

STABILITY OF *Pseudomonas putida* CULTURES DURING THE OFF-GAS TREATMENT OF TOLUENE

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Aromatic hydrocarbons emissions and accidental spills entail a potential risk to natural ecosystems and human health due to their high mobility, toxicity, and in many cases, carcinogenic effects (Fig 1). In this context, biological treatment methods constitute a well-established, low-cost alternative to conventional physical/chemical technologies for the destruction of organic contaminants. Environmental biotechnologies are based on the natural ability of microorganisms to utilize these toxic organic pollutants as carbon and energy source under mild conditions of temperature and pressure.



Figure 1. Toxic oil spill



Figure 2. Effect of Toluene on *P. putida* DOT -T1E

Biological methods exhibit however serious limitations when treating high concentrations of aromatic hydrocarbons, due to their inherent toxicity and mutagenic nature, which ultimately challenges microbial stability. Aromatic hydrocarbons can cause irreversible damage to cell membranes (loss of ions, metabolites, lipids, and proteins, dissipation of the pH gradient and electrical potential, etc.) followed by cell lysis and death (Fig. 2). Operational problems derived from microbial instability in processes treating toluene have been recently reported in literature. For instance, Song and Kinney (2005) reported a decline in the elimination capacity of biofilters subjected to high toluene loadings, likely due to the deterioration of the toluene degrading community, although the mechanisms responsible of this

deterioration were not identified. Likewise, Leddy et al. (1995) reported that the presence of benzoate and benzyl alcohol, commonly accumulating intermediates during toluene biodegradation, resulted in irreversible mutations in the toluene degradation pathways. There is however a lack of systematic studies addressing the influence of toluene-mediated mutations on the overall process performance (i.e. elimination capacity, removal efficiency, CO₂ production, etc.).

This project was launched in collaboration with the Group of Degradation of Toxic Organics from Estación Experimental del Zaidin (CSIC, Granada, Spain) to investigate the pernicious effects of toluene on both the macroscopic bioreactor performance and on the phenotype and genotype of the species *Pseudomonas putida*. The kinetics of microbial growth, metabolite production and the long term stability are being assessed under continuous cultivation using species of *P. putida* with different tolerance (*P. putida* mt-2, F1, DOT-T1E) and at different toluene loading rates. In addition, the role and stability of *P. putida* DOT-T1E efflux pumps will be evaluated.

More information available at:



<http://etuva.blogspot.com.es/>

Relevant publications

Hernandez M, Gouveia J, Segura A, Muñoz R Villaverde S, (2009) Addressing the role of the extrusion pump-bearing pGRT1 plasmid on toluene biodegradation by *Pseudomonas putida* DOT-T1E under real case scenarios. *Water Science and Technology*. 60.9: 2391-2398.

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