Although, Sunflower is now distributed almost worldwide. There are two types grown. The first is Oilseed. This small black seed is very high in oil content and is processed into sunflower oil and meal. It is also the seed of choice of most bird feeders. The second type is Non-oilseed (confectionery sunflower). This is a larger

black and white striped seed used in a variety of food products from snacks to bread. There are three types of sunflower oil available; Mid-Oleic, Linoleic and High Oleic sunflower oil. All are developed with standard breeding techniques. They differ in oleic levels and each one offers unique properties. With three types of sunflower oil available, sunflower oil meets the needs of consumer and food manufacturers alike for a healthy and high performance non-transgenic vegetable oil. Confection sunflower seeds are normally black with white stripes and provide consumers with in-shell seeds and kernel. In-shell means

the seed is left intact with the "meat" of the seed still in the shell. It is normally roasted and seasoned. High amounts of salts whether from soil or water are great problems to agriculture throughout the world [1, 2]. In many regions of the world, salinity stress may occur when crops are exposed to level of salt (Na^+ and Cl^-). The effect of sodium and chloride salts may be greater than those of other salts present in the soil or water [3, 4].

Materials and methods

The effect of salt stress on Sunflower hybrid (Samsung 600) was studied by performing an experiment in the botanical garden Department of Botany, Islamia College Peshawar. Seeds were sown in thirty pots. The plastic bag was kept in each pot. Then 5kg soil was taken in each pot. The experiment was conducted in a completely randomized design with over all five treatments. Each treatment had five replicates. Ten seeds were sown in each pot. The germination started at 21/3/2011 and completion date was 24/3/2011. It was the two leaf stage. At fourth leaf stage, uprooted few plants and weighed. The different doses of sodium chloride were given to the plants. After fifteen days of treatment the data was noted. The second treatment of different doses was given in the same manner. Second data was noted after 15 days of second treatment. The third data was noted at harvested stage.

Morphological characters:

Different treatments of sodium chloride given to sunflower affected morphological characters. It reduced the plant height and number of leaves. It also affected the fresh and dry weight of the plant. The low doses of NaCl showed less affects on morphological characters as compared to high doses which showed negative effects. It also reduced the head diameter and achenes number. All the collected data were analysed using LSD for comparison of means [3].

Statistical analysis

The experiment was set up in randomized complete block design with six replicates

per treatment. Data were statistically analyzed using ANOVA of Statistic 6 software, the significance of differences among means was carried out using the Least Significant Test (LSD) at $p = 0.05$. Analysis of variance was performed to examine the significant effect of all the parameters measured.

Results and discussion

The data showed significant result in case of number of leaves (Table 1). The control data for leaves showed non-significant result with 1.17g and 2.34g treatments but showed the significant result with 4.68g, 9.36g and 18.72g). Control data for plant height showed non-significant result with 1.17g, 2.34g and 4.68g while showed a significant result with 9.36g and 18.72g. The control data of plant fresh weight showed non-significant result with 1.17 g and 2.34g but showed significant result with 4.68g and 9.36g and 18.72g treatments. The control data of dry weight showed non-significant result with 1.17g and 2.34g but significant result with 4.68g, 9.36g and 18.72g treatments. The overall result showed that the leaves are highly susceptible to salt stress (Table 1) than other parameters of the test plant.

In case of number of leaves, the 1.17g data showed non-significant result with control, 2.34g and 2.34g but significant with 4.68g and 9.36g treatments. The height of plant of the 1.17g treatment showed a non-significant result with control, 2.34g and 4.68g treatments but showed a significant result with 9.36g and also showed a more significant result with 18.72g treatment.

The fresh weights of 1.17g treatment showed a non-significant result with control and 2.34g but it showed a significant result with 4.68g, 9.36g and 18.72g treatments. The dry weight of 1.17g treatment showed a non-significant result with control and 2.34g. 1.17g treatment showed a non-significant result with 4.68g, 9.36g and 18.72g treatments. In case of number of leaves, 2.34g treatment showed a non-significant result with control and 1.17g treatment. But 2.34g showed a more

significant result with 4.68g, 9.36g and 18.72g treatments. Plant height of 2.34g treatment showed a non-significant result with control, 1.17g, 4.68g and 9.36g treatments. But it showed more significant result with 18.72g dose.

The fresh weight of 2.34g treatment showed non-significant result with control and 1.17g, 4.68g and 9.36g but showed a more significant result with 18.72g dose. Number of leaves of 4.68g treatment showed a significant result with control, 1.17g, 2.34g and 18.72g treatments and showed a non-significant result with 9.36g treatment.

