present study, we investigated the effects of capsaicin on pulmonary metastasis of B16-F10 murine melanoma cells, as well as its mechanism of action. Capsaicin (i.p., 2.5mg/kg) suppressed the number of lung colonies (58%) in experimental lung metastasis assay. We studied the effects of capsaicin on B16-F10 melanoma cells growth, apoptosis and expression of VEGF and iNOS using western blot and immunohistochemistry. We found that capsaicin (i.p., 1.25, 2.5 mg/kg) inhibited the expression of iNOS and VEGF in the tumor lesions. DNA fragmentation, Caspase-3 activation and cleavage of PARP were observed after treatment with capsaicin dose- and time- dependent manner. We also observed in situ DNA fragmentation in the tumor lesions using the TUNEL method in animal model. TUNEL-positive cells were rarely found in tumor lesions of control mice, whereas many positive cells with marked fragmented nuclei were present in the tumor lesions of capsaicin treated mice(i.p., 0.625 ~ 2.5 mg/kg). Also, downregulation of bcl-2 expression was observed in capsaicin treated cells, but there was no difference in the expression of bax and p53. Taken together these results, capsaicin may prevent pulmonary metastasis of B16-F10 melanoma cells through apoptosis by decreasing the bcl-2 expression and increasing of caspase-3 activity and suppression of VEGF and iNOS.

[PA4-28] [04/17/2003 (Thr) 14:00 - 17:00 / Hall P]

Effect of skin and seed of Grape and on Dimethylnitrosamine-Induced Liver Damage in Rats

Shin MiOk°, Shin JiYoung, Yoon Sik, Moon JeonOk

College of Pharmacy, Pusan National University, Kumjeong-gu, Pusan 609-735, Korea and Department of Anatomy, College of Medicine, Pusan National University, Seo-Gu, Pusan 602-739, Korea

Polyphenolic compounds have been reported to exhibit a wide range of pharmacological properties. In this study, we investigated the hepatoprotective effect of skin and seed of grape which contain abundant polyphenol compounds on dimethylnitrosamine(DMN)-induced liver damage in rats. Ingestion of skin and seed of grape (10% diet, daily for 4 weeks) into the DMN-treated rats remarkably prevented the elevation of serum alanine transaminase, aspartate transaminase and alkaline phosphatase, and bilirubin levels. They also increased serum protein level and reduced the hepatic level of malondialdehyde in DMN-treated rats. Furthermore, DMN-induced elevation of hydroxyproline content was reduced by the ingestion of grape seed and skin which result was consistent with a histochemical analysis of liver tissue stained with Sirius red. In conclusion, these results demonstrated that the in vivo hepatoprotective effect of grape against DMN-induced liver injury, and suggest that grape may be useful in the prevention of liver damage

[PA4-29] [04/17/2003 (Thr) 14:00 - 17:00 / Hall P]

Single and 28-day Repeated Dose Toxicity Studies of Botulinum Toxin Type A in Mice and Rats

Jeon TaeWon°, Hyun SunHee, Kim NamHee, Lee SangKyu, Kim ChunHwa, Lee DongWook, Woo HeeDong*, Yang GiHyeok*, Jung HyunHo**, Jeong TaeCheon

College of Pharmacy, Yeungnam University, Kyungsan, 712-749, Korea, *Microbial Toxin Research Institute, Medy-Tox Inc., Cheonan, 336-708, Korea, **Division of Applied Biological Sciences, Sunmoon University, Cheonan, 336-708, Korea

Single and 28-day repeated dose toxicity studies of botulinum toxin type A(BTA) were carried out in ICR mice and SD rats, respectively. In the single dose toxicity study, BTA was injected intraperitoneally to male and female mice at a single dose of 40, 59, 89, 133 and 200 ng/kg. All animals died from 59 ng/kg. Some clinical signs were observed in most of both sexes from 59 ng/kg, but no signs were seen in all animals at 40 ng/kg. The results showed that the LD_{50} of BTA might be in the range of 40-59 ng/kg in both sexes. In the repeated dose toxicity study, the test material was administered intradermally for 28 days at doses of 0(control), 1.25, 2.5, 5.0 and 10.0 ng/head/50 $\mu\ell$ saline in male and female rats. BTA treatment significantly decreased the body weight gain rate in male of 5.0 ng/head and over and in female of 10.0 ng/head compared to control. One or more relative organ weights were increased significantly from 5.0 ng/head compared to control in both sexes. Serum biochemistry revealed increases in AST, ALT, CPK, total protein and albumin in male, and increases in AST and ALT and decreases in K^+ and Cl^- in female without dose-dependent manners. In the histopathological study, BTA treatment induced atrophy of skeletal muscle in both sexes from 2.5 ng/head. When the antibodies to toxin were determined in all animals, a significant increase in serum antibodies was observed from 5.0 ng/head. The results showed that the NOAEL of BTA might be 1.25 ng/head for 28-day repeated dose toxicity in rats. (Supported by a sub-grant from Ministry of Commerce, Industry and Energy for Medy-Tox Inc.).

[PA4-30] [04/17/2003 (Thr) 14:00 - 17:00 / Hall P]

Cadmium induces neurotoxicity via activation of JNK and c-JUN in human neuroblastoma cell

Kim Sun Don°, Moon CK, Jo Sangmee Ahn

Division of Brain Disease, Department of Biomedical Science, National Institute of Healt, and College of Pharmacy, Seoul National University, Seoul [122-701], Korea

Occupational exposure to cadmium (Cd) can result in brain disorders and olfactory dysfunction is the most well-known symptom. Recently Cd has been shown to induce apoptosis by activating MAPKs in various cell types. However, intracellular signaling pathways of Cd-induced cytotoxicity in neuronal cells is not known well. Thus, in the present study, we studied role of JNK and its well-known downstream transcription factor, c-JUN, in Cd-induced neuronal cell death. Treatment of SH-SY5Y cell, a human neuroblastoma cell line, with Cd caused cytotoxicity in a concentration- and time-dependent manner as measured by the MTT assay; LD50 was approximately 25 uM after 24h incubation with Cd. Cd-induced cytotoxicity involved apoptosis as determined by TUNEL staining. Western blot analysis showed that JNK was phosphorylated by Cd (1- 250 uM); its activation was seen as early as 1h after Cd exposure and thereafter sustained until 12h. In contrast, p38 was phosphorylated only at the high dose (100 uM) of Cd while Erk was not activated at all. In addition, c-JUN was also phosphorylated by Cd. Stable expression of dominant negative mutant construct of MKK4 reduced the Cd-induced cytotoxicity as well as c-JUN phosphorylation, suggesting that phosphorylation of JNK/c-JUN is responsible for cytotoxic effects mediated by Cd. Moreover, treatment of cells with N-acetyl-L-cysteine (1 mM), a free radical scavenger, increased cell viability significantly. Taken together, oxidative stress induced in cells by Cd may cause cytotoxicity, and phosphorylation of JNK and c-JUN is involved in cadmium-induced cytotoxicity in neuronal cells.
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[PA4-31] [04/17/2003 (Thr) 14:00 - 17:00 / Hall P]

PAH regualtion of CYP1 gene in MCF-7 & ZR-75-1 human breast cancer cells

Min KyungNan, Kim JaYoung, Kang SoHee, Seo MiJung, Sheen YhunYhong