Journal of Applied Pharmaceutical Science Vol. 2 (11), pp. 131-133, November, 2012
Available online at http://www.japsonline.com
DOI: 10.7324/JAPS.2012.21124
ISSN 2231-3354

Effect of salt stress on the growth and photosynthetic pigments of pigeon pea (*Cajanus cajan*)

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ARTICLE INFO

Article history:
Received on: 16/08/2012
Revised on: 20/09/2012
Accepted on: 05/10/2012
Available online: 30/11/2012

Key words:
Cajanus cajan, salt stress, growth, photosynthetic pigments, proline, water potential

ABSTRACT

The effect of salt stress on some physiological parameters in pigeon pea (*Cajanus cajan*) was studied under controlled conditions. The plants were treated with the solutions of 50 mM NaCl, 50 mM Na_2So_4, 100 mM NaCl and 100 mM Na_2So_4, starting at the appearance of first trifoliate leaf unfolded. Ground water was used for irrigation as the control. It was established that the applied doses of both salt types caused stress on the young pigeon pea plants, which found expression in the suppression of growth, photosynthetic pigment. The amount of proline in the tissues of the salt-treated plants was increased.

INTRODUCTION

At present about 20% of the world's cultivated land and approximately half of all irrigated land is affected by salinity (Zhu,2001). Therefore, salinity is one of the most significant abiotic factors limiting crop productivity (Munns, 1993; Gama et al., 2007). This is attributed to the fact that Na^+ competes with K^+ for binding sites essential for cellular function (Tester and Davenport, 2007) and the latter implication of these two macronutrients in salinity is thought to be the one of the factors responsible for the reduction of the biomass and yield components of plants. High concentration of salt in the root zone (rhizosphere) reduces soil water potential and the availability of water (Lloyd *et al*, 1989). As a result of this, reduction of the water content leading to dehydration at cellular level and osmotic stress is observed. The increased amount of Na^+ and Cl^- in the environment affects the uptake of many indispensable nutrients through competitive interactions and by affecting the ion selectivity of membranes. For example, external Na^+ negatively impacts intracellular K^+ influx, attenuating the acquisition of this essential nutrient by cells. The most important process that is affected in plants, growing under saline conditions, is photosynthesis. Reduced photosynthesis under salinity is not only attributed to stomata closure leading to a reduction of intercellular CO_2 concentration, but also to non-stomata factors. There is strong evidence that salt affects photosynthetic enzymes, chlorophylls and carotenoids (Stepien and Klobus, 2006).

The most studied compound under salinity stress is proline. The amount of proline usually increases under salinity (Munns, 2002). In most cases, salinity problems are linked to an excess of NaCl in the irrigation water, but sometimes other salts, like Na_2SO_4, are present. There are few studies on the effect of Na_2SO_4 on plant growth. The objective of the present study was to compare the physiological reactions of young pigeon pea plants to two salt types - NaCl and Na_2SO_4. It was assumed that the two salt types could have different effect with respect to plant growth and photosynthesis. Previous researches have suggested that peas are salt- sensitive plant.

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MATERIALS AND METHODS

Pigeon pea *(Cajanus cajan*) seed were surface sterilized with 0.2% $HgCl_2$ solution for 5 min with frequent shaking and then thoroughly washed with deionized water. The seeds were sown in plastic pots filled with 3 kg of 1:1:1 soil mixture containing red soil, sand and farmyard manure (FYM). Five seeds per pot were sown, and all the pots were watered with tap water up to 7 days after sowing. On 8 day onwards pots were irrigated with groundwater as control or with 50 mM NaCl, 50 mM Na_2So_4, 100 mM NaCl and 100 mM Na_2So_4 solutions. The plants were collected on seventh day after treatment and used for growth parameters, proline content and photosynthetic pigments. The plants were harvested on 7 days after treatment and the fresh weight of plants, dry weight of roots, shoots, and leaves and leaf area (LICOR Photoelectric Area Meter Model LI-3100, Lincoln, USA) were measured. The photosynthetic pigments in the leaves were determined spectrophotometrically (Arnon, 1949). Proline was estimated according to Bates et al. (1973).

