



Biodegradation of chlorpyrifos and 3,5,6-trichloro-2-pyridinol by *Cupriavidus* sp. DT-1

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HIGHLIGHTS

- ▶ A chlorpyrifos- and 3,5,6-trichloro-2-pyridinol (TCP)-degrading strain (DT-1) belonging to the genus *Cupriavidus* was isolated.
- ▶ The biodegradation pathway of TCP was examined.
- ▶ The *mpd* gene encoding an organophosphorus hydrolase was cloned.
- ▶ Strain DT-1 promotes the degradation of chlorpyrifos and TCP in soil.
- ▶ Strain DT-1 is a good candidate for studying the degradation mechanism of TCP.

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ABSTRACT

A bacterial strain, *Cupriavidus* sp. DT-1, capable of degrading chlorpyrifos and 3,5,6-trichloro-2-pyridinol (TCP) and using these compounds as sole carbon source was isolated and characterized. Investigation of the degradation pathway showed that chlorpyrifos was first hydrolyzed to TCP, successively dechlorinated to 2-pyridinol, and then subjected to the cleavage of the pyridine ring and further degradation. The *mpd* gene, encoding the enzyme responsible for chlorpyrifos hydrolysis to TCP, was cloned and expressed in *Escherichia coli* BL21. Inoculation of chlorpyrifos-contaminated soil with strain DT-1 resulted in a degradation rate of chlorpyrifos and TCP of 100% and 94.3%, respectively as compared to a rate of 28.2% and 19.9% in uninoculated soil. This finding suggests that strain DT-1 has potential for use in bioremediation of chlorpyrifos-contaminated environments.

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1. Introduction

Chlorpyrifos [O,O-diethyl O-(3,5,6-trichloro-2-pyridyl) phosphorothioate] is a broad-spectrum, moderately toxic organophosphate insecticide that is increasingly being used in agriculture because of the restrictions imposed on highly toxic organophosphate compounds (Tomlin and Council, 1994; McConnell et al., 1997). The insecticide inhibits acetyl cholinesterase in an irreversible manner and causes insect death (Karalliedde and Senanayake, 1989). The potential damage by chlorpyrifos to non-target organisms is high because acetyl cholinesterase is present in all

vertebrates (Sogorb et al., 2004). The half-life of chlorpyrifos in soil depends on soil type, climate, and other environmental factors. It is generally between 60 and 120 days, but can range from 2 weeks to over 1 year (Howard, 1991). Initially, it was observed to be hydrolyzed to TCP under alkaline conditions (Racke et al., 1996), but later, the involvement of microorganisms in the hydrolysis of chlorpyrifos was described (Singh et al., 2003).

TCP, a charged molecule at neutral pH, is mobile in soil as well as leachable into groundwater and surface water and thus can widely contaminate soil and aquatic environments (Manclus and Montoya, 1995). TCP is listed as a persistent and mobile pollutant by the US Environmental Protection Agency (Armbrust, 2001). TCP shows relatively high antimicrobial effects on microorganisms (Feng et al., 1997; Cáceres et al., 2007), which prevents its own degradation by microorganisms and also limits chlorpyrifos degradation.

Bioremediation has received increasing attention as a reliable and cost-effective approach to cleaning up polluted environments.

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