The plant height of 4.68g showed a significant result with only 18.72g treatment but showed a non-significant result with all other treatments. The fresh weight of 4.68g showed a significant result with control and 1.17g treatment but showed a non-significant result with 2.34g, 9.36g and 18.72g. The dry weight of 4.68g treatment showed significant result with control and 1.17g treatment. It showed a non-significant result with 2.34g and 9.36g treatments. It also showed more non-significant result with 18.72g treatment. Number of leaves of 9.36g treatment showed a significant result with control, 1.17g and 2.34g treatments but showed a

non-significant result with 4.68g and 18.72g treatment. Plant height of 9.63g showed a significant result with control, 1.17g and 2.34g treatment but showed a non-significant result with 4.68g and 18.72g treatment. In case of fresh weight 9.36g showed a significant result with control and 1.17g but showed a non-significant result with 2.34g and 4.68g and it also show a non-significant result with 18.72g treatment. The dry weight of 9.36g showed significant result with control and 1.17g treatment. It also showed a good significant result with 18.72g but showed a non-significant result with 2.34g and 4.68g treatments. The number of leaves of 18.72g showed a good significant result with control, 1.17g and 2.34g treatments but showed a non-significant result with 4.68g and 9.36g treatments. Plant height of 18.72g showed a significant result with the control, 1.17g, 2.34g and 4.68g treatments but showed a non-significant result with 9.36g treatment. The fresh weight of 18.72g showed a significant result with control 1.17g and 2.34g treatment but showed a non-significant result with 4.68g and 9.36g treatments. The dry weight of 18.72g showed a significant result control, 1.17g, 2.34g and 9.36g treatments but showed a non-significant result with 4.68g.

Table 1. Data after first treatment

Treatment	Number of leaves	Plant height	Plant fresh weight	Plant dry weight
T1-Control	9.600 A	14.000 A	8.948 A	1.488 A
T2-1.17g	8.400 AB	14.000 A	8.628 A	1.214 A
T3-2.34g	9.200 A	13.400 AB	8.322 AB	1.054 AB
T4-4.68g	7.600 BC	11.000 AB	5.426 BC	0.684 BC
T5-9.36g	7.600 BC	10.200 BC	5.428 BC	0.696 B
T6-18.72g	6.400 C	7.400 C	4.118 C	0.600 C

Figure 1. Data after 1st treatment

Table-2 shows data after second treatment. Control data number of leaves showed significant result with 19.72g treatment and non-significant result with all the treatments except 19.72g. Plant height of control showed a non-significant result with 1.17g treatment. It also showed a non-significant result with 2.34g and 4.68g treatments but showed a significant result with 9.36g 18.72g treatments. The fresh weight of control showed significant result with 18.72g and showed a non-significant result with all other treatments. The dry weight of control showed significant result with 1.17g, 2.34g, 4.68g, 9.36g and 18.72g treatments. In case of number of leaves, 1.17g treatment showed a non-significant result with all treatments except 18.72g. Plant height of 1.17g treatment showed a non-significant result with control, 2.34g and 4.68g treatment. It showed a significant result with 9.36g and 18.72g treatments. The fresh weight of 1.17g treatment showed a non-significant result with all treatments but show a significant result with 18.72g. In case of dry weight 1.17g treatment showed a significant result with all the treatments. The 2.34g treatment in case of number of leaves showed non-significant result with control, 1.17g, 4.68g and 9.36g treatments but showed a significant result with 18.72g. The plant

height of 2.34g treatment showed a non-significant result with control, 1.17g and 4.68g treatments but showed a significant result with 9.36g and 18.72g. The fresh weight of 2.34g showed a non-significant result with all the treatments except it only showed a significant result with 18.72g treatment. The dry weight of 2.34g showed a non-significant result only with 4.68g. It showed significant result with control, 1.17g, 9.36g and 18.72g treatments. In case of leaves, 4.68g treatment showed a non-significant result with all treatments except 18.72g which showed a significant result. In case of plant height, 4.68g treatment showed a non-significant result with control, 1.17g, 2.34g and 9.36g treatments but showed a significant result with 18.72g. The fresh weight of 4.68g treatment showed a non-significant result with all the treatments. Dry weight of 4.68g treatment showed a significant result with control and 1.17g, 9.36g and 18.72g treatments but showed a non-significant result with 2.34g treatment. In case of leaves, 9.36g treatment showed a non-significant result with all the treatments. Plant height of 9.36g treatment showed a significant result with control, 1.17g and 2.34g and 18.72g treatments but only showed non-significant result with 4.68g treatment. The fresh weight of 9.36g treatment showed non-significant result with control, 1.17g, 2.34g,