STATISTICAL ANALYSIS

Statistical analysis was performed using a one way ANOVA (for $P < 0.05$). Based on the ANOVA results, a Turkey test for mean comparison was performed, for a 95% confidence level, to test for significant differences among treatments. In the tables, different letters (a, b, c) express significant differences, with a representing the highest value.

RESULTS AND DISCUSSION

The salt-treatment of pigeon pea plants caused symptoms of phytotoxicity – the leaves became brown, yellow and dry. The root tips turned brown and a considerable inhibition of the root system growth was observed. From the data, it is evident that the toxicity in the chloride treatment is expressed more clearly in older leaves, while in the case of the sulphate treatment, a similar effect was observed with respect to younger leaves and the plants' tops .The results regarding the changes of the basic biometrical parameters of young pigeon pea plants indicate that the applied salt treatment causes a considerable inhibition of their initial development. The data on the accumulation and the distribution of the fresh and dry weight in the organs of the young plants are shown in Table 1. They change in accordance with the salt type and its concentration. In case of salt treatment with 50 mM NaCl and Na_2SO_4, the inhibition of the fresh and dry weight was within the limits of 17 – 23%. In both types of salt treatments the higher dose caused a stronger inhibition effect. This tendency was observed with respect to all parameters and was averagely at 50% below the control level (Fresh weight and Dry weight of plants). The addition of Na_2SO_4 to the nutrient environment caused a stronger inhibition effect. This tendency was more clearly expressed with respect to the plants dry weight. The dry weight changes in the different plant organs had similar values. Once more the dose of 100 mM Na_2SO_4 caused stronger inhibition than 100 mM NaCl. The applied salt treatment caused considerable changes in the ratios between the dry weight in the roots and in the above-surface organs (Dry weight of root/Dry weight of shoot), and these values were at about 30-40% above the control. A strong inhibition effect was also observed with respect to the leaf area. The reduction of this index at the dose of 100 mM was 64-67% below the control, for the chloride and sulphate treatments, respectively. Salinity type (NaCl and Na_2SO_4) would be expected to have an impact on plant growth, where a direct ion toxicity or nutritional imbalance occur (Jenifer and Franklin Janusz, 2002).

