A Technique for Testing and Selecting for Salt Tolerance in Sugarbeet

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Increased salt tolerance in sugarbeets (*Beta vulgaris* L.) would markedly increase sugar yields in many irrigated areas. Excessive salinity drastically reduces seedling stands and reduces plant growth in these areas. Sugarbeets are relatively sensitive to salinity at seed germination as compared with later stages of growth (2, 4, 6, 7, 15)[2]. At germination, the seedling stage is also subjected to the highest probable salt levels because of the close proximity of the planted seed to the soil surface where salt concentrations occur.

High temperatures drastically decrease seed germination (1, 3, 5, 14) and interact with salinity to reduce seed germination (1, 3). Francois and Goodwin (3) reported serious adverse effects of salinity on sugarbeet seed germination in the 25-35°C range. These temperatures occur during fall seed germination in irrigated desert environments (10). Other factors such as initial seed water content, substrata oxygen (9), and decortication or other forms of processing seed (11, 12) also affect seed germination. Maternal tissue of the seedball rather than embryo characteristics may establish the speed of germination (12).

A technique for evaluating the salt tolerance of a large number of sugarbeet lines was sought. Requirements for the technique were: (1) Field conditions should be simulated, (2) the results should be uniform and reproducible, and (3) it should permit selection and removal of seedlings for propagation. Sand culture was preferred to water culture because rooting was normal and seedlings could be selected after emergence from a uniformly highly saline planting medium.

Methods and Materials

Seed of the open-pollinated, self-sterile line C17 (8) was used to develop and evaluate the sand-culture technique. The seed was decorticated in a rice huller to simulate commercially processed seed. Ran-

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[2]Numbers in parentheses refer to literature cited.

[3]Dexon is a trademark name for a formulation of p-(Dimethylamino) benzenediazo sodium sulfonate manufactured by the Chemagro Corporation, Farbenfabriken Bayer AG (West Germany). Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture, and it does not imply approval to the exclusion of other products that may be suitable.

domly selected seeds were treated with p-(Dimethylamino) benzenediazo sodium sulfonate (Dexon)[3] immediately before testing. Temperatures of 24 and 34°C were maintained in a constant-temperature room, with temperature fluctuations of about ± 0.5°C. Fluctuations in sand temperature at seed depth were not detectable. Experiments at 24 and 34°C were not conducted concurrently, because only one constant-temperature room was available.

The composition of the salt solution was prepared to approximate a concentrated solution of Colorado River water. Salts were added in the proportion of: $NaHCO_3$, 27.9 g; NaCl, 15.8 g; $CaCl_2 \cdot 2H_2O$, 15.9 g; $MgSO_4 \cdot 7H_2O$, 49.0 g; $CaSO_4 \cdot 2H_2O$, sufficient for saturation. The salt solution was mixed and adjusted to approximately 26 mmhos/cm at the temperature for the test. The solution was permitted to equilibrate for 2 or more days at test temperature and was then readjusted to 26 mmhos/cm. Filtered Colorado River irrigation water, with an electrical conductivity of 1.3 mmhos/cm at 25°C, was used as a control solution at both test temperatures.

Technique for Sand Culture

Fine plaster sand was washed, dried, screened, and fumigated with methyl bromide. All equipment used in the test was treated with a disinfectant solution. Salt solution was mixed uniformly in the sand at 4.7%, air-dry basis, as recommended in Agriculture Handbook 30 (13). This step was important because preliminary experiments indicated that excessive water in the sand caused erratic seedling emergence. After it was mixed with the salt solution, the sand was immediately enclosed in plastic bags to prevent water evaporation. Sand in the amount of 5.8 kg was sieved into the bottom of metal flats 50 × 35 × 7 cm, and the sand was leveled. Rows were spaced 2 ½ cm apart, and 50 seeds were distributed evenly along each row. Sand (2.8 kg) was sieved over the seed with a 0.6-cm mesh screen and was leveled to a uniform depth. This produced a planting depth of about 1.3 cm. Each flat was enclosed in a double-folded 4-mil polyethylene bag, and the ends were sealed with masking tape.

