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Salinity Effects on Growth, Electrolyte Leakage, Chlorophyll Content and Lipid Peroxidation in Cucumber (*Cucumis sativus* L.)

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Abstract. A glasshouse experiment was carried out at Sultan Qaboos Univeriesty, Oman, to evaluate the performance of cucumber grown at varying salt stress levels viz; 0, 2, 3, 4, and 5 dSm^{-1}. The results revealed that salinity adversely affected the morphological, physiological and biochemical attributes of cucumber. The results exhibited that stem length and number of leaves per plants was substantially reduced at 5 dSm^{-1}. Moreover, the highest weakening in shoot and root dry weights were noted at 5 dSm^{-1} NaCl level in contrast to control treatment. The data set further narrated that the electrolyte leakage was enhanced remarkably with the increasing NaCl levels; however, substantially maximum electrolyte leakage was noted at 5 dSm^{-1}. Moreover, the same salinity level also decreased total chlorophyll contents than any other salinity level. Water potential of cucumber plant was decreased with increasing NaCl levels and maximum lower water potential was observed at 5 dSm^{-1} NaCl level. In conclusion, the varying levels of salt stress substantially reduced the morphological, physiological and biochemical; maximum reduction in all growth and physiological attributes were observed at salinity level 5 5 dSm^{-1} and this was positively correlated with increased salinity regimes.

Keywords: Cucumber, salt stress, climate change, growth

1. Introduction

Among different abiotic stresses, soil or water salinity is the major constraint in reducing vegetable growth and productivity in many vegetable producing areas of the world [1], [2]. Inadequate irrigation and intensive clearing of lands resulted in increased accumulation of concentrated salts in plant root zone.

Plants respond to salt stress in different phases, first the rapid osmotic stress phase that results in lower soil water potential due to higher sodium concentration in the root vicinity whereas the second is the prolonged ionic stress phase that causes the nutritional imbalance and direct toxicity of Na^+ ions that is present in the plant leaves. At plant level, both osmotic and ionic phases caused reduction in young leaf growth and senescence of older leaves, respectively [3]. The plants adapt varying mechanisms to cope with the deleterious effects of salt stress. Scientific findings highlighted inhibition of growth of many field crops particularly in vegetables for their high level of susceptibility to salt stress. Cucumber (*Cucumis sativus* L.) is a cucurbit annual and is well adapted to temperate climatic conditions [4]. In addition, it also grows well at high temperatures, humid conditions and excessive light intensity.

Agricultural statistics of Oman showed that cucumber is one of the most widely grown vegetable in Oman that occupies almost 90% of greenhouses under vegetable culture in the country [5], [6]. Constraints in production of cucumber in the country are both biotic and abiotic leading to decreased income of the farming community [7]. Salinity is one of the most deleterious abiotic factors retarding cucumber yields due to the intrusion of seawater into the coastal aquifer across the northern Oman. This resulted in raised salinity

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in agricultural lands and con depleted groundwater for agricultural production and soil borne salinization [8]. Reduction in growth and yield of cucumber in response to salinity exposure is well established but the reasoning on physiological grounds is little addressed. Such understanding to evaluate the effect of salinity on growth, electrolyte leakage, chlorophyll content and lipid peroxidation mechanism can help in ameliorating adverse impact of salinity on growth of cucumber and better productivity can be achieved.

2. Materials and Methods

A glasshouse experiment was conducted at Agriculture Experiment Station, Sultan Qaboos University, Oman, at 27/20 ±2 ℃ and 250 μmol $m^{-2}s^{-1}$ environment conditions. Three weeks older uniform seedlings were transplanted in 35cm plastic pots filled with equal amount of compost and vermiculite. Irrigation needs of the plants were monitored and equal amount of water was added at alternate days. Hoagland's nutrient solution was supplemented once a week to maintain uniform nutrition status [9]. Complete randomized design with nine replications was employed and salt stress levels were kept at 0, 2, 3, 4, 5 dSm^{-1}. After 14 days of salt exposure, measurements were made to observe and quantify effects of salinity on cucumber. After 14 days of treatments plant measurements were recorded and harvested samples were immediately transferred in a cool dry container to the laboratory for biochemical and other physiological analysis. Water potential of the middle leaf from each treatment was recorded for five replicates by using WP4C instrument. Leaf discs were excised according to instrument leaf chamber space and were immediately transferred into instrument chamber cavity and average readings were taken. All observations were made after 6 hours of watering [10]. Growth measurements for fresh and dry weight after 14 days treatment were taken as described by [1]. The chlorophyll meter or SPAD meter is a simple diagnostic tool that measures the greenness or relative chlorophyll content of leaves. Minolta SPAD-502 was used to measure the chlorophyll content throughout experiment. Lipid peroxidation was measured as the amount of MDA produced by the TBA reaction as described by [8]. Fresh leaf tissue about 100 mg was homogenized in a minimum volume of extraction medium KPi buffer and homogenate was centrifuged at 13000 g for one minute. The 200 µl sample supernatant was added to 3 ml reaction reagent and absorbance was recorded at 532 and 600 nm. Electrolyte leakage was measured using an electrical conductivity meter Jenway Model 4330 as described by [11]. Leaves were excised and washed with demonized water. After drying with filter paper, 1 g fresh weight of leaves were cut into small pieces (about 1 cm^2) and then immersed in 20 ml demonized water and incubated at 25 ℃. After 24 hours, electrical conductivity (EC1) and after 48 hours (EC2) of the bathing solution was recorded of same samples.

