

bisphosphate carboxylase/oxygenase (RubisCO) activity was also detected in cell-free extracts prepared from all cells, except *M. tuberculosis*, grown on CO. The RubisCOs in cell-free extracts of *M. smegmatis* and *M. neoaurum*, however, did not cross react with antibody raised against *Mycobacterium* sp. strain JCI RubisCO when it were subject to Western blot analysis. It was found that the mycobacterial species grown on methanol did not exhibit activities of methanol dehydrogenase, hydroxypyruvate reductase, and hexulose phosphate synthase, except that *M. gastri* showed hexulose phosphate synthase and NDMA-dependent methanol dehydrogenase activities.

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Characterization of Recombinant Gal β 1,4GlcNAc α 2,6-sialyltransferase Expressed in Insect Cells

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Gal β 1,4GlcNAc α 2,6-sialyltransferase (ST6Gal I) catalyses the reaction responsible for the attachment of sialic acid to N-linked glycoproteins. A truncated form of ST6Gal I, lacking 71 amino acids from N-terminus was produced in insect cells by using baculovirus expression system. For secretion out of the cell and easy purification, signal peptide of a mouse-derived IgM (20 amino acids) and part of protein A was inserted in front of ST6Gal I gene in frame. Immunoblot analysis done with rabbit antisera showed that the recombinant ST6Gal I was secreted into the culture media indicating that mammalian IgM signal peptide is effective for secretion of recombinant protein into the media in insect cells. The recombinant ST6Gal I was purified by immunoaffinity column and its biochemical characteristic was analyzed. The

recombinant ST6Gal I retains a biological activity that catalyzes the transfer of sialic acid from CMP-NeuAc to the carbohydrate groups of asialofetuin. These results suggest that massive amount of biologically active form of sialyltransferase could be produced from baculovirus expression system.

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Characterization of Recombinant Gal β 1,3GlcNAc α 2,3-sialyltransferase Expressed in Insect Cells

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Gal β 1,3GlcNAc α 2,3-sialyltransferase (ST3Gal III) catalyses the reaction responsible for the attachment of sialic acid to N-linked glycoproteins. A truncated form of ST3Gal III, lacking 28 amino acids from N-terminus was produced in insect cells by using baculovirus expression system. For secretion out of the cell and easy purification, signal peptide of a mouse-derived IgM (20 amino acids) and part of protein A was inserted in front of ST3Gal III gene in frame. Immunoblot analysis done with rabbit antisera showed that the recombinant ST3Gal III was secreted into the culture media indicating that mammalian IgM signal peptide is effective for secretion of recombinant protein into the media in insect cells. The recombinant ST3Gal III was purified by immunoaffinity column and its biochemical characteristic was analyzed. The recombinant ST3Gal III retains a biological activity that catalyzes the transfer of sialic acid from CMP-NeuAc to the carbohydrate groups of asialofetuin. These results suggest that massive amount of biologically active form of sialyltransferase could be produced from baculovirus expression system.