dependent manner. In addition, DA-125 inhibited cancer cell migration and colony formation, and also exhibited the inhibitory activities of invasion and motility with a matrigel and type I collagen assay. These results suggest that DA-125 inhibits tumor cell invasion and metastasis by suppression of MMPs and TIMPs in tumor cells. Further, cell adhesion molecules (CAMs) such as vascular cell adhesion molecule-1 (VCAM-1) and intercellular cell adhesion molecule (ICAM) have been reported to play an important role in cancer metastasis via the adhesive interaction between tumor cells and endothelial cells. We examined the effects of DA-125 on CAMs expression and its transcriptional regulatory mechanism in human microvascular endothelial cells (HMEC-1). Dose-dependent suppression of CAMs mRNA levels was observed in DA-125-treated HMEC–1. Taken together, anti-metastatic and anti-invasive effect of DA-125 might be an additional mechanism as a promising anticancer agent.

[OA4-1] [2003-10-11 09:45 - 10:00 / ASEM Hall Meeting Room 208]

Dihydrosphingosine 1-phosphate: New Biomarker for Fumonisin B1 Toxicity

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Fumonisins are a family of mycotoxins produced from Fusarium verticillioides. Most of fumonisin B1 (FB1) toxicities can be explained by its ability to alter sphingolipid metabolism by inhibiting ceramide synthase. At least, the elevation in dihydrosphingosine (DHS) mediates the earliest toxicity of FB1. Some tissues such as kidney and liver, may be most affected by FB1 because they shows high rates of de novo sphingolipid synthesis. Recent review on FB1 toxicity by A.H. Merrill Jr. et al. suggested the possible role of dihydrosphingosine 1-phosphate (dihydroS1P), which sometimes elevated in cell- or tissue specific manners. In this study, we demonstrated that FB1 accumulated not only DHS but dihydroS1P, which was typically observed in FB1-sensitive pig kidney epithelial cells (LLC-PK1 cells). Moreover, dihydroS1P was suggested as a new indicator for FB1 exposure in rat plasma while sphingoid bases ratio was still useful in other organ tissues. For further study to elucidate the toxic mediator of FB1, we used mouse F9 embryonal carcinoma cells, which exhibits SPL -/- stable transformant (SPL lyase KO) and murine S1P phosphohydrolyase (mSPP1) stably overexpressed transformant (SPL lyase KO + mSPP1). Surprisingly, overexpression of mSPP1 in SPL -/- stable transformant showed strong resistance to FB1. Conclusively, the FB1 toxicity may be mainly mediated by endogenous dihydroS1P, which possibly exerted antagonistic action to S1P in intracellular mode.

[OB3-1] [2003-10-11 10:00 - 10:15 / ASEM Hall Meeting Room 208]

In vivo evidence for brain-to-blood efflux transport of taurine and regulation of this transport by tumor necrosis factor-α at the blood-brain barrier

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The purpose of this study is to examine whether the efflux system for taurine from brain to blood is present on the blood-brain barrier (BBB) using the brain efflux index (BEI) method and taurine transport system is regulated by CNS cell damage with oxidative stress agent such as diethyl maleate (DEM) or tumor necrosis factor-α (TNF-α) in vivo. [^{3}H]Taurine was microinjected into parietal cortex area 2 (Par2) of the rat brain, and was eliminated from the brain with efflux transport rate of 1.22 10^{-2}/min, and the process is saturable with a K_m of 43.5 μM. This process was significantly inhibited by taurine transport inhibitors, such as unlabeled taurine, β-alanine, betaine, nipecotic acid and γ-aminobutyric acid (GABA). In addition, the effect of DEM or TNF-α on [^{3}H]taurine transport was investigated. [^{3}H]Taurine uptake was increased and efflux was reduced by pre-treatment with DEM or TNF-α. Also, [^{3}H]taurine efflux was decreased by TNF-α in time- and dose-dependent manner. In conclusion, the efflux pump for taurine at the BBB reduced taurine concentration in the brain interstitial fluid and this process was carrier mediated and also, was regulated by oxidative cell damage.

