Advanced Screening Method for The Selection of Salt Tolerant Rice and Barley Using Agar Plate

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Abstract

It was clearly illuminated the detectable pH were ranged from 4.0 to 5.2 or from, 6.0 to 5.2. At initial pH 6.0, the badge seed became to circumference fluctuates of the pH which will add cation like NH4+. This seemed to be caused by germination of seed and exhaust passage of the H+ together. The germination of rice was almost advanced in the normality from the NaCl under 250mM. In at the initial the germination was quickly progressed under 250mM Nacl. Especially in barley, the area of changed color were largest in untreated control, and then 250mM, 100mM, and 50mM in order. The area of changed color initial pH 4.0 was smaller than pH 6.0. The change in rice relatively less than in barley. The color changed area in rice were 50mM > 0mM > 100mM > 250mM in order.

In rice, grown with CaCl2, there were few changes as like as treated with NaCl. However, pH change of rice was most fast in 50mM. The color of NH4+ agar medium treated with CaCl2 fainted or changed to yellow compare with or without Nacl.

Key words: Bio Assay, Agar Plate, Salt Stress, RGB, Rice(Oryza sativa L.), Barley(Hordeum vulgare var. hexastichon)

Introduction

Salt stress, one of the major abiotic stresses, reduced agricultural productivity. Therefore, many efforts have been tried to find the mechanisms about salt tolerance and to produce salt tolerant crops. Field assessment of salt tolerance in crop plants were very difficult because of the variability of salinity levels and the enormous environmental factors within soil. Evaluating tolerance is made more complex by variation in sensitivity to salt during the life cycle (Flowers, 2004). Agar technique with dye indicators, measuring the spatial variability of pH changes in the rhizosphere, have been applied to qualitative assay of metabolic root functionality of seedling (Braccini et al, 2000; Kopittke et al, 2004). Some resercher provided beautiful images of pH changes and variation along the root system. Others coupled the dye indicator/agar gel technique with the use of an optical densitometer to provide a more quantitative estimate of pH changes in the rhizosphere (Gregory and Hinsinger, 1998). The pH changes on agar plate were photographed using digital camera. For the standardization, agar media were adjusted to the desired. Kim(2003) had suggested that the phytotoxicity can be identified through atar plate method. On the basis of color, each standardized plate were monitored and digitized on a computer. Ku et al.(2004) had reported that the numeric RGB values on a program photoshop could be directly used to evaluate the chlorophyll contents in rye leaf. The present investigation studies nondestructive evaluation of agar plate technique for the selection of salt tolerant crops.

Materials and Methods

Seeds of rice and barley were rinsed with deionized water and then soaked in 5% NaOCl for disinfection. For germination test, 5 to 7 seeds were immersed on 1% agar plate with 0~250mM Salt(NaCl, CaCl2) and pH indicators. For seedling test, one seedling was immersed on 1% agar plate with 0~250mM Salt(NaCl, CaCl2) and pH indicators. Bromocresol purple with 1 mM NH4Cl and bromocresol blue with 1mM NaNO3 were used to detect the pH changes during the germination and seedling growth on agar plate in the range from 6 to 4 or from 4 to 6, respectively. The changes of colors on the agar plate were captured by digital camera. Colors were transformed to numeric RGB (red/green/blue) values using Sigma Scan Pro 5 program, then the relationships between the pH and RGB values were obtained. Finally, the changes of (RGB Area) values on the agar plate during the germination and the seedling growth of gramineous crop were compared within treatments.
Results and Discussion

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Fig. 1. Changes in pH(6.0 →4.0) during the germination of barley seeds under NaCl.

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<th>NaCl Treatment</th>
<th>CaCl₂ Treatment</th>
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Fig. 2. Changes in rhizospheric pH(4.0→6.0 and 6.0→4.0) of rice seedlings grown in NaCl and CaCl₂.

Conclusion

Color indicators bromocresol purple and cromocresol blue may differentiate pH range from 5.2 to 6.0 and from 4.2 to 6.0, respectively. The pH changes near root were detected within 2 hours. The tests of germination and young seedling on agar plate with pH indicators can rapidly detect the salt stress.

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References