4.68g and 18.73g treatments. The dry weight of 9.36g treatment showed non-significant result only with 18.72g treatment but showed more significant result with all other treatments. The number of leaves of 18.72g treatment showed a significant result with 1.17g, 2.34g and 4.68g treatments while it showed a non-significant result with control and 9.36g treatment. In case of plant height, 18.72g

treatment showed a significant result with the rest of treatments. Fresh weight of 18.72g treatment showed a significant result with control, 1.17g and 2.34g treatments while it showed a non-significant result with 4.68g and 9.36g treatments. The dry weight of 18.72g showed a significant result with control, 1.17g, 2.34g and 4.68g treatments and non-significant result with 9.36g treatment.

Table 2. Data after second treatment

Treatment	Numbers of leaves	Plant height	Plant fresh weight	Plant dry weight
T1-Control	14.400AB	44.200A	74.280A	8.040A
T2-1.17g	14.800A	44.600A	73.140A	6.240B
T3-2.34g	15.200A	43.600A	64.860A	4.240C
T4-4.68g	15.200A	42.400AB	56.240AB	4.280C
T5-9.36g	12.800AB	35.200B	57.680AB	2.320D
T6-18.72g	12.00B	22.200C	37.480B	1.180D

Figure 2. Data after 2nd treatment

Table-3 shows data after harvest. In control, numbers of leaves showed non-significant result with all the treatments. Plant height in control showed a non-significant result with 1.17g treatment. Control plant height showed a significant result with 2.34g, 4.68g 9.36g and 18.72g treatments. Fresh weight of control showed a non-significant result with 1.17g treatment and showed significant result with 2.34g, 4.68g and

9.36g treatments. Control fresh weight showed more significant result with 18.72g treatment. Control dry weight showed significant result with all other treatments. The number of achenes of control showed non-significant result with 1.17g and 2.34g treatments a more significant result with 4.68g, 9.36g and 18.72g treatments. Head diameter of control showed a non-significant result with 2.34g treatment.

Control head diameter showed a significant result with 1.17g, 4.68g, 9.36g and 18.72g treatments.

In case of number of leaves, 1.17g treatment showed a non-significant result with rest of the treatments. Plant height of 1.17g treatment showed a non-significant result with control and 2.34g treatment. It showed significant result with 4.68g, 9.36g and 18.72g treatments. The fresh weight of 1.17g treatment showed a non-significant result with control while it showed a significant result with 2.34g, 4.68g, 9.36g and 18.72g treatments. In case of dry weight, 1.17g treatment showed a significant result with control and non-significant result with rest of the treatments. Head diameter of 1.17g treatment showed a significant result with control and 2.34g treatments while it showed a non-significant result with 4.68g, 9.36g and 18.72g treatments. Number of achenes of 1.17g treatment showed a significant result with 9.36g and 18.72g treatments while it showed a non-significant result with control, 2.34g and 4.68g treatments. In case of number of leaves, 2.34g treatment showed a non-significant result with rest of the treatments. The plant height of 2.34g treatment showed a significant result with control but it showed a non-significant result with rest of the treatments. Fresh weight of 2.34g treatment showed a significant result with other treatments. Dry weight 2.34g treatment showed a significant result with control and 18.72g treatments and non-significant result with 1.17g, 4.68g and 9.36g treatments. Head diameter of 2.34g treatment showed a non-significant result with control while showed a significant result with 1.17g, 4.68g, 9.36g and 18.72g treatments. Achene number of 2.34g showed a non-significant result with control, 1.17g, 4.68g and 9.36g treatments while it showed a significant result with 18.72g treatment. Number of leaves of 4.68g treatment showed a non-significant result with rest of the treatments. Plant height of 4.68g treatment showed a significant result with control and 1.17g

