External Antioxidants; an Effective and Affordable Way to Overcome Salinity Stress in Plant

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ABSTRACT

Salinity stress is one of the threats for plants in deserts and some parts of the world. Plants exposed to NaCl suffer from oxidative stress, and their growth disrupts through DNA damage and enzyme inactivation. It seems that exogenous antioxidants can increase plant tolerance to salinity. Therefore, the effect of fruits peel extract such as pomegranate, sweet lemon and pistachio as extracted antioxidants on improving germination and growth of susceptible seeds to salinity, such as beans, lettuce and fennel were examined by using polar and semi-polar solvents, and ultrasonic bath and two positive (absence of sodium chloride and extracts) and negative (presence of sodium chloride and absence of the extracts) control groups.

1 Introduction

One of the most important environmental issues in Iran is desertification, which reduces the potential for biological production of saline soils [1]. The high tensions in the environment and soil of deserts and arid areas make vegetation vulnerable. One of these tensions is salinity, which negatively affects plant growth and weakens it. All plants can't withstand the salinity of the environment. A few species can survive in these conditions. Therefore, the vegetation of these areas is reduced [2, 3, 4].By reducing the absorption of water by root or by disturbing the ionic balance, salinity has an inhibitory effect on growth, production and metabolic processes of plants, such as photosynthesis, respiration and biochemical activity of the plant. Most environmental stresses, such as dryness and salinity, lead to the production of reactive oxygen species (a variety of free radicals), oxidative stress in plant cells, and ultimately cell death [4]. There are several ways to cope with this problem and help the plants grow in saline soils, such as; the use of osmoprotectants, the transfer of the gene of plants resistant to saline sensitive plants, and the use of Antioxidants [5,6,7]. During the optimum plant growth, the balance between the formation of free radicals and their use are strongly controlled by the antioxidant defense system of the plant [7,8]. The results obtained in this research can be used to regenerate degraded ecosystems, prevent soil erosion, and also convert saline lands to arable land in Iran, which leads to an increase in the quality and quantity of agricultural products.

2 Materials and Methods

Using studies from other researchers on the subject, seeds of three herbs, including beans, lettuce and fennel, were selected as salinity sensitive plants and their sensitivity was re-examined. Pomegranate, citrus limetta, and pistachio peels were selected as three high oxidant sources because of their antioxidant activity. Selected sources were exposed to air, and extract from 10 g each using four solvents with different polarity: aqueous the ability of various solvents to extract bioactive compounds from the peel fruit; through a chromatography of a paper layer using silica-coated aluminum and methanol and dichloromethane solvents; and studying the formation of bands of extracts by UV light, was done. The extracts were dried using nitrogen gas so that the chemical solvents with the extracts did not enter the growth environment of the plants and did not damage the seeds. Each dried extract was mixed with distilled water and, to prevent the entry of any microorganisms and impurities, the solution was passed through a sterilization filter (0.22 μm). Seeds of selected plants were sterilized with diluted bleach under biohood. After three times washing, three seeds from each plant were entered into the growth medium containing 50 mM NaCl. The protective effect was studied in two groups of control- a positive control sample (absence of sodium chloride and extracts) and a negative control sample (presence of sodium chloride and the absence of extracts)- and treatment group- presence of a variety of extracts- to compare the growth rate of the seeds in the treatment and control groups (especially with negative control samples). In order to investigate the antioxidant ability of the extracts in antioxidant activity with DPPH, a clear solution of 200 μg of extracts extracted in methanol was prepared. 10 μl of extract with 200 μl of DPPH solution was heated at ambient temperature and in the dark for 30 minutes. Finally, absorption of each well was read at 570 nm. In order to focus on the research process, based on the results obtained in the initial test, repeat testing on bean seeds (its cost-effectiveness in broad applied scale in the case of lettuce with high water requirements and faster germination in the presence of extracts, unlike fennel) as a salinity sensitive plant, and citrus limetta (the greater effect of its polarized extracts on the growth of bean seeds, compared with other polar extracts) that it was used as an antioxidant source, was done.

3 Results and Discussion

According to the results, the type and amount of bio-compounds extracted by solvents used in different polarities are completely different. Aqueous extract of citrus limetta, improved seedling growth of beans (3 seeds of 3 seeds, length of buds of 5 cm) and its ethylated extracts increased the growth of lettuce seeds (3 seeds of 3 seeds, 3 cm long buds) significantly compared to the negative control group. The aqueous extract of pistachios, significantly increased the growth of bean seeds (1 seed of

of 3 seeds, length of 2 cm buds) and its ethyl acetate extracts improved growth of lettuce seeds (3 seeds of 3 seeds, length of bud 3 cm) and fennel (2 seeds of 3 seeds, 1 cm length). Superior extracts showed significant antioxidant activity in the DPPH test. According to this study, the pistachio ethyl acetate extracts (74%), the citrus limetta (69%) and the aqueous extract of citrus limetta (55%) have antioxidant quality. (Fig.1, 2 and 3)

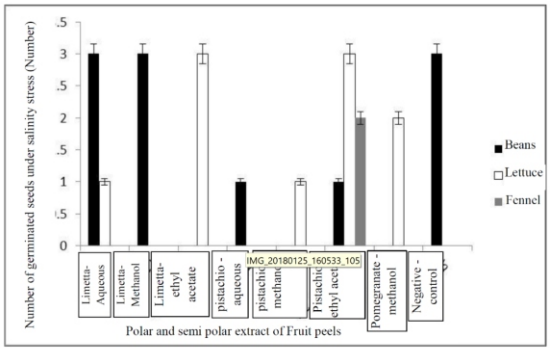

Fig.1: Number of germinated seeds of each plant under salinity stress (repeat = 3)

