

Desulfurization Activity Evaluation of Recombinant *Klebsiella Oxytoca* ISA4

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Background & Objectives: In order to minimize the adverse effect of sulfur emissions from power plants and transportation fuels regulations have become increasingly strict. Oil refinery technology use physicochemical processes, such as hydrodesulfurization (HDS) to reduce sulfur content of diesel. However, polyaromatic sulfur heterocycles such as dibenzothiophene (DBT) are recalcitrant and the least reactive to HDS. Biodesulfurization (BDS) has been studied as a potential technology to complement or alternative the traditional HDS approaches. In this process, microorganisms have been applied which can selectively remove sulfur atoms from organosulfur compounds.

Methods: A Bacterium designated as *Klebsiella oxytoca* ISA4, was isolated from oil-contaminated soil of Ahvaz, selected as the host due to its inability to catabolize DBT as a sole source of carbon and energy, solvent-tolerant and biosurfactant production characteristics. To improve biodesulfurization activity, the dszABC cluster from *Rhodococcus erythropolis* IGTS8 was amplified and cloned using pVLT31 vector in indigenous *K. oxytoca* ISA4 to express under control of tac promoter. In addition, the Gibbs' assay and GC analysis were applied to measure the amount of 2-HBP production in this recombinant strain and compared its characteristics with those of *R. erythropolis* IGTS8 as a standard strain and *Pseudomonas auroginosa* pTSOX4 as a positive control. Duncan comparison tests were used to detect significant differences at a confidence level of 95% ($P < 0.05$).

Results: Further studies by Gibbs' assay and GC analysis revealed that recombinant *K. oxytoca* ISA4 harboring dszABC genes exhibited the highest desulfurization ability (48%) in comparison with *R. erythropolis* IGTS8 (42%) and *P. aeruginosa* pTSOX4 (46%) within 72 h in stationary phase.

Conclusion: This bacterium is a promising biocatalyst for desulfurizing system due to ability in production of biosurfactant which accelerates emulsification of organic phase. In addition, its desulfurization capability was higher than *R. erythropolis* IGTS8 according to analytical & statistical studies.

Keywords: Biodesulfurization; DBT; *Klebsiella Oxytoca*; DszABC