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Simple Germination And Root Elongation Bioassay For Early Detection Of Salt-Tolerant Genotype

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Introduction

Sustainable agriculture development is limited worldwide by the lack of fresh-water resources. Thus, arid and semi-arid regions are relying on saline water irrigation to assure appropriate crop yields. Though, constant saline water irrigation entails modifications of soil properties, lowers plant productivity, and limits crop growth. Saline stress impacts particularly on plant early stages, such as seed germination and seedling development. Hence, finding salt tolerant crops can assure satisfactory yields and diminish pressure on fresh-water reserves.

Here we propose a novel technique to investigate the physiological traits altered by saline stress, easily enabling the early detection of salt-tolerant genotypes.

Our experiment was performed with castor bean (*Ricinus communis* L.), a tropical plant belonging to the spurge family that can establish on marginal lands, employed in several industrial products, and used in biodiesel production.

Materials and Methods

Plastic containers used were originally designed as boxes for sugared almonds (30 x 40 x 240 mm). The bottom of each box was closed with waterproof tape, while the longer side was closed with normal tape, leaving 2 centimeters between the two tapes (Fig.1). Ten seeds for each tested genotype were sown individually in plastic containers (30 x 40 x 240 mm). The containers filled with silica sand. Twenty-five containers were placed in a plastic tray (150 x 200 x 60 mm) that provided standing position and acted as water reservoir. The trays were placed in a growth chamber (25°C, 12/12 d/n, 40% of humidity) and moved every two days to avoid position bias. The plastic containers were watered with saline water (0, 4 or 8 dS m⁻¹) until field capacity. At emergence, germinations were counted every day. After 15d plants were harvested, and length of the main root, shoot, and fresh weight was measured. Each plant was then photographed to allow digital imaging analysis (Fig.2). Images were analysed with the software ImageJ. Then, plants were dried at 50°C until constant weight. Three replicates for each saline level were performed, thus 30 seeds for each genotype and each saline level were used.

Preliminary Results

The experiment revealed its functionality by screening the most salt tolerance castor genotypes (Fig. 3). While some varieties do not germinate at all, other were fully capable to develop. Root elongation, shoot length and germination percentage allows a further screening for the most adapted to saline water irrigation. Moreover, different index can be easily evaluated, e.g., Relative Root Elongation (i.e., the ratio of root length of the saline treatment vs. the control), or Mean daily germination. The use of sand allows easy root cleaning. The containers can be reused multiple time, decreasing experimentation costs.



Fig. 1. Plastic boxes used for the bioassay

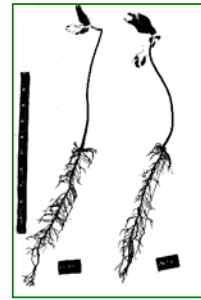


Fig. 2. Image analysis of seedlings

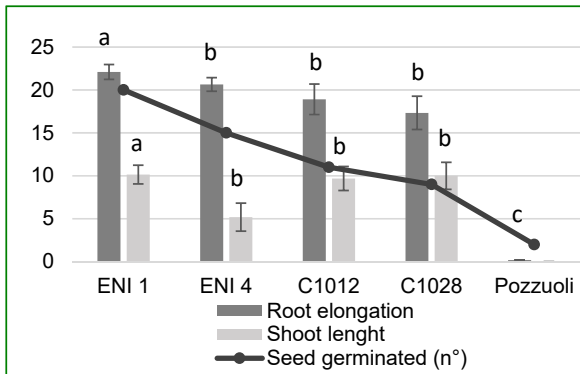


Fig. 3 Roots and shoots length (cm) and number of germinated seeds for each variety, treated with saline water (4dS m⁻¹). Mean values with the same letter are not different ($p < 0.05$).

Conclusions

The identification of salt tolerant crops is an imperative mission in the climate change era. Here we present a simple, low-cost methodology to identify salt tolerant genotypes. Plants are differently sensitive to salt stress. Thus, for further research is important to study other plant species, less and more tolerant of castor bean. This kind of experiment could be used to study roots architecture, roots diameters and other physiological characteristics. Moreover, it could be also utilized for ecotoxicological test with potentially toxic elements.