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In galactose metabolic pathway : there are three inborn metabolic disorders galactokinase deficiency (galactosemia type II), galactose-1-phosphate uridyl transferase(GALT) deficiency (galactosemia type I), uridine diphosphate galactose-4-epimerase deficiency (galactosemia type III). Among these disorders GALT deficiency is the most severe and common. Infants with GALT deficiency fail to metabolize galactose-1-phosphate. As a consequence, galactose-1-phosphate and galactose are accumulated in blood in which GALT enzyme plays the role of a pathognomonic marker. In the previous paper, we reported a reversed-phase HPLC method using 8-Amino-2-naphthalenesulfonic acid as derivatization reagent for the determination of galactosemia. But, this method has the defects such as a relatively longer pretreatment, the reduction of sensitivity. We developed an advanced diagnostic method for galactosemia by shortening pretreatment and increasing the sensitivity.

[PD4-8] [ 2003-10-10 14:00 - 17:30 / Grand Ballroom Pre-function ]

### **Indirect chiral separation of $\alpha$ -arylmethylpropionic acids by liquid chromatography**

**Min ChungSik**<sup>o</sup>, Jang SeungJae, Choi BoKyung, Kim YoungLim, Jung HaeYun, Bak KyungMin, Lee KyungHee, Jo KeangIn, Gu YouNi

*KFDA*

A various  $\alpha$ -arylmethylpropionic acids(profen) have been widely used as non-steroidal anti-inflammatory drugs for the relief of acute and chronic rheumatoid arthritis and osteoarthritis, as well as for other connective tissue disorders and pains. Example is fenoprofen, ibuprofen, ketoprofen, and naproxen. All are chiral and, except for naproxen and ibuprofen, are marketed in racemic form. Enantioseparations of profens have been of considerable interest because their anti-inflammatory and analgesic effects have been attributed almost exclusively to their (S)-enantiomer. A simple method for determination of optical purity of (+) and (-)- $\alpha$ -arylmethylpropionic acids has been developed. By means of EEDQ,  $\alpha$ -arylmethylpropionic acids was coupled to (S)-naphthylethylamide. The diastereoisomeric derivatives was then separated by normal-phase liquid chromatography. And separation process of diastereoisomeric isomer was interpreted by molecular mechanics and quantum mechanics calculation of diastereoisomeric conformation.

[PD4-9] [ 2003-10-10 14:00 - 17:30 / Grand Ballroom Pre-function ]

### **Studies on the tyrosinase inhibitory compound of *Potentilla bifurca* L. var. *glabrata* Lehm**

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Tyrosinase is an important enzyme involved in the transition steps from tyrosine to melanin. Inhibition of the tyrosinase activity could block melanin formation from tyrosine and thus prevent melanin pigmentation on skin. This may contribute to the development of new whitening agent that would be useful in the prevention of pigmentation. In this study, we isolated tyrosinase inhibitory compound from BuOH fraction of *Potentilla bifurca* L. var. *glabrata* Lehm by activity guided fractionation method. Based on spectroscopic data, the active compound was identified as a quercetin 4"-O-glucopyranoside.

[PD4-10] [ 2003-10-10 14:00 - 17:30 / Grand Ballroom Pre-function ]

### **Physical properties and determination of eupatilin, a new antigastric agent, by high performance liquid chromatography**

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Eupatilin is a major active component of *Artemisia Herba Extract* having a potent antigastric effect. We investigated the physical properties of eupatilin using high performance liquid chromatography. Solubility, stability & partition coefficient of eupatilin were investigated. pH-stability of eupatilin was examined over the broad range through pH1-9 at 37°C & it has good stability above the broad range pH. The solubility of eupatilin was extremely low but the value of logP was more than 2. Also, a high performance liquid chromatographic method was developed for the determination of eupatilin in rat plasma. The method involved deproteinization of biological sample with the same volume of acetonitrile, 0.2M zinc sulphate, and 0.15M barium hydroxide. The mobile phase employed was ammonium acetate buffer(1% ammonium acetate and 0.5% acetic acid) – acetonitrile (58:42,v/v) and the flow rate was 1.0 ml/min. The quantitation limit of eupatilin in rat plasma was 10 ng/ml. No interferences from endogenous substances were found.