3. Results

Results showed that the NaCl treatments affected morphological traits (stem length and number of leaves) adversely and this effect was more pronounced at 5dS m^{-1} (Fig. 1).

The decrease in shoot length was 38%, 45% and 53% at 2, 3 and 5 dS m^{-1} of salt stress respectively compared to non-treated plants. The data regarding about the number of leaves also showed a gradual decrease with increasing the salt levels and the maximum decrease in number of leaves was recorded at 5dS m^{-1} NaCl level as compared to the control plants (Fig. 1).

The data showed that the electrolyte leakage was significantly increased with the increasing NaCl levels (Fig. 2). The maximum electrolyte leakage was recorded at 5 dS m^{-1} NaCl level compared to the control plants. The MDA (malondialdehyde) reactive product (lipid per oxidation) showed significant effect of treatments. The salt stress induced MDA increase was 50%, 66% and 72% at 2, 3 and 5 dS m^{-1} of NaCl levels respectively (Fig. 2).

Total leaf chlorophyll contents significantly decreased with an increasing NaCl levels. The decrease in total chlorophyll contents was 12%, 21% and 30% at 2, 3 and 5 dS m^{-1} of salt stress respectively compared to non-treated plants (Fig. 2). The water potential of cucumber plant was decreased by increasing NaCl levels. The maximum lowered water potential was observed at 5 dS m^{-1} NaCl level (Fig. 2).

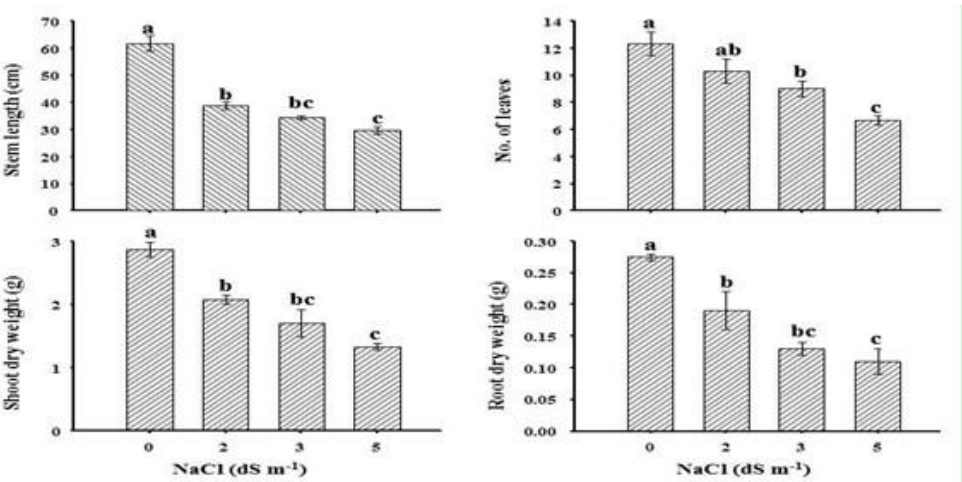

Fig. 1: The influence of different NaCl levels (0, 2, 3, 4, and 5 dS m^{-1}) on shoot length, number of leaves, shoot and root dry weights of cucumber plant. Data are the mean±standard error. Bars showing same letter do not differ significantly at 5% probality.