Data presented in this paper were obtained in a standard test with several experimental sugarbeet lines. Fifty seeds of each line were planted. The lines were randomized within the flat. Each flat represented one replicate, and there were eight replicates. Seedlings were counted 3, 7, 10, 14, 21 and 27 days after initiation of each test. At each date, the flats were carefully unwrapped to prevent seedling breakage, seedling counts were taken, and the flats were rewrapped as quickly as possible to prevent water loss. Early-emerged seedlings were removed for propagation during seedling counts.

Technique for Petri Dishes

Seed germination tests were conducted in petri dishes, and the results were compared with those in the sand cultures. The petri dishes were sterile plastic, 87 mm in diameter and 1.3 cm deep. Eight replicates of 50 seeds each were used. Blotters were standard 7.5 cm squares with 100 depressions. The blotters were soaked in the salt solution and then blotted to remove the "glisten appearance." After inserting the bottom blotter in the petri dish, seed was distributed uniformly and the top blotter applied. Blotters were changed on the ninth day for the tests at 24°C and were changed weekly for the tests at 34°C. The petri dishes were labeled for identification and randomized in a tray, which was then covered with aluminum foil. Seedling counts were taken after 3, 7, 10, 14, 21, and 27 days, which corresponded with counts made for the sand cultures. The total number of seedlings per 50 seedballs and the number of seedballs germinated were recorded for a comparison with the sand-culture technique.

Results and Discussion

Data obtained in sand culture and in petri dishes are shown in Figure 1. Seedlings developed slightly faster in petri dishes than in

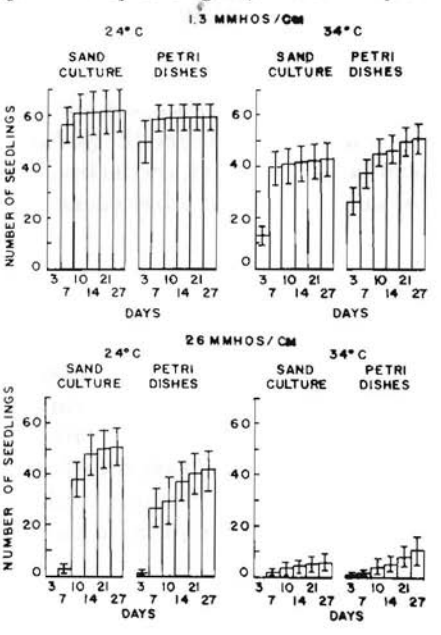

Figure 1.—Seedling emergence in sand culture and total seedlings germinated in petri dishes at 2 salinity levels and 2 temperatures on several dates after tests were started. The bars indicate the standard deviation of the mean for 8 replicates of 50 seeds each.

sand culture, which is indicated by differences in seedling counts at 3 and 7 days on the low-salt treatments. However, seedling numbers were not different after 7 days. Seedling emergence was nearly complete after 14 to 21 days.

The effects of high salinity and high temperatures were about equal in sand and in petri dishes. The high-salinity treatments substantially reduced the rates of seed germination and seedling emergance and reduced the number of seedlings. A temperature of 34°C reduced the number of seedlings, compared with a temperature of 24°C. The decrease was greater at the higher salinity level. Analysis of variance indicated highly significant ($P = 0.01$) reductions in seedling numbers caused by the interaction between salinity and temperature.

The sand-culture technique provided a simple, uniform and reproducible method for testing the salt tolerance of a large number of sugarbeet lines. Seedling emergence in the sand-culture technique was equal to seed germination in petri dishes. The standard deviation of the mean was no greater with the sand-culture technique than with petri dishes. Mold was never a problem, as it sometimes is in petri dishes. The sand provided adequate water and space for root development before the transfer of seedlings to other containers for propagation. Transferred seedlings had a nearly perfect survival rate. Water loss during the tests was minimized by enclosing the metal flats in a plastic bag. The sand lost less than 40% of its water content in 28 days, including water uptake by the seedlings. Since the temperature and salinity level can be easily varied, the technique is readily adapted to the evaluation of salt tolerance of other economic plants in addition to sugarbeets.

Summary

A simple and reproducible technique in sand culture was developed for evaluating the salt tolerance of sugarbeets at the stages of seed germination and seedling emergence. Mean germination and the standard deviation of the mean with sand culture were the same as with petri dishes. Sand culture permitted easy transfer of seedlings to other containers for propagation. The technique could be used to evaluate the salt tolerance of other crops in addition to sugarbeets.

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