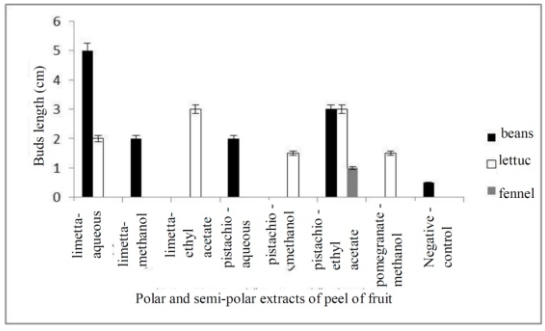

Fig.2: Seed length of each plant under salinity stress (repeat = 3)

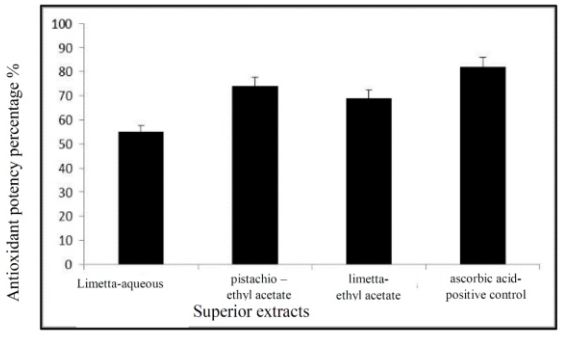

Fig.3: Antioxidant potency of superior extracts using DPPH, ascorbic acid and methanol was used as positive and negative control. (Repeat = 3)

According to the results of the repeated test, the effect of the bio-compounds extracted by the polar solvents used, namely, water and methanol, is different. The aqueous extracts of citrus limetta peel and methanolic extracts of citrus limetta peel significantly improved growth of bean seeds (9 seeds of 10 seeds with average length of buds of 56.9 mm and 8 seeds of 10 seeds with an average length of 19.5 mm buds) compared to the negative control group. (Fig.4, 5 and 6).

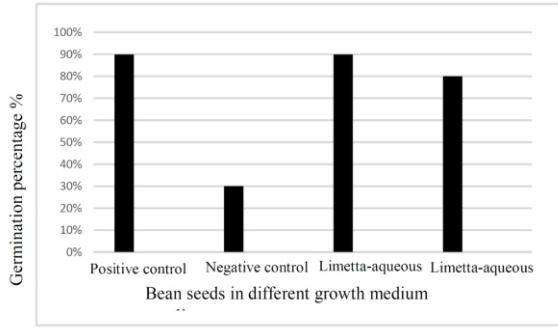

Fig.4: Germination percentage of bean seeds in different growth medium

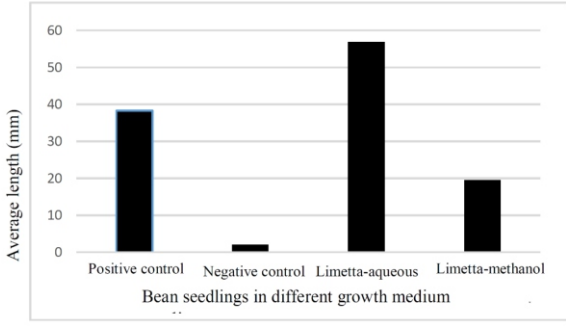

Fig.5: Average length of seedlings per mm

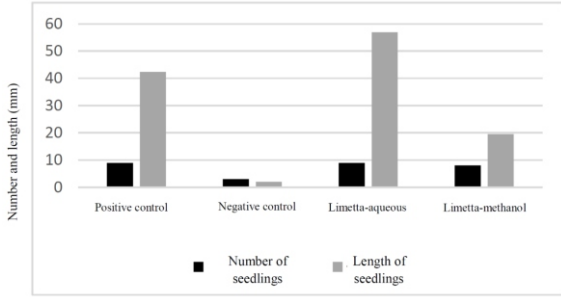

Fig.6: Comparison of bean seed function in different growth media

According to the observations, antioxidants in selected extracts increased the tolerance of saline-sensitive plants. The compounds of aqueous and ethyl acetate extracts have a significant effect on the growth of bean and lettuce seeds. Therefore, the compounds that affect the growth of bean seeds are polar, and the compounds that affect the growth of lettuce seeds are semi-polar. The factor that distinguishes this project from other research is extracting antioxidants from natural wastes which makes it very economical and applied solution in environmental issues. One of the important results of this study is the effect of antioxidants on plant tolerance to salinity, which is consistent with the results of other investigations.

4 Conclusions

According to the results, vegetable wastes, especially peel of pigmented fruit, are rich and affordable sources of antioxidants that can be extracted using various solvents and increase tolerate salinity sensitive plants to oxidative stress caused by salinity and growth retardation.

Some suggestions are made to continue this research; 1) Antioxidants are extracted with different solvents. 2) Experiments are conducted on different plant species with

different resistance. 3) Test are carried out on various bean species (Navy bean and Cowpea, etc.), which have different resistance to salinity. 4) In a separate project, instead of peeling pigmented fruits, antioxidant compounds from their nuclei are extracted. One of the limitations of this research was the lack of time for a sampling of natural saline soils and the plants were artificially subjected to salinity stress. And another limitation was the lack of time to carry out the experiment in the soil.

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