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Biodegradation of 2,4-Dichlorophenol by Pseudomonas chlororaphis Strain Isolated from Activated Sludge Sample from a Wastewater Treatment Plant in Durban, South Africa

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Abstract : Agricultural and industrial activities have led to increasing production of xenobiotics such as 2,4-dichlorophenol (2,4-DCP), a derivative of 2,4-dichlorophenoxyacetic acid (2,4-D), which is a widely used herbicide. Bioremediation offers an efficient, cost-effective and environmentally friendly method for degradation of the compound through the activities of the various microbial enzymes involved in the catabolic pathway. The aim of this study was to isolate and characterize bacterial isolate indigenous to contaminated sites in Durban, South Africa for 2,4-DCP degradation. One bacterium capable of utilizing 2,4-DCP as sole carbon source was isolated using culture enrichment technique and identified as Pseudomonas chlororaphis strain UFB2 via PCR amplification and analysis of 16S rRNA gene sequence. This isolate was able to degrade up to 75.11% of 2,4-DCP in batch cultures within 10 days, with the degradation rate constant of 0.14 mg/l/d. Phylogenetic analysis revealed the relatedness of this bacterial isolate to other Pseudomonas sp. previously characterized for chlorophenol degradation. PCR amplification of the catabolic genes involved in 2,4-DCP degradation revealed the presence of the correct amplicons for phenol hydroxylase (600 bp), catechol 1,2-dioxygenase (214 bp), muconate isomerase (851 bp), cis-dienelactone hydrolase (577 bp), and trans-dienelactone hydrolase (491 bp) genes. Enzyme assays revealed activity as high as 21840 mU/mg, 15630 mU/mg, 2340 mU/mg and 1490 mU/mg obtained for phenol hydroxylase, catechol 1,2-dioxygenase, cis-dienelactone hydroxylase and trans-dienelactone hydroxylase, respectively. The absence of catechol 2,3-dioxygenase gene and the corresponding enzyme in this isolate suggests that the organism followed ortho-pathway for 2,4-DCP degradation. Furthermore, the absence of malaycetate reductase genes showed that the bacterium may not be able to completely mineralize 2,4-DCP. Further studies are required to optimize 2,4-DCP degradation by this isolate as well as to elucidate the mechanism of 2,4-DCP degradation.

Keywords : biodegradation, catechol 1,2-dioxygenase, 2,4-dichlorophenol, phenol hydroxylase, Pseudomonas chlororaphis
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