Calcium and Oxalic Acid Contents of Sugar Beet Plant in Salinity Stress

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***Abstract*—**Sugar beet plants were grown in peat-perlite substrate by using a non-recirculating nutrient film growing system. The effect of NaCl addition at 0 (control), 25, 50 and 75 mM concentrations to nutrient solution on calcium and oxalic acid contents in the leaf and root tissues of sugar beet plant were examined. NaCl applications decreased leaf and root dry matters and calcium contents in the leaf and root tissues of sugar beet. Oxalic acid content was increased in the leaf tissue while decreasing in the root tissue by NaCl applications. Physiologically active oxalic acid amount was only determined in the root tissue at the highest NaCl level. Although Ca deficiency induced by low plant Ca uptake due to high salinity, physiologically active oxalic acid accumulation in the root tissue of sugar beet may be a limiting factor on Ca availability and plant productivity in salinity stress.

***Keywords*—** Salinity, calcium, oxalic acid, sugar beet.

I. Introduction

Oxalic acid is a common constituent of plants and is found in almost all plant families, occurring as free acid, soluble salts of potassium, sodium and magnesium and as insoluble salts of calcium and iron. High oxalic acid concentrations in leaves of plant species, especially those used as green leafy vegetables in the daily diets, have been of concern because of the harmful health effects associated with the intake of high oxalic acid [1].

Oxalate-rich foods are usually minor components in human diets but are sometimes important in seasonal diets in certain areas of the world. Plants such as spinach and beetroot are well known for containing higher concentrations of oxalates than other plants [2].

Oxalic acid is not only effects diet mineral availability but also effects plant Ca metabolism. Calcium present in plant foods exists primarily as a complex in which it is bound to substances such as oxalate, phytate, fiber, fatty acid, protein, and/or other anions. Most studies suggest that oxalate is an antinutrient that renders calcium and sometimes other minerals unavailable for nutritional absorption by human. Most of the findings showed that the general conclusion that the more calcium in the form of the oxalate salt, the less calcium available for nutritional absorption [3].

Ca is a very immobile essential macro element once deposited in a tissue. Ca immobility may result from ion complexes as oxalate or other insoluble forms and from binding to the cell wall making it unavailable for transport [4].

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Ca deficiency causes of black tip at the apex of the petiole of sugar beet. The prevalence and severity of black-tip formation has been consistently associated with factors causing formation of insoluble Ca compounds such as phosphate or oxalate at the growing tip [5]. Excessive concentrations of soluble salts in the substrate have also been associated with black-tip formation and can cause a decreased Ca uptake even when the measurable Ca ratio (Ca/soil solution salts) appears to be adequate [6]. Ca is concentrated in the form of calcium oxalate and may be precipitated in plant cells and thus oxalic acid limits Ca availability for continued shoot growth [7].

Another factor affecting plant Ca uptake is soil salinity. Salinity is a major factor in reducing plant growth and productivity. In saline conditions plant Ca uptake significantly be reduced and the rate of Ca uptake decreased linearly with increasing salinity [8]. Understanding the physiology of high oxalic acid occurring plants under salinity conditions is important for an effective approach to the salinity and calcium nutrition problem.

II. Materials and Methods

The experiment was carried out in greenhouse under controlled and standardized condition. Sugar beet seeds (Beta vulgaris L. Cv. Evita) were sown in perlite and after germination seedlings were irrigated and allowed to grow approximately 5 cm of shoot length. These seedlings were transferred to 10 L pots containing washed peat + perlite mixture (1:1) substrate as one plant per pot and received basal nutrient solution plus the treatments of NaCl. Each pot had a hole at the bottom to facilitate drainage when renewing the nutrient solution.

The composition of basal nutrient solution was: 1.25 mM KH_2PO_4, 2.00 mM $MgSO_4$, 4.20 mM $Ca(NO_3)_2$, 3.60 mM KNO_3, 1.50 mM NH_4NO_3, 1.87 mM K_2SO_4. Iron (Fe-EDDHA), Boron ($Na_2B_4O_7.H_2O$), Manganese ($MnSO_4.H_2O$), Zinc ($ZnSO_4.7H_2O$), Copper ($CuSO_4.5H_2O$), Molybdenum ($Na_2MoO_4.2H_2O$) were also included at the rates of 40, 30, 10, 4, 0.75, 0.5 μmol l^{-1}, respectively. NaCl was applied to basal nutrient solution at 0 (control), 25, 50 and 75 mM . Each NaCl treatment was replicated four times in a randomised plot design.

Pots were irrigated every day with nutrient solutions contained above mentioned NaCl levels. Pots were leached with distilled water every three irrigation to prevent salt accumulation and then nutrient solution was added as previously mentioned. All plants were harvested and weighed during 2 months of growth. All sugar beet plants were

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removed from the pot and divided two parts as the upper half (leaves) and lower half tissues (roots) and then samples were washed with distilled water.

Plant material was dried at 65 °C, weighed, blended and wet ashed with HNO_3+$HClO_4$. In wet ashed samples total Ca was determined by atomic absoption spectrofotometry. In dried plant samples total oxalic acid was determined by $KMnO_4$ titration medhod [9]. Physiologically active oxalic acid was calculated as the excess of equivalent amount of total oxalic acid than that of calcium [10].

Statistical analysis were performed by using SPSS-16 for windows program.

