

Overproduction of trehalose by recombinant *Escherichia coli* in the presence of validamycin A and high osmolarity

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In this presentation, we explored the trehalose production in E.coli by over-expressing trehalose biosynthetic operon (otsBA) encoding trehalose-6-phosphate phosphatase and trehalose-6-phosphate synthase as well as inhibiting the action of trehalase in the cells. These genes were amplified by PCR, sequenced and cloned into pTrc99A vector. The recombinant DNA was introduced into E.coli cells. The productivity of trehalose was investigated in the fermentation of recombinant E.coli cells in M9 media with or without high osmolarity or validamycin A. In vitro enzyme assays using cell-free extract were also performed to verify the action of trehalase when trehalose degradation occurred. It was found that trehalase activity in the cells was the highest under the high osmotic pressure. Addition of trehalase inhibitor, validamycin A into medium inhibited the trehalase action in the cells and thus significant enhancement of trehalose production in the recombinant cells was observed under the osmotic pressure.