Rotavirus predominantly occurs the sporadic diarrhea in infants and young children. To prevent rotaviral diarrhea, many kinds of orally administered vaccines against each of the epidemiologically important serotypes have been developed. However, the developed vaccines were not complete for preventing the rotaviral diarrhea until now. Therefore, we screened the inhbitory substances from some traditional herbal medicines on the infectivity of rotavirus. Among tested 60 kinds of herbal medicines, the fruit of *Citrus aurantium* had the most potent inhibitory activity on rotavirus infection. The active components of the fruit of *Citrus aurantium* were neohesperidin and hesperidin. Their 50% inhibitory concentrations were 25 and 10 μM, respectively. These active herbal extracts and the isolated active compounds are believed to contribute to the prevention of the rotaviral illness in some degree.

[PC2-2] [04/21/2000 (Fri) 14:50 - 15:50 / [1st Fl, Bldg 3]]

Biotransformation of Rhaponticin from the Rhizome of Rheum undulatum by Human Intestinal Bacteria and Their Anti-allergic Activity

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During the screening program from discovering hyaluronidase-inhibitory substances from natural products, *Rheum undulatum* (Family Polygonaceae) was found to show inhibitory activity for the activation of hyaluronidase. Rhaponticin isolated from the rhizomes of *Rheum undulatum* (Family Polygonaceae) is metabolized to rhapontigenin and chrysophanol by human intestinal microflora, respectively. Most intestinal bacteria isolated from human feces catalyzed these metabolic pathways. Among rhaponticin and its metabolite, rhapontigenin had the most potent inhibitory activity on a hyaluronidase, a histamine release from mast cell and PCA reaction. The inhibitory activity of rhapontigenin was more potent than that of disodium cromoglycate, one of commercial anti-allergic drugs. These results suggest that rhaponticin in the rhizomes of *Rheum undulatum* should be a prodrug that has an extensive anti-allergic property

[PC2-3] [04/21/2000 (Fri) 14:50 - 15:50 / [1st Fl, Bldg 3]]

Two Novel α-L-Rhamnosidase from Quercitrin-hydrolyzing Fusobacterium K-60

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Recently we isolated *Fusobacterium* K-60, a bacterium transforming quercitrin to quercetin, from human intestinal microflora. We tried to purify α-L-rhamnosidase from *Fusobacterium* K-60, comparing its properties to those of the previously purified enzymes. *Fusobacterium* K-60 produced two kinds of α-L-rhamnosidases, cytosolic and membrane enzymes. The cytosolic enzyme hydrolyzed naringin and poncirin but not quercitrin. Whereas, the membrane enzyme did vice versa. The cytosolic enzyme was purified to homogeneity by 70% ammonium sulfate fractionation, butyl toyopearl, hydroxyapatite, Sephacryl S-300, Q-sepharose column chromatography. The specific activity of purified α-L-rhamnosidase was 2.89 μmole/min/mg protein and its molecular weight was calculated to be 150 kDa by gel filtration. From gel filtration data, it seems to be composed of four identical subunits of 40 kDa with pI and optima pH values of 5.2 and 5.5-7.0, respectively.

[PC2-4] [04/21/2000 (Fri) 14:50 - 15:50 / [1st Fl, Bldg 3]]

Purification and Characterization of Two Novel Chondroitinases from Bacteroides stercoris HJ-15

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Bacteroides stercoris HJ-15, which is human colon Gram-negative rod cell, had been known to degrade acharan sulfate and heparin. Recently it was found to produce chondroitinases and We tried to purify the chondroitinase. *B. stercoris* HJ-15 was cultured in 10 L of tryptic soy broth containing chondroitin sulfate A, collected and sonicated, followed by centrifugation at 18,000 rpm for 60 min. at 4℃. The supernatant was applied into QAE-cellulose, CM-Sephadex C-5, hydroxyapatite, phosphocelluose, and Sephacryl S-300 coumn chromatography. Two kinds of chondroitinases, chondroitinase ABC and chondroitinase AC were purified. They were consisted of monomer and dimer subunit, respectively. The specific activities, molecular weight and optimal pH of chondroitinase ABC were 45.7 μmole/min/mg, 114 KDa and pH 7.0. The specific activities, molecular weight and optimal pH of chondroitinase AC were 57.0 μmole/min/mg, 84 KDa and pH 5.8. They were inhibited by Ni^{+2} , Mg^{+2}, Zn^{+2}, Cu^{+2} and Co^{+2}, and Cu^{+2}, Pb^{+2}, Zn^{+2}, Ni^{+2} and Co^{+2}, respectively. These findings suggest that the biochemical properties of the purified enzymes were different from the previously purified enzymes.

[PC2-5] [04/21/2000 (Fri) 14:50 - 15:50 / [1st Fl, Bldg 3]]

Biochemical characterization of HrcA in Sreptococcus pneumoniae.

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Heat shock response plays a central role in cellular adaptation to stresses and hostile environments. Of several heat shock preteins(hsps), DnaK and GroEL play a key role in the folding of nascent protein chains and in the refolding of proteins after thermal damage. HrcA acts as a repressor of groEL and dnaK operon in Streptococcus pneumoniae or other gram positive organisms.
To elucidate the biochemical nature of the HrcA, the HrcA was overproduced in E.coli and purified. we determined stability of HrcA in various conditions of PH and temperatures. Also biophysical nature of HrcA in native condition was determined by nondenaturing PAGE analysis. HrcA was found to form dimer in the absence of CIRCE and PI value and molecular weight were 5.05 and 39.3 kDa, respectively. To identify the binding of HrcA to CIRCE element, the band shift assay was performed. To determine the effect of metal ion on binding of HrcA to CIRCE, several metal ions were supplemented to band shift assay buffer.
HrcA bound to CIRCE element, especially hairpin-loop structure, which is the promoter site of groESL and dnaK operon. The binding between HrcA and CIRCE element was stimulated by supplementation of calcium and zinc. Also supplementation of calcium affected synthesis of GroEL after heat shock.

[PC2-6] [04/21/2000 (Fri) 14:50 - 15:50 / [1st Fl, Bldg 3]]

Molecular Characterization of HSP104 in Steptococcus pneumoniae

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Several stress conditions induced the synthesis of heat shock protein(HSP)s in S. pneumoniae. One