Molecular Genetical Analysis of Mechanisms of Salt Tolerance in Plants

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Summary

Plants associated with the ability of photosynthesis utilizing solar energy bring foods and oxygen gas to us, without which animals including the human cannot sustain their lives. Although irrigation is needed to make dry desert areas green, accumulation of salts derived from sandy soil on the land surface by desiccation is a serious problem even when irrigation is partially possible. Irrigation with diluted sea water may be available in areas closed to sea. In any cases, it is desired to confer the salt tolerance ability on plants. Therefore, we intended to characterize genes regulating to make plants salt tolerant and further bestow that ability on cultivating plants. There seems to be a limitation of biochemical approaches with analysis of proteins due to the possible trace amounts and instability of proteins responsible for expression of genes for salt tolerance. To overcome this limitation, we have applied genetical methodology to the model plant *Arabidopsis thaliana*, which is suited for mutagenesis and the subsequent gene cloning by complementation.

It is speculated that the rate of formation of the photosynthetic machinery is reduced under salt stress conditions, resulting from suppression of constitutive expression of genes for photosynthesis. We have focused on well known genes for photosynthesis, the genes for the small subunit of ribulose 1,5-bisphosphate carboxylase/oxygenase (rubisco) (*RbcS*) and for chlorophyll *a/b*-binding proteins (*Cab*, *Lhc*), especially their gene members *RbcS-3B* and *Cab1* (*Lhcb1*At3*) of *A. thaliana*. In order to monitor their expression, several reporter genes were placed under the controls of their promoter regions including their upstream sequences. Chlorophyll contents of *A. thaliana* decreased and the plant died when it was exposed to 200 mM NaCl for 12 days or 250 mM NaCl (approx. a half of NaCl concentration in sea water) for 7 days on solid medium containing elementary minerals. The expression of *RbcS-3B* and *Cab1* as demonstrated with the reporter genes is dramatically reduced prior to the death of *A. thaliana* under such salt conditions. The 8,200 lines of M2 plants after ethylmethanesulfonate (EMS) treatment and 4,900 lines of T-DNA insertion mutagenesis have been screened for salt tolerance or salt hypersensitiveness, resulting in finding a few mutants of salt tolerance (ste) and of salt hypersensitiveness. We are continuously screening EMS-treated lines.

There are two mechanisms proposed for salt tolerance, the accumulation of osmoticants in the cells and the strengthened activities of ion or water pumps. We are analyzing which mechanism is involved in the ste mutant candidates. Organisms have been evolved under sea conditions to adapt to living on land, expecting us that land plants may potentially have the ability of salt tolerance which must be usually suppressed. Therefore, it is not unexpected that plants may become salt tolerant if genes for repressing the ability is disrupted by mutagenesis.