treatments and non-significant result with 2.34g, 9.36g and 18.72g treatments. Fresh weight of 4.68g treatment showed a significant result with control, 1.17g, 2.34g and 18.72g treatments while it showed a non-significant result with 9.36g treatment. Dry weight of 4.68g treatment showed a significant result with control while it showed a non-significant result with rest of the treatments. Head diameter of 4.68g treatment showed a significant result with control and 2.34g treatments and a non-significant result with 1.17g, 9.36g and 18.72g treatments. Number of achene of 4.68g treatment showed a significant result with control and 18.72g treatments while it showed a non-significant result with 1.17g, 2.34g and 9.36g treatments. In case of number of leaves, 9.36g treatment showed a non-significant result with other treatments. Plant height of 9.36g treatment showed a significant result with control and 1.17g treatment while showed a non-significant result with 2.34g, 4.68g and 18.72g treatments. Fresh weight of 9.36g treatment showed a significant result with control, 1.17g, 2.34g and 18.72g treatments while it showed a non-significant result with 4.68g treatment. Dry weight of 9.36g treatment showed a significant result with control but it showed a non-significant result with 1.17g, 2.34g, 4.68g and 18.72g treatments. In case of head diameter, 9.36g treatment showed a significant result with control and 2.34g treatments while it showed a non-significant result with 1.17g, 4.68g and 18.72g treatments. Number of achenes of 9.36g treatment showed a significant result with control and 1.17g treatment while it showed a non-significant result with 2.34g, 4.68g and 18.72g treatments. In case of number leaves, 18.72g treatment showed a non-significant result with rest of the treatments. Regarding the plant height, 18.72g treatment showed a significant result with control and 1.17g treatments while it showed a non-significant result with 2.34g, 4.68g and 9.36g treatments. Fresh weight of 18.72g treatment showed a significant result with

control, 1.17g, 2.34g, 4.68g and 9.36g treatments. Dry weight of 18.72g treatment showed a significant result with control and 2.34g treatments while it showed a non-significant result with 1.17g, 4.68g and 9.36g treatments. Head diameter of 18.72g treatment showed a significant result with

control and 2.34g treatments while showed a non-significant result with 1.17g, 4.68g and 9.36g treatments. In case of achenes number, 18.72g treatment showed a significant result with control, 1.17g, 2.34g and 4.68g treatments while showed a non-significant result with 9.36g treatment.

Table 3. Data after harvest

Treatment	Numbers of Leaves	Plant height	Plant fresh weight	Plant dry weight	Head diameter	Achene's number
T1-Control	29.20 A	4.660 A	202.800 A	83.562 A	14.0 A	980.000 A
T2-1.17g	28.80 A	4.366 AB	182.800 A	52.702 BC	10.40 B	795.000 AB
T3-2.34g	85.60 A	3.734 BC	153.100 B	61.286 B	12.80 A	693.333 ABC
T4-4.68g	27.60 A	3.748 C	106.172 C	53.602 BC	10.90 B	648.333 BC
T5-9.36g	25.80 A	3.500 C	106.240 C	45.222 BC	9.60 B	460.00 CD
T6-18.72g	23.00 A	3.218 C	80.384 D	37.170 C	9.60 B	346.667 D

Figure 3. Data after harvest

Proximate analysis of seeds

Moisture

Sunflower seeds analysis showed that the highest value of moisture was 6.1% in the 9.36g treatment while lowest value was 5.1% 2.34g treatment (Table 4). It shows that the moisture content may increase with increase in salt stress

Ash

Ash content was increased in the seeds as the salt stress was increased. The highest value (7.5%) of ash content was analyzed in

2.34g treatment. It was followed by 7.4% 4.68g and 18.72g treatments. The lowest values i.e. 7.3%, 7.3% and 7.2% of ash content was recorded in control, 2.34g and 9.36g treatments respectively (Table 4).

Proteins

The maximum protein contents (24.1%) were found in 18.72g treatment and minimum in control (23.3%). Protein contents were increased with increased concentration of NaCl dose which may has

influenced the gene expression positively (Table 4).

Crude fats

Maximum crude fats (55.7%) were recorded in 18.72g treatment. Crude fats were progressively increased as the salt doses were increased. The lowest values (51.1%) of fat contents were recorded in control (Table 4).

Crude fibers

The crude fibers showed decreased tendency as the salt stress was increased.

The highest value of the crude fibers were analysed in control (5.1%), the lowest (4.2%) in 9.36g treatment (Table 4).

Carbohydrates

The highest value of carbohydrates were recorded in control (7.5%) while the lowest value (2.7%) in 18.72g treatment. The salt stress affected the carbohydrates contents and considerably decreased the accumulation of the carbohydrates in the seeds (Table 4).

