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Isolation of two phytochelatin synthases (*PCSs*) from rice

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Objectives

We have tried to identify heavy metal tolerance genes from rice (*Oryza sativa*) and expressed them in *saccharomyces cerevisiae* and *Arabidopsis thaliana* for heavy metal tolerance.

Materials and Methods

1. Material

Plant - *Oryza sativa*, *Arabidopsis thaliana*.

Yeast strain - *ycf1* mutant (DTY167) and wild type (DTY165).

Agrobacterium strain GV3101

2. Methods:

We transformed rice cDNA library in yeast *ycf1* mutant that show hypersensitive to Cadmium as compared with wild type yeast. To isolate heavy metal resistance genes, resistant colonies were isolated in 50 μ M cadmium media. Two full-length *PCS* cDNAs were expressed in *ycf1* mutant and wild type. Transgenic yeast strains were tested tolerance to cadmium.

Results and Discussion

Two partial rice phytochelatin synthesis (*PCS*) cDNAs, *OsPCS1* and *OsPCS2*, were identified by cadmium resistance screening in yeast mutant. In an attempt to know the heavy metal resistance of yeast cells, these two full-length *PCS* cDNAs were expressed in *ycf1* mutant (DTY167) and wild type (DTY165). Transgenic yeast strains were selected and analyzed for tolerance to cadmium. The result showed that two *OsPCS* genes could confer enhanced cadmium tolerance in both *ycf1* mutant and wild type yeast cells. Interestingly the gene expression of two *PCS* genes was highly induced by heavy metal in rice. The localization of *OsPCS* in cytosol was determined by the localization of *OsPCS1* tagged with green fluorescent protein in the yeast and plant cells by using confocal fluorescence microscope.

In this report, we discuss the application of *PCS* genes to construct higher plants with highly increased accumulating abilities of heavy metals for phytoremediation.

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