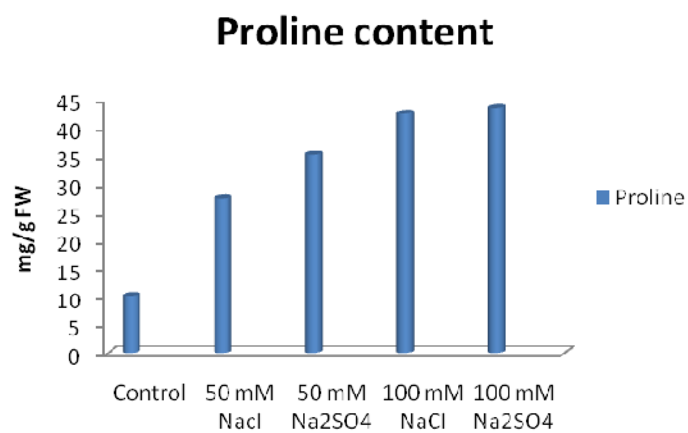

Fig. 1: Effect of salinity on proline content of *Cajanus cajan.*

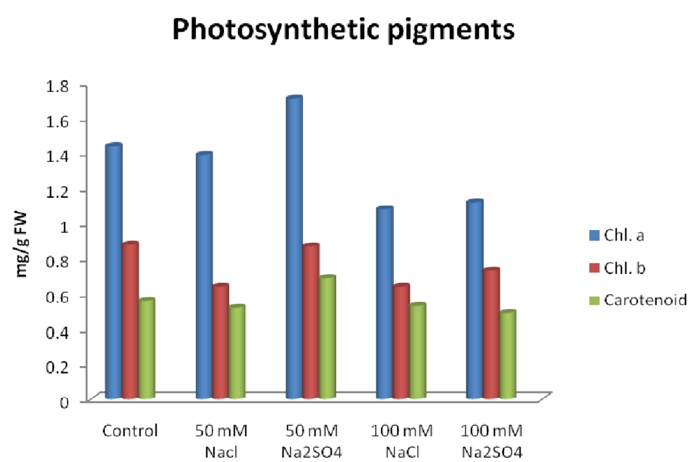

Fig. 2: Effect of salinity on photosynthetic pigments of *Cajanus cajan*

Proline accumulation is an important physiological index for plant response to salt stress (Shi and Yin, 1993), as well as to other types of stress. The relationship between proline accumulation in the shoots of bean plants and the two salinity types is shown in Fig: 1. It is evident that proline content increased parallel with the rise of the salinity levels. In the case of higher salinity level (100 mM) the proline concentration was increased 3-3.5 times compared to the control. This result demonstrates that sulfate stress can cause higher accumulation of proline than the chloride type. This fact suggests that the induction of proline synthesis is related not only to changes in the water potential and to the salinity type – chloride and sulfate, but also resulted from metabolism interruption by high-stress intensity or from an

Table. 1: . Effect of salinity on growth parameters of *Cajanus Cajan.*

Growth parameters	Control	50 mM NaCl	50 mM Na_2so_4	100 mM NaCl	100 mM Na_2so_4
Fresh weight of plants (FW)	10.24 a	8.92 b	8.55 b	5.42 c	4.90 c
Dry weight of plants (DW)	1.29 a	1.04 b	1.03 b	0.59 c	0.55 c
DW/FW	0.13 a	0.12 a	0.12 a	0.11 b	0.10 b
Dry weight of root (DWr)	0.24 a	0.23 a	0.25 a	0.09 b	0.14 b
Dry weight of shoot (DWsh)	0.30 a	0.20 b	0.21 b	0.13 c	0.15 c
Dry weight of leaves	0.75 a	0.61 b	0.58 b	0.31c	0.28 c
DWr/DWsh	0.30 a	0.28 b	0.32 a	0.31a	0.33 a
Leaf Area (cm^2)	258.95 a	184.45 b	165.15 b	93.65 c	79.73 c

Mean values are the average of three replicates. Within the same column, values flanked by the same letters are not significantly different for *P=0.05* following one-way ANOVA test..

adaptive response with special physiological function. The increased levels of proline, under salt stress, have been reported in two wheat cultivars (Khatkar and Kuhad, 2000). It was suggested that proline accumulation may be caused by increased proteolysis or by decreased protein synthesis. The higher concentration of proline under salt stress is favorable to plants as proline participate to osmotic potential of leaf and thus to osmotic adjustment (Cachorro et al., 1995; Hasegawa et al., 2000). Besides the role of osmolyte, proline can also confer enzyme protection and increase membrane stability under various condition. The photosynthetic pigments are some of the most important internal factors, which in certain cases can limit the photosynthesis rate The data presented in Fig: 2 shows that the applied treatment tended to diminish the content of all pigments and to a certain extent changed their ratios. The decrease of Chl.a content at 100 mM NaCl and Na_2SO_4 was considerably different from the controls and was 75% and 77%, respectively. The decreased content of both chlorophyll and carotenoids could probably be a result from the mineral deficiency. Total chlorophyll content in both salt-stressed plants declined with the salinity increase. It was established that Carotenoid decreased to a lesser extent than Chl. Ion accumulation in leaves adversely affected Chlorophyll content (Meloni et al., 2003). The observed decrease of Chlorophyll content in the plants grown under saline conditions may be attributed to both of the increased degradation and the inhibited synthesis of that pigment (Garsia-Sanchez et al, 2002). Interestingly, Chl.a was less sensitive or better protected against salt stress compared to Chl b.

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How to cite this article:

P. Amuthavalli and S. Sivasankaramoorthy. Effect of Salt Stress on the Growth and Photosynthetic Pigments of Pigeon Pea (*Cajanus Cajan).* J App Pharm Sci. 2012; 2 (10): 131-133.