

**B304** Diversity and Phylogeny of Oligotrophic Bacteria from Forest Soil

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Sixty bacterial strains used are selected from the obligate oligotrophic Gram-negative bacteria isolated from forest soil at Sendai, Japan. The strains were clustered based on the cellular fatty acid composition for further selection. The strains representing cluster 4a contained C16:0 and C18:0 acids predominantly. 3OH-C14:0 and 3OH-C16:0 acids were also found as the characteristic components. Their quinone system is Q-8. Seven strains of this cluster were divided into three DNA homology groups. They are accommodated in the cluster of the genus Burkholderia in the Proteobacteria beta subdivision. Cluster 3 was characterized by the presence of branched-chain fatty acids, iso-C15:0, iso-C17:1, and iso-C17:0. Based on the 16SrDNA sequence analysis, the two representative strains of cluster 3 showed the close relation to the genera, Xanthomonas and Stenotrophomonas, but were not included in these genera. The isolates with Q-10 are corresponded to the two large groups in Proteobacteria alpha subdivision. One was incorporated in the genus Bradyrhizobium cluster, which also includes Agromonas, a genus for oligotrophic bacteria. The oligotrophic bacteria studied here showed diversity and indicated the close relationship to established genera of non-oligotrophic bacteria.

**B305** Detection and identification of substituted nitrotoluenes as intermediates of bacterial degradation of TNT

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Degradation of 2,4,6-trinitrotoluene (TNT) was studied using a 1.5-L stirred tank reactors and a bacterial culture which had been originally derived by enrichment with s-triazine herbicide, atrazine. Complete depletion of TNT was achieved within 14 days of incubation. High pressure liquid chromatographic methodology was used to measure TNT and it also resolved two intermediates, 2-ADNT (2-amino-4,6-dinitrotoluene) and 4-ADNT (4-amino-2,6-dinitrotoluene). Gas chromatography-mass spectrophotometry (GC-MS) was used to verify the intermediates. Total ion chromatogram (TIC) displayed four major and a few minor peaks. The major TIC peaks yielded positive identification based on mass/charge by MS for TNT (TIC peak A at 8.591 min), 4-ADNT (TIC peak B at 9.963 min), and 2-ADNT (TIC peak C at 10.301 min), respectively.