[PD4-11] [ 2003-10-10 14:00 - 17:30 / Grand Ballroom Pre-function ]

### **Cyclooxygenase Inhibitory Activity of Ginsenosides from *Panax ginseng***

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*P. ginseng* C.A. Meyer is one of the most widely used herbal medicine in Asia. It has been used for the treatment of many disorders. Its major constituent is known to be ginsenosides, and there are many documents about bioactivities of ginsenosides such as anti-oxidant, anti-tumorigenic, anti-fatigue, and anti-inflammatory activities. Some of these activities are supposed to have some correlation with inhibitory action of cyclooxygenase (COX). Ginsenosides from *P. ginseng* and sapogenins were evaluated for their inhibitory effects against both cyclooxygenase-1 and -2 (COX-1 and -2). Inhibitory activity was evaluated by measuring prostaglandin E<sub>2</sub> (PGE<sub>2</sub>) production from arachidonic acid with an ELISA reader. As a results, Rg<sub>3</sub>(S), and Rg<sub>5</sub> and Rk<sub>1</sub> showed COX-2 inhibitory activity in a selective manner (COX-1: IC<sub>50</sub> = >100, 77.01 µg/mL, COX-2: IC<sub>50</sub> = 35.47, 18.6 µg/mL). Protopanaxatriol (PPT) showed moderate activity on COX-1 and -2 (COX-1: IC<sub>50</sub> = 39.16, COX-2: IC<sub>50</sub> = 35.56 µg/mL), while Re, Rg<sub>3</sub> (R), and protopanaxadiol (PPD) showed little activity.

[PD4-12] [ 2003-10-10 14:00 - 17:30 / Grand Ballroom Pre-function ]

### **HPLC Analysis of Phytosphingosine and Its Metabolites in Mammalian Cells with TCPO- H<sub>2</sub>O<sub>2</sub> Chemiluminescence Reaction**

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Sphingolipids has been known to induce apoptosis, cell proliferation, differentiation and migration in a variety of cell types . Recently, its phosphate form was suggested that they may act both as an agonist ligand to S1PRs and a second messenger in intracellular action. Phytosphingosine(PHS) is not easily detected due to trace component of cellular lipids in mammalian and human tissues while this is a major sphingolipid in yeast and plants. We therefore developed highly sensitive and reproducible analytical method for PHS and its phosphate by oxalic acid bis(2,4,6-tri-chlorophenyl) ester(TCPO)-hydrogen peroxide(H<sub>2</sub>O<sub>2</sub>) chemiluminescence. The NDA derivatives of PHS exhibited stable fluorescences and was enhanced their detectability at low concentrations by post-column chemiluminescence detection with TCPO- H<sub>2</sub>O<sub>2</sub> . The dried lipid extracts or sphingoid base standards for the calibration curve were dissolved in 40 µl of ethanol. NDA derivatization was accomplished by adding the following stock solutions: 40 µl 0.05M NaHCO<sub>3</sub> / 0.1M NaOH buffer(pH 10.5), 20 µl 13% (w/v) NaCN, and 20 µl 0.5%(w/v) NDA. The tube was tightly sealed with PTFE film and heated at 67°C in a water bath for 90 min, glycine was added to stop the derivatization reaction. We successfully measured the amount of PHS and PHS-1-P in LLC-PK<sub>1</sub> cells. Collectively, this method can be thus used to detect and distinguish PHS and PHS-1-P with high sensitivity from other sphingolipids in mammalian cells.

[PD4-13] [ 2003-10-10 14:00 - 17:30 / Grand Ballroom Pre-function ]