III. Results and Discussion

NaCl applied to nutrient solutions decreased plant dry matter and fruit yield. At the application of 75 mM NaCl, plant dry matter was sharply dimished (Figure 1). The findings on plant yield decrease by NaCl applications are in concordance with previous results [11].

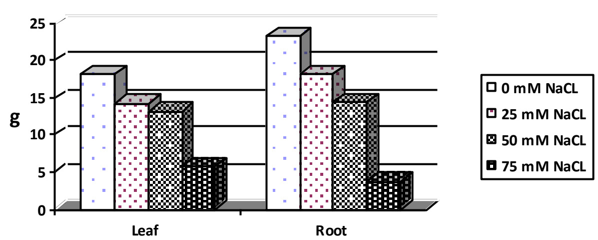

Fig 1. Leaf and tuber dry matter amounts (g/pot) of sugar beet grown in different NaCL levels.

Oxalic acid contents were decreased both in the leaf and root tissues by the applications of NaCl. In control treatment, oxalic acid content was the highest rate in the sugar beet tissues (Figure 3).

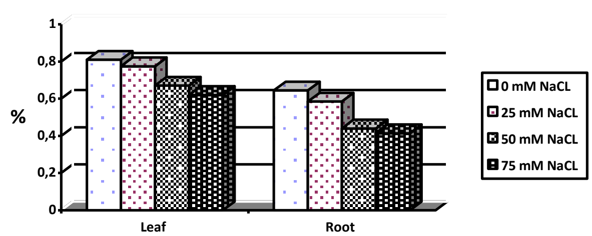

Fig 2. Leaf and tuber oxalic acid contents (%) of sugar beet grown in different NaCL levels.

Ca contents in the leaf and roots of sugar beet were decreased by the applications of NaCl (Figure 3). Typical Ca deficiency symptoms in sugar beet leaves were recorded by the applications of NaCl to nutrient solution. No deficiency symptoms were observed in control application. The data that decreasing Ca content was related with the salt applications confirm the previous findings [12]. Leaf tissue contained higher content of Ca than the root tissue.

It is thought that the incidence of Ca deficiency is not only increased depending on the low Ca content of the sugar beet in salinity stress, but also oxalic acid complexes with Ca and reduces its metabolic availability and translocation to upper tissues. It is reported that higher concentrations of oxalic acid in the upper portions of susceptible Burley tobacco varieties be observed and it fixed Ca and reduced its translocation to and metabolic accessibility in apical tissues [13].

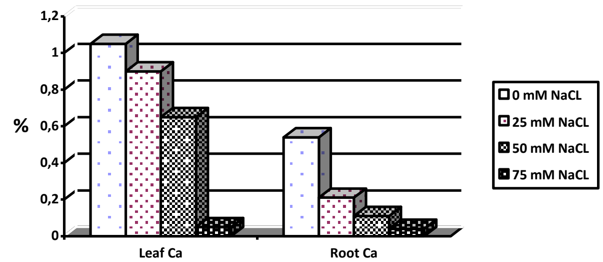

Fig 3. Leaf and tuber calcium contents (%) of sugar beet grown in different NaCL levels.

Physiologically active oxalic acid (PAOA) was a stoichiometric value and calculated as the excess of equivalent amount of total oxalic acid than that of calcium [10] and represents potential health hazards oxalate rich foods. PAOA was recorded in the highest NaCl application (75 mM) in the leaf tissue, and physiologically active oxalic acid amount was increased by NaCl applications (Figure 4). Physiologically active oxalic acid in the leaves tissues at 75 mm NaCl level and in the root at NaCl tretaments were occured as a result of lower Ca content at this level. Although oxalic acid content was also decreased by NaCl treatments, this subsidence rate was lower than for Ca.

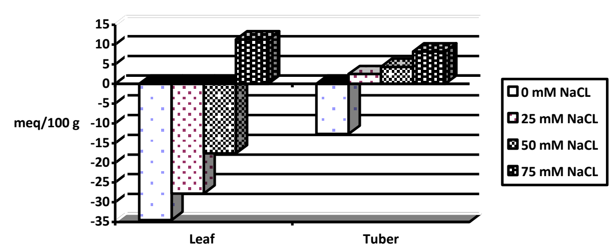

Fig 4. Leaf and root PAOA amounts of sugar beet grown in different NaCL levels.

Physiologically active oxalic acid is equal to soluble oxalate which is a stoicheiometric value and means a free form of oxalic acid which is not fixed hardly in insoluble form by divalent cations. In this meaning physiologically active oxalic acid is thought to be an important factor limiting Ca availability and mobility in plant tissues. High concentrations of oxalic acid in stems have resulted in precipitation of Ca before it reached such organs as leaves and fruits [14].

IV. Conclusion

In saline conditions plant Ca uptake is mostly depressed by Na or other metallic cations. On the other hand, due to highly significant relations between insoluble Ca and oxalic acid in the oxalate rich plants, Ca is mostly precipitated in the plant

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tissues and limiting its availability for continued shoot growth. In addition to Ca starvation induced by low Ca uptake due to high salinity, higher oxalic acid synthesis in the sugar beet may also be thought as a limiting factor on Ca availability and plant productivity.

The strategies including the improvement of lower oxalate accumulating plant varieties, and well-organized fertilization programs for low oxalate plant cultivation should be planned for effective plant nutrition in saline conditions and low oxalate food consumption for the people.

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