CLONING AND EXPRESSION OF A SERINE RACEMASE GENE HOMOLOGUE OF THE GREEN ALGA CHLAMYDOMONAS REINHARDTII AND CHARACTERIZATION OF THE GENE PRODUCT.

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A unicellular green alga *Chlamydomonas reinhardtii* (*C. reinhardtii*) has served as a model system to study many fundamental biological processes. We demonstrated that some D-amino acids have no inhibitory effect on the growth of *C. reinhardtii* and the green alga has alanine racemase and D-threonine aldolase. The homologous gene of serine racemase was found on the genome sequence of *C. reinhardtii*. In this study, a homologous gene of serine racemase on the genome of *C. reinhardtii* was cloned and expressed in *E. coli* cells, and the gene product was purified and characterized.

Total RNA was extracted from *C. reinhardtii* cells. Sense and antisense primers were designed for PCR based on the upstream and downstream regions of the putative gene for serine racemase. First strand cDNA was synthesized from the mRNA and the antisense primer. Amplification of nucleotides between the two primers was performed with the cDNA. The fragment (*ser-h*) was sequenced. The deduced protein consisted of 340 amino acids with a molecular weight of 35,300.

The amino acid sequence of the protein showed similarities to the reported serine racemases; *Oryza sativa*, 55%; *Mus musculus*, 52%; *Schizosaccharomyces pombe*, 39%. A modified serine racemase homologous (*ser-h*) whose codons were optimized for E. coli was synthesized and used to construct pET24/*ser-h*' and to transform BL21 (DE3). SDS-PAGE of the crude extract revealed that the gene product was overexpressed. The gene product was purified to electrophoretic homogeneity from the recombinant cells using ammonium sulfate fractionation and Column chromatography. Further characterization and crystallization of the enzyme are currently under study.