

Studies on the inhibition activities of various adenosine derivatives on S-adenosylhomocysteine hydrolase

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The inhibitory activities of various analogues of adenosine (Group I, Group II, Group III, Group IV, Group V) were assayed by using recombinant human placental SAH hydrolase. The activity of the SAH hydrolase was determined by measuring the formation of AdoHcy from Ado and Hcy. AdoHcy was analyzed by HPLC using C18 reverse-phase column. The peak of AdoHcy was monitored at 258 nm. Among the tested compounds, fluoroneplanocin A (LJ-276) was the most potent inhibitor.

[PC2-10] [2003-10-10 09:00 - 13:00 / Grand Ballroom Pre-function]

Genomic Fingerprinting of genera Bifidobacterium using Microbial UniPrimer Kit

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The genera Bifidobacterium is a member of the normal intestinal flora in humans, and important in food industry. In order to test the genetic identity of this bacterial genera, four primers originated from rice genome (SRILS Microbial UniPrimersTM kit) were used in molecular typing of 7 Bifidobacterial species and 20 isolates from various source. SRILS Microbial UniPrimersTM kit were effectively applied to genomic fingerprinting of various organism such as plant, animal and microorganism. Using a total set of four primers, it was demonstrated that it may be possible to distinguish all strains and isolates of genera Bifidobacterium. Furthermore, application of this technique may also be reproducible as well as useful and faster than other methods such as phenotypic or biochemical analyses in molecular typing of this bacterial genera. Thus, PCR-fingerprinting method using SRILS UniPrimersTM kit may be applicable in the identification of isolates from various sources or classification of this bacterial genera.

[PC2-11] [2003-10-10 09:00 - 13:00 / Grand Ballroom Pre-function]

Inhibitory Effect of Ginseng on Infection and Vacuolation of Helicobacter pylori

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Panax ginseng C.A. Meyer (Family Araliaceae) was treated at low (60°C, LT), mild (100°C, MT) and high (120°C, HT) temperatures, some components (panaxytriol, ginsenosides and polysaccharides) were isolated, and their inhibitory effects on growth, infection and VacA vacuolation of Helicobacter pylori (HP) were investigated. The molecular weights of polysaccharides were decreased according to the increasing processed temperature. Ginseng polysaccharides inhibited the HP infection into KATO III cells, but did not inhibit HP growth and VacA vacuolation of HeLa cells. HT polysaccharides showed the most potent inhibition with IC₅₀ values of 6.8 mg/ml. Ginseng saponins did not inhibit the infection of HP into KATO cells. However, 20(S)-Protopanaxadiol showed the most potent inhibition of HP growth and vacuolation of HeLa by VacA toxin with IC₅₀ values of 0.05 and 0.067 mg/ml.

[PC2-12] [2003-10-10 09:00 - 13:00 / Grand Ballroom Pre-function]

Screening of New Antibiotics Inhibiting Bacterial Peptide deformylase (PDF)

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Peptide deformylase (PDF) is essential and unique to bacteria for cytoplasmic protein synthesis, but not required in eukaryotes, thus making it an attractive target for the discovery of novel antibacterial drugs. Protein synthesis in eubacteria, under normal conditions, is initiated by formyl-methionyl-tRNA. PDF removes the formyl-group of N-formylmethionine of newly synthesized polypeptides to produce a mature protein. In this study, a pdf gene from *Staphylococcus aureus* 6538p was cloned in pET-14b vector and transformed in *Escherichia coli* BL21 (DE3). PDF protein was overexpressed by addition Isopropyl- β -D-thiogalactopyranoside (IPTG). NH₂-terminal His-tagged PDF protein was purified by nickel-nitrilotriacetic acid (Ni-NTA) metal-affinity chromatography. Enzymatic activity of purified 6xHis-tagged PDF was tested on the substrate, formyl-Methionine-Alanine-Serine (fMAS), by formate dehydrogenase-coupled spectrometric assay of peptide deformylase. For the discovery of new PDF inhibitors from chemical libraries and culture broths of soil bacteria, a target-oriented screening system using a 96-well plate was developed. About 50,000 commercial chemical libraries were tested in this screening system, and about 100 chemicals (0.2 %) among them showed an inhibitory activity against PDF enzyme. This result shows that a new screening system can be used for the discovery of new PDF inhibitors.

[PC3-1] [2003-10-10 09:00 - 13:00 / Grand Ballroom Pre-function]

Anti-angiogenic activity of wilfoside glycosides isolated from *Cynanchum wilfordii*

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Wm was isolated from *Cynanchum wilfordii* (Asclepiadaceae) as a mixture form of polypregnane glycosides that included wilfoside K1N and wilfoside C1N. In the present study, we investigated the anti-angiogenic effect of wilfoside glycosides using in vivo and in vitro assay systems. We first demonstrated that concentrated conditioned media obtained from Wm-treated HepG2 human hepatoblastoma cells blocked the angiogenic activation of Wm-untreated concentrated conditioned media, suggesting that Wm may have an inhibitory effect on tumor-induced angiogenesis. In addition, Wm decreased both neovascularization of chick embryos in the chorioallantoic membrane assay and basic fibroblast growth factor-induced vessel formation in the mouse Matrigel plug assay. Interestingly, the angiogenesis inhibition of Wm was the most dramatic in comparison with those of wilfoside K1N and wilfoside C1N, indicating that wilfoside K1N and wilfoside C1N may have stronger effects when they are co-treated in a mixture form. Moreover, wilfoside K1N reduced tube formation and proliferation of human umbilical vein endothelial cells. Taken together, our present study suggests that wilfoside glycosides may be strong angiogenic inhibitors with a potential of therapeutic application on hypervascularizing tumor cells.

[PC3-2] [2003-10-10 09:00 - 13:00 / Grand Ballroom Pre-function]

In vitro neural differentiation of human embryonic stem cells

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Human embryonic stem (ES) cell lines derived from the inner cell mass of human blastocysts have potential to differentiate into any cell types. We have established in vitro neural differentiation of human ES cells. After the formation of embryoid bodies (EBs), the differentiating EBs formed neural tube-like rosettes in the presence of basic fibroblast growth factor (bFGF). The rosettes were selectively isolated by the treatment of dispase and cultured in a medium for human neural precursors in the presence of bFGF. Finally, after the human neural precursors were cultured in the absence of bFGF, the neural precursors differentiated into three neural lineages, neurons, astrocytes, and oligodendrocytes. The in vitro neural differentiation of human ES cells provides a powerful tool for both basic neuroscience research and therapeutic application.