Abstract

The biodegradation of petroleum products resulting from microorganism activities is the process in which hydrocarbons and other petroleum compounds are disposed. Therefore, the improper and uncontrollable growth of microbes leads to the microbial contamination of the petroleum and gas reservoirs and the bio-degradation of fuel and corrosion which ultimately causes failure in the storage and transportation industries of petroleum products. Considering this point that the biggest microbial problem in petroleum industry is related to the contamination of petroleum products, the need for a reliable but low cost method for detection of microorganisms in the petroleum industry is sensible. In this study, the identification of *Pseudomonas stutzeri* bacteria as a case of contaminating microorganism of petroleum reservoirs was evaluated by multiplex PCR and colorimetric gold nanoparticles assay. In multiplex PCR technique, designed primers for *catA* and *nirP* genes were used and the results indicated the proliferation of two bands of 512 bp and 249 bp for *catA* and *nirP* genes respectively. The study of sensitivity and specificity of multiplex PCR reaction using 5 bacteria was done. The results respectively indicated the specificity of 100% and sensitivity of 0.048 ng/µL and 0.048 ng/µL of genomic DNA for *catA* and *nirP* genes. Then, probes were designed to perform colorimetric assay with gold nanoparticles. Sensitivity of reaction was compared by using NaCl and KCl solutions that results in 0.0005 ng/µL and 0.0001 ng/µL of genomic DNA for NaCl and KCl respectively. The reaction specificity was 100%. The results of this study indicated that the colorimetric technique on gold nanoparticles was more sensitive than multiplex PCR technique. Moreover, the KCl electrolyte led to high sensitivity of colorimetric technique.

Keywords: biodegradation of petroleum compounds, *Pseudomonas stutzeri*, Multiplex PCR, gold nanoparticles, *catA* gene, *nirP* gene

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