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Characterization of Myxobacteria Extracts Against Biological Activity

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Myxobacteria extracts were determined for application in cosmetic field. Firstly, tyrosinase activity was determined and shown in No. 1952 having 20% inhibition. For screening of anti-bacteria activity, growth inhibition of Staphylococcus aureus, Staphylococcus epidermidis, and E. coli K12 were determined. The extracts didn't inhibited growth of Staphylococcus aureus and Staphylococcus epidermidis, but did the growth.

Keywords: biological activity, Myxobacteria, tyrosinase

Purification and Characterization of Cold-Active β-N-Acetylglocosaminidase from Arctic Bacterium Pseudoalteromonas isschenkowii KOPRI 22718

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136 marine bacteria showing chitinolytic activity were isolated from 47 sediment samples of Kara Sea, Arctic. Among them, a psychrotolerant strain (KOPRI 22718) was selected for its cold-activity at 5°C. The sequence analysis of the 16S rRNA affilies KOPRI 22718 to Pseudoalteromonas isschenkowii. An exo-acting chitinase (~100 kDa) was homogenously purified from the culture supernatant of KOPRI 22718 through ion exchange and gel filtration chromatography. The purified chitinase (W-Chi22718) exhibited the highest activity toward pNP-GlcNAc and produced GlcNAc monomer as end-product from chitin oligosaccharides (GlcNAc-GlcNAc)n, due to its β-N-acetylglycosaminidase activity. W-Chi22718 displayed chitinase activities at 0-37°C (optimal temperature of 30°C), and maintained its activities at pH 6.0-9.0 (optimal pH of 7.6). Interestingly, W-Chi22718 exhibited a relative activity of 13% and 35% at 0 and 10°C, respectively, in comparison to 100% at optimal 30°C, which is comparable with those of the previously characterized, cold-adapted, bacterial chitinases. Among the main cations and protein denaturing reagents, W-Chi22718 activity could be enhanced by K+, Ca2+, and Fe3+, but completely inhibited Cu2+ and SDS.

Keywords: Arctic; psychrotolerant, cold-active, exochitinase, β-N-acetylglycosaminidase

Biological Characterization of Glucosidase Isolated from Soil Bacteria

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Glucosidase from previously isolated bacteria was determined and isolated. The glucosidase showed activity at congo red and active staining in native gel. Reactive product was determined in HPLC analysis, and compared with Rg3, reactive product control. Reactive product showed Rg3 peak at the same time. From these results, glucosidase have biotransformation of Rb1 to Rg3, and shows typically biological activity of glucosidase.

Keywords: glucosidase, biotransformation, active staining

Noble Substrate Specificity of UDP-Galactose-4-Epimerase from Escherichia coli

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Sugar isomers have broad substrate specificity, i.e., ribose-5-phosphate isomerase catalyzes not only phosphate sugar but also common sugar as substrate for isomerization. Natural sugar-4-epimerase, in contrast, has been reported catalyzing only a nucleic acid activated substrate, which means its catalytic site has a space for UDP recognition. To understand substrate specificity of sugar-4-epimerase forward common sugars, UDP-galactose-4-epimerase (galE) from Escherichia coli, which catalyzes interconversion from UDP-galactose to UDP-glucose in the galactose metabolism in vivo, has been chosen. Since the GaLE gene has been revealed x-ray crystal structure including catalytic residues and co-factor (NAD+) recognition site, it is considered suitable to protein engineering and evolution purpose for changed substrate recognition. The galE gene was cloned into a pET20b and express protein was purified using Ni-affinity chromatography. The purified GaLE was found having a catalytic activity for D-galactose and the optimal temperature and pH were 35°C and 7.5, respectively. Under the optimal condition, the Km and kcat values were exhibited 26.4 mM and 0.0155 min-1 for D-galactose. When it was compared with kinetic parameter of UDP-galactose, Km was lower 90 fold and kcat was higher 3x106 fold. Among all D-aldohexose and D-ketohexose, galE indicated 4-epimerization activity for glucose, galactose, fructose, tagatose, psicose, and sorbose as substrates. The specific activity for galactose, glucose, psicose, sorbose, fructose and tagatose were 0.47, 0.32, 15.44, 2.08, 2.82 and 9.67 U/mg, respectively. Substrate specificity evolution of GaLE based on 3D-structure analysis and protein engineering are under investigation for 4-epimerization of common sugars.

Keywords: 4-epimerization, common sugar, nucleic acid activated sugar