

crystalline cellulose and mainly generate cellobiose, which is a product inhibitor for these enzymes. We have isolated 4 endoglucanases, 2 exoglucanases, and β -glucosidase from *Trichoderma* sp. C-4, a strain with high cellulolytic activity to determine an optimum hydrolytic condition for cellulose. Combination of endoglucanase 1 and 2 gave highest hydrolytic rate. Addition of exoglucanase 2, exoglucanase 1, and β -glucosidase showed the increase of activity. The presence of 2 endoglucanases, 2 exoglucanases, and β -glucosidase is enough for solubilization of cellulose in vitro, although the roles of remaining 2 endoglucanases are not elucidated clearly.

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Investigation on the Physiological Role of Maltogenic Amylase in *B. subtilis*

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The *bbma* gene isolated from *B. subtilis* SUH4-2, an isolate from Korean soil, encoded a maltogenic amylase and it found to be a homologue of the *yvdF* gene of *B. subtilis* 168 with unknown function, which located in a gene cluster involved in maltose/maltodextrin utilization. *bbma* promoter-*lacZ* gene fusion study in *B. subtilis* 168 suggested that the promoter was most active when the cells were cultured in the medium containing β -cyclodextrin (β -CD), moderately active in the maltose medium, and relatively less active in the starch medium. The promoter was under catabolite repression. Based on the results, the *bbma* gene product was likely to be involved in maltose and β -CD utilization when other sugars, which are readily usable as energy source, are not available. In order to test this

hypothesis, mutagenesis of the *bbma* gene was carried out by Campbell type recombination in both *B. subtilis* 168 and SUH4-2 strains using an internal *bbma* gene fragment. The resulting mutants grew poorly in culture medium containing either 2% β -CD or 2% maltose, but grew as well as wild type strains in 2% starch medium. The β -CD hydrolyzing activities of maltogenic amylase decreased significantly in both strains when they were grown in β -CD or maltose medium. However, temporal expression of the β -CD hydrolyzing activity was slightly different in the two *Bacillus* strains investigated. The results obtained in this study support our working hypothesis on the physiological function of maltogenic amylase in *Bacillus*.

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한국산 버섯 추출물로부터 α -glucosidase의 저해제 탐색과 분리

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버섯 추출물에서 α -glucosidase (당분해효소)의 활성을 저해하는 추출물을 선별한 후 그 추출물로부터 저해제를 분리하였다. 한국의 야산에서 채취한 버섯을 MeOH, 65°C H₂O, 100°C H₂O에서 추출한 후 이 추출물을 이용하여 α -glucosidase의 활성을 억제하는 추출물을 선별하였고, 이로부터 층분리, Silica 크로마토그래피, C-18 MPLC, TLC, 이온교환 크로마토그래피 (NaCl 농도구배, pH 구배), SEPHADEX G-10 겔여과 크로마토그래피, C-18 HPLC등을 통해 저해제를 분리하였다. 그 결과 *Phallus impudicus* (달뚝버섯)에서 α -glucosidase의 저해활성이 가장 높았으며, 여러 단계의 크로마토그래피법을 통해 순수한 물질로 분리해 내었다. 더 많은 노력과 연구를 통하여 아직 밝혀지지 않은 이 물질의 확실한 구조를 밝혀냄으로써 확실한 당뇨병 치료제의

개발에 한 단계 더 나아가려 한다.

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Molecular Cloning of a Nucleoside Diphosphate Kinase (NDPK) Gene in *Aspergillus nidulans* and Biochemical Characteristics of *Aspergillus* NDPK Protein

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NDPK catalyzes the transfer of the terminal phosphate group of a nucleoside triphosphate to a nucleoside diphosphate. Besides to NDPK activity, several other functions including suppression of metastasis in human, phytochrome responses in plants, and regulation of differentiation have been reported. Human NDPK was first isolated as a tumor metastasis suppressor, Nm23-H1. Six isotypes of Nm23 were known to date. Among them, Nm23-H1 and Nm23-H2, also known as the c-myc transcription factor · PuF, coexist as hetero-hexamers. Autophosphorylation and serine/threonine specific protein phosphotransferase activity were also reported for Nm23. However, relevant functions of Nm23 on the various cellular processes are hardly known. To investigate a possibility of using *Aspergillus nidulans* as a microbial model system for Nm23, a Nm23 homolog gene (*ankA*) has been isolated and various biochemical properties of recombinant as well as native NDPK purified using ATP-Sepharose affinity chromatography were examined. *Aspergillus* NDPK of 154 amino acids with 65% sequence similarity to human Nm23 was coded from

an ORF of 462 bp, interrupted by four introns located on chromosome II. The 1.2 Kb transcript was detected in northern analysis. No evidence on the existence of other isotypes was obtained from non-stringent Southern and western analyses. *Aspergillus* NDPK was existed as a homo-tetramer (78 KD) judged from gel filtration chromatography. NDPK and autophosphorylation activities were demonstrated and both enzymatic activities were more thermostable than human NDPK-A. [Supported by a grant from KRF (#1998-001-F00771)].

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Growth of Several Mycobacterial Species on Carbon Monoxide and Methanol

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Mycobacterium sp. strain JC1 DSM 3803 has been known to grow on carbon monoxide (CO) and methanol as the sole source of carbon and energy. We found in this experiment that several other mycobacteria tested such as *M. tuberculosis*, *M. neoaurum*, *M. parafortuitum*, *M. gastri*, *M. peregrinum*, *M. phlei*, *M. smegmatis* and *M. vaccae* were also able to grow on carbon monoxide as sole carbon and energy sources. The bacteria, but *M. tuberculosis*, also utilized methanol as the sole carbon and energy sources. CO-DH assay, CO-DH staining by activity, and Western blot analysis using antibody against *Mycobacterium* sp. strain JC1 CO-DH revealed that CO-DH is present in cell-free extracts prepared from all the mycobacteria grown in 7H9 medium supplemented with CO. Ribulose