and cell proliferation of bisphenol A in the presence of a rat liver S9 mix containing cytochrome P 450 enzymes and Cu(II). In the present study, we found that BPA in combination with Cu(II) exhibited a enhancement in cytotoxicity, which was inhibited by reacitve oxygen species scavenger. For cell proliferation assay MCF-7 cells were seeded on a 96-well multi-well-plate at 1.5 x 10(3) cells per well. After 24hr cultivation, the S9 mix and Cu(II) was added to the wells as an S9 mix group (+S9), and medium was added to the other wells as a none-S9 mix group (-S9), then 5 different concentrations of various BPA were added to each well. After 5 days, a sulforhodamine B (SRB) assay was conducted to measure cell proliferation. +S9 mix group enhanced the proliferation of MCF-7 cells at much lower concentrations than -S9 mix group which was inhibited by the ROS scavenger. These results suggest that reacitve oxygen species reacts with Cu(I) leading oxidative stress. Also the formation of reacitve oxygen species induced by BPA was dose-dependently by inhibited by tamoxifen, which suggests that the effect of BPA was estrogenic action via estrogen receptors.

[PA3-16] [10/18/2001 (Thr) 14:00 - 17:00 / Hall D]

Toxicity Identification Evaluation of Water Pollution using in vitro bioassay

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So far, investigation of environmental pollution has been achieved in field study. This remains the most exhaustive approach, current dimensions of environmental researches and their inherent complexity require that relatively inexpensive and simple laboratory procedures are developed to make possible the screening of large numbers of sites and samples. At this point, micro-bioassay has been highlighted. The purpose of this study is to evaluate the water pollution using micro-bioassay. Micro-bioassay methods were optimized and validated for the sensitive and quantitative determination of total toxic effects of the water samples. EROD bioassay was focused to detect PAHs, PCBs and dioxinlike components in the water. The EROD bioassay was executed in rat hepatoma cell line, H4 II E cell lines. 50L of river water was adsorbed using XAD-2 resin column. Pollutants adsorbed to the XAD-2 resin were extracted by elution with methanol (sample I), and with ethyl acetate (sample II). Toxic effects of extracts were determined by micro-bioassay methods.

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

Oxidative damage by bisphenol A induced lipid peroxidation and apoptosis

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It is very important to understand the ROS generation of estrogen-like chemicals. Of such chemicals, we were especially interested in bisphenol A for its wide spreadness in environmental and structual similiarity with aneuploidogenic stilbene estrogen diethylstilbestrol. The purpose of the this study was to evaluate the lipid peroxidation and DNA fragmentation by bisphenol A in the presence of a rat liver S9 mix containing cytochrome P 450 enzymes and Cu(II) in HaCaT cell lines. The specific content of malondialdehyde, an end product of lipid peroxidation, was also found to increase with concentration. The fragmentation of intact DNA, a parameter of apoptotic cell death, was evaulated qualitatively by agarose gel electrophoresis analysis and quantitatively by diphenylamine reation method. BPA induced apoptotic cell death in a dose-dependent manner. When HaCaT cells were exposed to 50uM BPA for 48h, the DNA fragmentaion was significantly increased to 54%. The effect of radical scavenger on the apoptotic cell death induced by BPA was investigated. The DNA fragmentation induced by BPA was significantly inhibited by addition of ROS scavenger to the culture medium. Also we examined the enzyme activities of Cu,Zn-SOD, Mn-SOD, catalase, and GPx in the cells. The activities of Cu,Zn-SOD, glutathione peroxidase, Catalase were found to decrease with concentration. However, the activity of