[OC1-1] [2003-10-11 10:15 - 10:30 / ASEM Hall Meeting Room 208]

Augmentation of constitutive nf-κb activation by bcl-2 in pc12 cells: implications for

protection against oxidative stress

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A substantial body of evidence indicates that reactive oxygen intermediates (ROIs) are implicated in pathogenesis of diverse human diseases, including cancer, diabetes and neurodegenerative disorders. Oxidative stress induced by ROIs often causes cell death via apoptosis that is regulated by a plenty of functional genes and their protein products. In the present work, we have investigated the role of bcl-2 in protecting against oxidative death induced by hydrogen peroxide in cultured rat pheochromocytoma (PC12) cells. When PC12 cells were treated with hydrogen peroxide, they underwent apoptotic death as determined by characteristic morphological features and internucleosomal DNA fragmentation. Hydrogen peroxide treatment also led to the decreased mitochondrial membrane potential, increased Bax expression, activation of caspase-3, and cleavage of poly(ADP-ribose)polymerase. Transfection with the anti-apoptotic bcl-2 rescued PC12 cells from oxidative cell death caused by hydrogen peroxide. Addition of NF-κB inhibitors, pyrrolidine dithiocarbamate or L-tosylamido-2-penetyl chloromethylketone to the media aggravated hydrogen peroxide-induced PC12 cell death. PC12 cells overexpressing bcl-2 exhibited relatively high constitutive DNA binding and transcriptional activities of NF-κB, compared with the vector-transfected control cells. In addition, sustained NF-κB activation was observed in the bcl-2 overexpressing cells, which was accompanied by the constitutive activation of extracellular signal-regulated kinase 1/2. However, bcl-2 overexpression did not cause any significant alterations in the activity of either c-Jun N-terminal kinase and p38 mitogen-activated protein kinase. The ectopic expression of bcl-2 caused elevated level of cellular glutathione and expression of γ-glutamate-cysteine ligase and catalase, which were inhibited by NF-κB inhibitors. These results suggest that NF-κB plays a role in bcl-2-mediated protection against hydrogen peroxide-induced apoptosis in PC12 cells through augmentation of antioxidant capacity.

[OC1-2] [2003-10-11 10:30 - 10:45 / ASEM Hall Meeting Room 208]

Celecoxib inhibits phorbol ester-induced expression of cyclooxygenase-2 and skin-tumor promotion in mouse skin: p38 and AP-1 as possible molecular targets

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Celecoxib, the selective cyclooxygenase-2 (COX-2) inhibitor, has recently been reported to reduce the formation of polyps in patients with familial adenomatous polyposis. This specific COX-2 inhibitor also protects against experimentally induced carcinogenesis, but molecular mechanisms underlying its chemopreventive activities remain largely unresolved. In the present work, we found that celecoxib inhibited 12-O-tetradecanoylphorbol-13-acetate (TPA)-induced expression of COX-2 in female ICR mouse skin when applied topically 30 min prior to TPA as determined by both immunoblot and immunohistochemical analyses. In another study, celecoxib attenuated the DNA binding activity of activator protein-1 (AP-1) through suppression of c-Jun and c-Fos expression in TPA-treated mouse skin. Under the same experimental conditions, celecoxib inhibited both the catalytic activity and phosphorylation of p38 mitogen-activated protein (MAP) kinase in mouse skin. While the p38 inhibitor SB203580 blocked TPA-mediated AP-1 activation and c-Jun expression, the MEK1/2 inhibitor U0126 was not inhibitory despite suppression of c-Fos expression in mouse skin. Furthermore, SB203580 markedly blunted COX-2 expression induced by TPA. An anti-tumor promoting activity of celecoxib was examined in a two-stage mouse skin carcinogenesis model with 7,12-dimethylbenz[a]antharcene as an initiator and TPA as a promoter. Topical application of celecoxib (10 mmol) prior to each TPA treatment reduced the multiplicity of papillomas by 32% at 18 week. All the tumors analyzed exhibited elevated COX-2 levels compared with surrounding normal tissues or vehicle-treated control skin. The COX-2 expression was found to be repressed in the papillomas from celecoxib-pretreated mice. Taken together, down-regulation of COX-2 by blocking activation of p38 MAP kinase and AP-1 may account for the anti-tumor promoting activity which celecoxib exerts in mouse skin tumorigenesis.

[OC2-1] [2003-10-11 10:45 - 11:00 / ASEM Hall Meeting Room 208]

Characterization of Muations in DNA Gyrase and Topoisomerase IV Involved in