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The production of 1,3-propanediol from mixtures of glycerol and glucose by a *Klebsiella pneumoniae* mutant deficient in carbon catabolite repression

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HIGHLIGHTS

- We prepared a *crr* mutant strain of *Klebsiella pneumonia* mutant, in which carbon catabolite repression (CCR) was abolished.
- Production of 1,3-propanediol from glycerol upon fermentation of the *crr* mutant increased by the presence of glucose.
- This is the first report to show that elimination of CCR in *K. pneumoniae* strains used for 1,3-PD biosynthesis.

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ABSTRACT

In the present study, mutant strain of *Klebsiella pneumoniae* with deletion of the *crr* gene encoding $EIIA^{Glc}$ (a component of the glucose-specific phosphoenolpyruvate-dependent transferase system [PTS]) was prepared. This eliminated the ability of the strain to mediate carbon catabolite repression (CCR). Production of 1,3-propanediol (1,3-PD) from glycerol by the *crr* mutant strain was enhanced (compared to that of the parent) in the presence of glucose. Using molasses as a co-substrate of glycerol, the maximum yield of 1,3-PD was 60.4% greater (81.2 g/l) than that obtained when glycerol was used alone, under optimum fermentation conditions.

1. Introduction

1,3-Propanediol (1,3-PD) is a platform substrate of the chemical industry and can be synthesized either from petrochemical or from renewable resources such as crude glycerol derived from the biodiesel industry. The market for 1,3-PD was initially rather small, but is currently over 100 million pounds per year and growing rapidly (Kraus, 2008). 1,3-PD monomer is useful in the manufacture of polyurethanes, various cyclic compounds, and the polyester polytrimethylene terephthalate (PTT). PTT can be much more resistant to dirt than are polyethylene threphtalate (PET) and polybutylene threphtalate (PBT) (Biebl et al., 1999; Xiu et al., 1999; Zeng and Hiebl, 2002).

Microbial production of 1,3-PD has attracted worldwide research interest because petroleum availability is limited. DuPont and Genencor have developed a glucose-based process using a recombinant Escherichia coli strain carrying the genes required for production of glycerol by *Saccharomyces cerevisiae* and *Klebsiella pneumoniae* genes mediating the biosynthesis of 1,3-PD from glycerol (Nakamura and Whited, 2003). However, it is necessary to supply coenzyme B_{12} as a cofactor of an enzyme involved in 1,3-PD biosynthesis; this likely increases production costs (Mendes et al., 2011). Coenzyme B_{12} biosynthesis is limited to a few representatives of microorganisms, such as *Clostridium, Enterococcus, Lactobacillus*, and *Klebsiella* (Sakai et al., 2007). This coenzyme supply is not required when *K. pneumoniae*, a natural producer of 1,3-PD from glycerol, is employed; the strain can synthesize the costly coenzyme *de novo* (Luo et al., 2011). Currently, vast amount of crude glycerol is produced as the major byproduct of biodiesel manufacture. Glycerol fermenting *K. pneumoniae* are thus in

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