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P24

ISOLATION, SELECTION AND IMPROVEMENT OF A *Methylobacterium* spp. STRAIN FOR THE BIOREMEDIATION OF ANTHROPOGENIC ORGANIC COMPOUNDS

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Abstract

The widespread environmental contamination due to the placing of large amounts of persistent chemicals in soil resulting from urban, industrial and agricultural activities, is a problem that is becoming increasingly important in the world. The technologies based on the use of microorganisms able to degrade the pollutant charge, allow to accelerate the natural detoxification processes in the soil environment and represent an alternative method of environmental remediation compared to the harmful traditional methods (incineration and chemical treatment). For this aim we isolated, characterized and selected 14 indigenous bacteria strains obtained from soil of the industrial site ex-ACNA (Associate National Chemical Companies) in Cengio (Savona, Italy) contaminated by different classes of organic compounds (PAHs). The isolation was carried out using a minimal selective solid medium containing 0.5% of Contaminated Soil Aqueous Extract (CSAE) extracted from the soil mentioned above, with or without addition of natural nutrients (1% of soil extract obtained from freshly collected meadow soil). Further biotechnological selection allowed to detect a bacterial strain able to grow in solid medium containing 1.5% of CSAE as sole nutrient source. This strain was characterized and identified by phenotypic (morphological and biochemical tests) and molecular methods (16S rDNAs sequence analysis of ribosomal genes) as belonging to *Methylobacterium* spp. (97%).

The ability of *Methylobacterium* spp. strain to grow in the highly contaminated habitat was enhanced using serially enrichment strategy in liquid media containing increasing concentrations of CSAE (up to 40%), with or without addition of nutrients (1% soil extract obtained from meadow soil). During enrichment experiments, the growth of the bacteria in the liquid culture was determined by spread plate count method using minimal solid media containing the same amount of CSAE. Moreover, to demonstrate the degradative capacities of *Methylobacterium* spp. strain, GC-MS analysis of the centrifuged broth cultures supernatant containing 40% of CSAE, was performed. *Methylobacterium* spp. was able to remove partially or completely some pollutants from liquid medium containing CSAE. These results demonstrate the effectiveness of selective ecological strategy that employs indigenous strains naturally present in highly contaminated soils, able to express their potential biodegradation of xenobiotic organic compounds of industrial origin and their potential use to remediate contaminated soil.