Table 4. Proximate Analysis of seeds

Treatment	Moisture (%)	Ash (%)	Proteins (%)	Crude fats (%)	Crude fibers (%)	Carbohydrates (%)
T1-Control	5.6	7.3	23.3	51.1	5.1	7.5
T2-1.17g	5.5	7.3	23.9	53.4	4.6	5.3
T3-2.34g	5.7	7.5	24.0	52.7	4.9	5.2
T4-4.68g	5.8	7.4	23.4	51.4	4.3	7.2
T5-9.36g	6.1	7.2	23.7	54.9	4.2	3.9
T6-18.72g	5.4	7.4	24.1	55.7	4.7	2.7

Figure 4. Proximate Analysis of seeds

Discussion

In the present study increased salt stress (NaCl) had inhibitory effect on the growth of sunflower plant but low dose of NaCl

had less inhibitory influence on the plant growth. Increasing salinity levels caused delay in seedling emergence as a result of reducing cell division and plant growth metabolism[4]. At the highest NaCl

concentration, seedling emergence was inhibited and some of seeds did not emerge at all [5].

Various doses of Sodium chloride decreased the height of plant, number of leaves, fresh and dry weight of plant, head diameter and number of achenes. The high doses of NaCl decreased number of achenes and plant height. Many investigations on several plant species have been carried out to determine the response of plants to salt stress [6- 8]. According to Asraf *et al.*, [6] NaCl has an inhibitory effect on sunflower seed germination and its effect on germination showed time course dependence for absorption of Na and Cl by the hypocotyls. Similarly, Cakmak *et al.*, [8] found that sunflower seedling growth was affected by NaCl treatments. The result of this study is in line with our present finding.

The number of leaves (Table 1 & Table 2) shows a significant result but Table 3 shows a non-significant result. Plant height shows a significant result with 9.36g and 18.72g treatments (Table 1 & 2) but data control show a non-significant result with 1.17g 2.34g and 4.68g treatments (Table 1 & 2). Plant height of control (Table 3) shows a non-significant result with 1.17g treatment. Control data of plant height shows a significant result with 2.34g, 4.68g, 9.36g and 18.72g treatments. Fresh and dry plant weight was also reduced significantly by different NaCl treatments. NaCl also showed negative effect on head diameter and achenes number of sunflower.

This current study can be compared to the work of Munns, 1993 [18] who stated that salt deposit in the root growing medium is the main reason for physiological drought and subsequently reduce cell division and/or enlargement in the root growing region and ultimately reduce root growth. High levels of salinity reduced leaf number, subsequently aerial parts and plant dry weight. This is likely due to the interference of salinity stress with the phytohormones biosynthesis and action [9]. Furthermore, there is evidence that high levels of salinity

inhibit growth of plants by retarding leaf primordia initiation [10]. Reduced leaf number and area lead to a low level of photosynthesis and photosynthetic production and consequently lower plant production and biomass [11].

In the present study, salt stress had a negative effect on yield and yield related attributes of sunflower. Our results support the work of some researchers. They reported that salt-induced inhibition in seed yield and yield components have been reported in different plant species e.g. sunflower [12] and wheat [13]. Generally, salt-induced inhibition in yield is associated to decline in chlorophyll contents, stomatal and non-stomatal limitations for gas exchange, high level of Cl⁻ and Na⁺ in plant tissues, oxidative damage [14-16].

Table 4 shows proximate percent composition of sunflower seed. Increasing the salt concentration, the moisture contents of seeds were also increased. Similarly, the ash was increased with increase in salt treatment. The protein and fats percentage was increased with increasing in NaCl doses. Crude fibers and carbohydrates are considerably reduced with the increase in the salt stress. Growth reduction due to salinity is well documented [17, 18] and is mainly attributed to water deficit due to lower water potential in the root zone, nutritional imbalance and specific ion toxicity arising from higher concentration of Na⁺ and Chloride ions. [4]. Different physiological trades such as selectivity for potassium, exclusion of Na⁺ and Cl⁻ ions, osmotic adjustment by accumulation of organic solutes have all been related to salt tolerance of crop plants [19]. There is also possibility that NaCl treated plants utilize energy for the osmotic adjustment processes at the expense of growth [20]. Plant subjected to abiotic stresses underwent impairment of electron transport systems of membranes that caused increase in reactive oxygen species (ROS) production [21]. ROS can rapidly attack all types of biomolecules such as nucleic acids, proteins, lipids and amino acids [22]

leading to irreparable metabolic dysfunction and cell death. Therefore, antioxidant enzymes can protect plant cells from injury. Responses of these enzymes activate the essential component of the plant antioxidative defense [23].

Authors' contribution

Conceived and designed the experiment: WM Khan, Performed the experiment: S Karim, Analyzed the data: MS Khan, Provided the Sunflower hybrid: S Ali, Wrote the paper: N Umar.

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