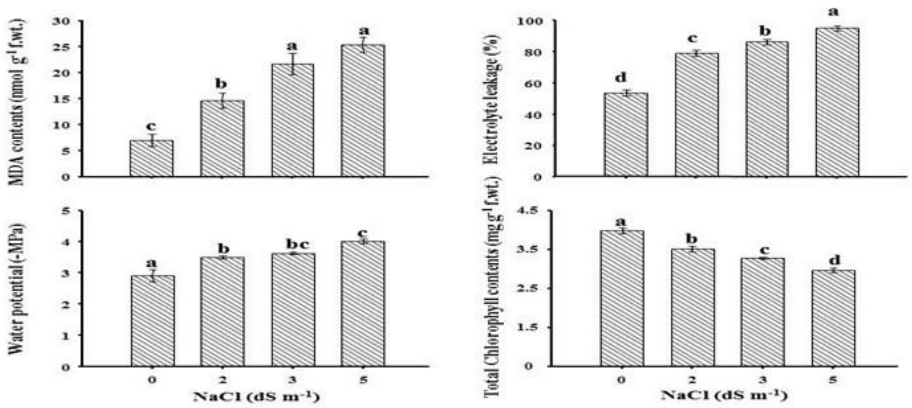

Fig. 2: The influence of different NaCl levels (0, 2, 3, 4, and 5 dS m^{-1}) on MD A contents, electrolyte leakage, water potential and total chlorophyll contents of cucumber plant. Data are the mean ±standard error. Bars showing same letter do not differ significantly at 5% probality.

4. Discussion

Crop plants vary in their ability to tolerate the salt stress [12]. Cucumber (*Cucumis sativus* L.) is highly susceptible to salt stress [13]. Currently little information is available about the effects of salt stress on cucumber plant growth and development. The present study was conducted to study the effect of salt stress on cucumber plant. The results showed that salinity reduced the number of leaves in cucumber plant. Reduction in number of leaves is a common attribute of plants growing under salt stress [14].This decrease under salt stress may be due to the reduction in turgor potential that is pre-requisite for cell elongation in plants [15] and due to reduced turgor pressure [16].

The results of present study demonstrated a decline in overall growth of plants due to reduced dry root, shoot weight, number of leaves and stem elongation. These results also showed that the effect of salt treatments was more pronounced on root dry weight than shoot dry weight. This significant effect of salt stress on cucumber root vs. shoot may be due to more water loss from the root than shoot during salt stress. The decreased effect of salts on shoot dry biomass may be due to stomata closure that controls the water loss from the shoot. In fact, NaCI treatment cause osmotic changes in the leaf tissue or effect on leaf CO_2 concentration that leads towards stomata closure [13]. In general, many previous studies on most crops showed same results [17], [14]. However, [18] reported that the reduction in leaf area and stunned root and shoot growth may depend on both cell division and cell elongation which can be directly affected by the salinity.

Electrolyte leakage was enhanced with increasing salinity levels as compared to the control cucumber plants. Similarly, [13] observed the same increasing trend of electrolyte leakage in salt sensitive cucumber cultivar as compared to the salt tolerant cultivar. This phenomenon was already observed by several authors in cucumber, [19] rice [11] and sugar beet [20]. In this study, NaCl-stress led to a significant reduction in the leaf chlorophyll contents. This effect agrees with earlier work [21]. The decreased levels in chlorophyll content under saline stress are commonly phenomenon and it may be due to different reasons like an inhibition of chlorophyll biosynthesis from an activation of the chlorophyllase [22], [23] and membrane deterioration that resulted salinity mediated chlorophyll degradation [24].

Water potential is an important physiological parameter for determining the water status of the plants [22]. In our experiments the water potential of cucumber plants decreased linearly with increasing salinity levels. The more negative value of water potential with increasing NaCl levels suggested a decreased water uptake due to the increased salt concentration in the external environment. This result was found to be similar that was done before for leaf water potential where lower water potential was observed with increasing salinization [25], [26]. However, some researchers reported contrary findings.

Lipid peroxidation levels in leaves assessed as the content of MDA that is a more reliable indicator of salt stress tolerance. In our present study there was an increase in MDA content under salt treatment. The content of MDA, a decomposition product of polyunsaturated fatty acids produced during peroxidation of membrane lipids that increase significantly with increasing salt stress. MDA content and membrane permeability is used as biomarkers for lipid peroxidation [27]. It has been suggested that a decrease in membrane stability reflects the extent of lipid per oxidation caused by ROS [28]. Thus, lipid peroxidation of the cell membrane components is caused by reactive oxygen species which generated oxidative stress to plant. We suggest further studies to examine such deleterious effects on wide range of cucumber cultivars grown in Oman.

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