

## Simultaneous Degradation of *p*-nitrophenol and Phenol by *Nocardioides* Sp. Nsp41

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A bacterium which can degrade *p*-nitrophenol (PNP) and phenol simultaneously was isolated from an industrial wastewater and identified as *Nocardioides* sp. NSP41. The presence of phenol enhanced the PNP degradation rate, which may be due to the increased cell mass by assimilation of phenol. In the low phenol concentration, 4-nitrocatechol was transiently accumulated during the simultaneous degradation of PNP and phenol. Strain NSP41 degraded PNP via a hydroquinone pathway, while phenol was degraded via a catechol pathway. When grown on a mixture of PNP and phenol, the simultaneous coexistence of the hydroquinone and catechol pathway in strain NSP41 was demonstrated by resting cells and cell extracts.

In order to enhance the efficiency of PNP degradation, the immobilized cells of *Nocardioides* sp. NSP41 were used. In the immobilized cell cultures, although simultaneous degradation of PNP and phenol was maintained, the specific PNP and phenol degradation rate decreased. However, a high volumetric PNP and phenol degradation rate could be achieved by immobilization because of the high cell concentration. Furthermore, when the immobilized cells were reused in the simultaneous degradation of PNP and phenol, they did not lose their PNP- and phenol-degrading activity for one week in semi-continuous cultures.

### Introduction

Nitroaromatic compounds have been used as dyes, pesticides, and explosives. Among these nitroaromatic compounds, *p*-nitrophenol (PNP), a major metabolite resulting from the microbial degradation of parathion or methylparathion, widely occurs as an environmental pollutant during the production of dyes, pesticides, and pharmaceuticals (Popov 1965). Because of its toxicity, PNP is listed as US EPA priority pollutant. Thus, nitrophenols resulting from industrial activities should be eliminated before they enter the environment.

In polluted environments, organic pollutants frequently occur in mixtures with other natural as well as synthetic organic compounds. Because PNP biodegradation can be greatly affected by the presence of other organic compounds, especially toxic aromatic compounds (Zaidi and Mehta 1995), the study on the simultaneous degradation of toxic aromatic compounds is important from a practical aspect.

In this study we identified different metabolic pathways of PNP and phenol degradation by a newly isolated *Nocardioides* sp. NSP41 and investigated the simultaneous degradation of PNP and phenol by freely suspended and immobilized *Nocardioides* sp. NSP41.

## Materials and Methods

### Microorganism

*Nocardioides* sp. NSP41 was isolated from industrial wastewater by selective enrichment as previously described (Cho et al. 1998). This strain was described as a new species of the genus *Nocardioides*, *Nocardioides nitrophenolicus* (Yoon et al. 1999).

### Resting cells experiments

Resting cells were obtained by growth in the mineral salt medium with PNP or phenol. The cells were harvested and incubated with substrates in a rotary shaker. The disappearance of substrates and the formation of metabolites were determined by HPLC and GC/MS.

### Enzyme assays

Nitrophenol oxygenase activity was determined by measuring the nitrite released from the substrate (Kadiyala and Spain 1998). Catechol 1,2-dioxygenase and catechol 2,3-dioxygenase activities were determined spectrophotometrically

### Immobilization

The cells were immobilized in gel beads of calcium alginate. Gel beads of about 1.0-1.2 mm in diameter were obtained by blowing air through the outer tube of a manufactured atomizer (Cho et al. 2000)

### Analytical methods

Cell growth was monitored by measuring the optical density at 600 nm. The concentrations of substrates and metabolites were determined by isocratic reverse-phase HPLC. For the determination of the nitrite ion concentration in the culture fluid, the photometric method was used.

## References

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