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GENE FOR DEGRADATION OF ORGANOMERCURIAL COMPOUNDS AND ITS APPLICATION TO BIOREMEDIATION OF MERCURY CONTAMINATION

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Abstract

Organomercury lyases (MerBs) are key enzymes in bacterial degradation and detoxification of organomercurial contaminants, and essential for bioremediation of environmental pollution caused by several types of organomercurials. MerBs are encoded by merB-genes in mer operons of environmental mercury-resistant bacteria. Each bacterial mer operon contains none to three merB genes. In this study, first, organomercury removal capabilities and specificities of *Bacillus megaterium* MB1, *Bacillus megaterium* MK64-1, *Bacillus cereus* RC607, *Staphylococcus aureus* RN23, and *Pseudomonas* sp. K-62 that are possessing merB genes were tested as wild-type organomercurial-resistant bacterial strains. Second, to investigate the substrates specificities of MerB enzymes, the different merB genes, i.e., merB1, merB2, and merB3 genes from *B. megaterium* MB1, merB1 and merB2 genes from *Pseudomonas* sp. K-62, and a merB gene from *S. aureus* RN23, were cloned into *Escherichia coli* with the mercuric ion reductase gene (merA) from *B. cereus* RC607 and the organomercury removal (volatilization) capabilities and specificities of the *E. coli* recombinants were investigated. Five chemical species of organomercurial compounds, i.e., methylmercury chloride (MMC), ethylmercury chloride (EMC), and phenylmercury acetate (PMA), thimerosal (TH) and p-chloromercuribenzoate (PCMB) were used as substrates for the removal by the wild-type mercury resistant bacterial isolates and the recombinant *E. coli* strains. The assay results showed that all *Bacillus* species used in the assay degraded and volatilized MMC, EMC, TH and PCMB rapidly, *S. aureus* RN23 volatilized MMC, EMC and TH but slightly volatilized PCMB, and *Pseudomonas* sp K-62 volatilized EMC, TH and PCMB but slightly volatilized MMC. MerBs cloning assay using *E. coli* strain showed that the merB1 gene from *B. megaterium* MB1 conferred the highest removal abilities against MMC, EMC, TH and PCMB, while the merB1 gene from *Pseudomonas* sp. K-62 conferred the highest removal ability against PMA. The recombinant *E. coli* strain that produced MerB1 protein of *B. megaterium* MB1 conferred the highest mercury volatilization abilities to MMC, EMC, and TH, while the recombinant strain that produced MerB3 protein of *B. megaterium* MB1 conferred the fastest mercury volatilization activity against PCMB. The recombinant strain that produced MerB2 protein of *B. megaterium* MB1 could slightly decompose EMC, TH, and PCMB but not MMC and PMA. These results suggested that there are organomercurial substrate specificities among these six MerB enzymes. The substrate specificities indicate importance of selection of the appropriate bacterial strains or merB genes to apply them in bioremediation engineering for cleaning up specific organomercurial pollution sites.
