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An Obligately Methylophilic Isolate Producing Extracellular Polysaccharide during Growth on Methanol

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An obligately methylophilic bacterium which produces extracellular polysaccharide (EPS) was isolated from water samples from a creek near Nanji island, Seoul, through methanol-enrichment culture technique. The isolate was nonmotile and Gram-negative rod. The colonies were small, pale-yellow, and raised convex with entire circular margin. The cell did not produce spores and capsular materials. The cell was obligately aerobic and exhibited catalase, but no oxidase, activity. Plasmid, carotenoids, and poly- β -hydroxybutyric acid were not found. The guanine plus cytosine content of the DNA was 50-55%. The isolate was found to grow only on methanol and methylamine. Growth factors were not required. Growth was optimal ($t_d = 2.4$ h) at 35-40°C and pH 6.5 in a mineral medium supplemented with 0.5% (v/v) methanol, 25 mM phosphate and 1.0 g/l ammonium sulfate. No growth was observed at over 50°C. Growth was inhibited by tetracycline and streptomycin. Methanol was assimilated through the ribulose monophosphate pathway. EPS was produced most abundantly (1.4 g/l) in cells growing at the mid-stationary growth phase at 30°C in a mineral medium (pH 6.5) supplemented with 1.0 % methanol (v/v) and ammonium sulfate which was added in the C/N molar ratio of 24.6.

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Purification and Characterization of Ribulose Biphosphate Carboxylase/Oxygenase from *Acinetobacter* Sp. Strain JCI DSM 3803

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Ribulose biphosphate carboxylase/oxygenase (RubisCO) was purified to homogeneity by conventional method from cells of *Acinetobacter* sp. strain JCI DSM 3803 grown on carbon monoxide (CO) and methanol, respectively. Molecular weight of the native enzymes were estimated to be 520,000. Sodium dodecyl sulfate-gel electrophoresis of the enzymes revealed two nonidentical subunits of molecular weights 53,500 and 15,000. The K_m of CO₂ and ribulose biphosphate were 36.7 mM and 3.7 mM, respectively. The V_{max} of CO and ribulose biphosphate were 296.1 nmol/mg protein/min and 770 nmol/mg protein/min. The enzymes from methanol- and CO-grown cells showed difference in the susceptibility to protease treatment and also in the pattern of one-dimensional peptide map. The enzymes, however, were found to be identical in immunological properties and also in N-terminal amino acid sequences of the large- and small-subunits, but were found to have no immunological relationship with RubisCOs of *Hydrogenophaga pseudoflava*, *Pseudomonas carboxydohydrogena*, and *Oligotropha carboxidovorans*.