

Mn-SOD were unchanged. This indicated that elevated oxidative stress caused by an imbalance between the production and removal of ROS and free radicals occurred in cells.

[PA3-18] [10/18/2001 (Thr) 14:00 - 17:00 / Hall D]

Determination of PCBs in Korean Adipose tissues and Endocrine Disrupting Effects

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Polychlorinated biphenyls(PCBs) are members of the halogenated aromatic group of environmental pollutants. Because of their unique physical and chemical properties, notably their stability and widespread use, PCBs are sidely distributed and transported throughout the global environment. In fact, residues of PCBs have been identified in air, water, aquatic and marine sediments, and human tissue samples. Although the mechanism of the effects of these PCBs on estrogenic function are still not entirely understood, the toxicities of the PCBs have been studied intensively. Some PCBs exert dioxin-like activities mediated through the aryl hydrocarbon receptor and some congeners are hypothesized to possess endocrine disruptive potential and to induce CYP1A. We examined antiestrogenic potentials of some PCB congeners(PCB 52, 118, 138, 153, 180)in vitro which detected in Korean adipose tissues. As a result, PCB 118, 153 inhibited aromatase activities using tritiated water release assay in JEG-3 cell line. PCB 118, 138, 153 induced CYP1A activities using ethoxyresorufin o-deethylase bioassay in H4IIE cell line. And PCB 118, 138, 153, 180 showed antiestrogenic activities by E-Screen assay in MCF-BUS cell line. This study demonstrated that PCB congeners could have and antiestrogenic activities and affect estrogen biosynthesis depend on their structure.

[PA3-19] [10/18/2001 (Thr) 14:00 - 17:00 / Hall D]

Solvent Microextraction of Methamphetamine and its metabolite, amphetmaine, in urine with simultaneous back-extraction

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Solvent microextraction was developed as a sample preconcentration for the determination of methamphetamine and its metabolite, amphetamine, in urine. Although liquid-liquid extraction(LLE) and solid-phase extraction(SPE) have been used most commonly for the preconcentration and cleanup of samples prior to the GC or GC/Mass analysis, their relative poor selectivity to the target drugs, analogues, or impurities produced poor resolution between the target drugs and impurities and resulted the low detection limit in GC or GC/Mass analysis. Furthermore the use of the relatively large amount of extracting organic solvent for the extraction of the target drugs from urine in LLE might be harmful to thamphetamine testers. Solvent microextraction employs a microliter size liquid membrane and receiving phase. The small apparatus of the solvent microextraction was composed of 2.0 mL reaction vessel, stirrer, Teflon ring, 1.0 mL syringe, stirrer bar, and Teflon stopper. The n-octane liquid membrane was confined inside a small Teflon ring and layered over 1.0 mL urine or aqueous sample which was already adjusted to alkali with 6N-NaOH. The receiving droplet of 0.05M-NaH₂PO₄(pH = 2.3) was suspended in the n-octane liquid membrane from tip of a microsyringe needle. When the sample was stirred the basic drugs like methamphetamine and amphetamine in urine diffused into the n-octanof phase because the basic drugs were in the form of molecular state, not ionized state, under alkaline conditions. Successively the molecular basic drugs in organic phase diffused into the acidic aqueous microdroplet suspended on the needle because the basic drugs turned into the ionized form in the acidic aqueous media. After extraction of the basic drugs in the aqueous media for ten minutes, the microdrop was taken back into the syringe and transferred into the 2.0 mL reaction vial. The microsyringe was rinsed out with ethanol 2 times then added in the above solution. The solution was evaporated under nitrogen stream. The residue was derivatized with trifluoroacetic anhydride and then injected into the GC/MS. The MS spectra by microextraction and by common LLE were compared each

other. The MS spectra by LLE included many other peaks due to impurities and analogues (some metabolites) as well as the targeted drugs. However, the MS spectra by microextraction included only the target drugs and the other peaks were almost disappeared. The resolution and selectivity of the targeted drugs on MS-spectra were much improved by the solvent microextraction. Those improvement in resolution and detection limit of the target drugs in microextraction were due to its simultaneous back extraction. Furthermore the recovery by solvent microextraction were almost the same as that by LLE. The detection limit of MS by the solvent microextraction was ng unit, which was 10 times more sensitive than that by LLE when the detection limit was defined with 3 times of the background signals.

Poster Presentations – Field A4. Toxicology

[PA4-1] [10/18/2001 (Thr) 14:00 – 17:00 / Hall D]

Establishment of bioassay to detect estrogenic flavonoids using stable MCF-7-ERE cell and MCF-7 cell proliferation assay.

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Stable MCF-7-ERE cells, in which pERE-Luc reporter gene has been stably integrated into the genome of the MCF-7 cells, were used to detect the estrogenic activity of various dietary flavonoids in either pure chemical or mixtures. Estradiol (E2) induced luciferase activity in dose dependent manner and this activity was inhibited by tamoxifen (Tam) concomitant treatment. A large series of flavonoids showed estrogenic activities, corresponding to EC50 values between 0.2 and 9 microM and their mixtures didn't show additive or synergistic effects. And we could find some structure and activity relationship. First, 4-methoxylation and catechol structure decreased estrogenic activities. Second, hydroxylation of 3 position reduced estrogenic effect. Third, glycosides of flavonoids showed weak estrogenic activity or no activity. Interestingly, when tested at high concentrations, genistein, kaempferol, biochanin A and chrysin elicited luciferase induction higher than that of the maximum induction by estradiol. And these effects of genistein and kaempferol could not be fully inhibited with tamoxifen. The estrogenic activity of the dietary flavonoids was further investigated using MCF-7 cell proliferation assay. In this system, several flavonoids were capable of mimicking natural estrogens and thereby induced cell proliferation. Among the investigated flavonoids, 7 compounds elicited the significant cell proliferation, whereas remaining flavonoids were weak estrogenic or devoid of estrogenic activity.

[PA4-2] [10/18/2001 (Thr) 14:00 – 17:00 / Hall D]

Study of resveratrol and its derivatives on the regulation of gene expression in MCF-7 cells transfected with either pERE-Luc or pCYP1A1-Luc.

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Resveratrol (trans-3,4',5-trihydroxystilbene), which is a polyphenolic compound found in a variety of plants such as grapes and wine, has been reported to have a variety of anti-inflammatory, anti-platelet, and anti-carcinogenic effects. Recently resveratrol was reported to serve as an estrogen agonist in MCF-7 cells. Based on its structural similarity to diethylstilbestrol, a synthetic estrogen, we examined whether resveratrol and its derivatives might be estrogenic using stable MCF-7-ERE cells. Resveratrol functioned as a superagonist at high concentrations (i.e., produced a greater maximal transcriptional