Department of Environmental and Health Chemistry, College of Pharmacy, Chung-Ang Univ., Seoul, Korea

It become evident that Ca^{2+}-dependent release of arachidonic acid (AA) and subsequent formation of bioactive lipid mediators such as prostaglandins and leukotrienes in red blood cells (RBCs) can mofify physiological functions of neighboring RBCs and platelets, and thus influence hemostssis and thrombosis. Here we identified a novel type of cytosolic PLA_2 in bovine and human RBCs and purified it to apparent homogeneity with a 14,000-fold purification. The purified enzyme, termed $rPLA_2$, had a molecular mass of 42 kDa and revealed biochemical properties similar to group IV $cPLA_2$, but showed different profiles from $cPLA_2$ in several column chromatographies . Moreover, $rPLA_2$ did not react with any of anti-$cPLA_2$ and $sPLA_2$ antibodies and examined by matrix-assisted laser desorption/ionization time-of-flight mass spectrometric analysis. Divalent metal ions tested exhibited similar effects between $rPLA_2$ and $cPLA_2$, whereas mercurials inhibited $cPLA_2$ but had no effect on $rPLA_2$. Antibody against the 42 kDa protein not only precipitated the $rPLA_2$ activity, but also reacted with the 42 kDa protein from bovine and human RBC in immunoblot analysis. The 42 kDa protein band was selectively detected in murine fetal liver cells as a type of progenitor cells of RBCs. Finally, we found that EA4, a derivative of quinone newly developed as an inhibitor for $rPLA_2$ in vitro, inhibited a Ca^{2+} ionophore- and mechanical stress-induced AA release from bovine and human RBCs. Furthermore, Ca^{2+} ionophore- and exogenous AA-induced AA release inhibited by PD 98059, MAP kinase inhibitor. Our results altogether demonstrate that the 42 kDa $rPLA_2$ is identified as a novel form of Ca^{2+}-dependent PLA_2 and responsible for the Ca^{2+}-dependent AA release from human and bovine RBCs and modulated by MAP kinase at least in part.

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

Protective effects of paeonol on cultured rat hepatocytes exposed to Br-A23187

Yi JungHeon°, Bae KiHwan, Oh Kiwan

College of pharmcy, Chungbuk Natl Univ. and Chungnam Natl Univ.

Br-A23187 (Ca2+ ionophore) is frequently used as a model of Ca2+-dependent cell killing as Br-A23187 induces both necrotic and apoptotic cell death. The aim of this study was to evaluate the protective effects of paeonol isolated from Paeoina Moutan on cultured rat hepatocytes exposed to Br-A2187. Cell killing was assessed propidium iodide fluorometry. Br-A23187 caused dose-dependent cell killing. Br-A23187 (10μM)-induced cell killing was decreased in the presence of paeonol (20, 50 and 100 μM). On the other hand, Br-A23187-induced [Ca+2]i level was increased dose-dependently. However, paeonol decreased Br-A23187-induced [Ca+2]i level in dose-dependent manner. Additional measurements of enzyme activities were made to know whether paeonol inhibits those enzyme activities such as LDH, S-GOT and S-GPT on cultured hepatocytes.
Therefore, the present results indicate that paeonol has protective effects against Br-A23187-induced hepatocytotoxicity in rats, indicating paeonol decreases [Ca+2]i level and inhibits enzyme activities related to hepatotoxicity.

[PA4-11] [10/18/2001 (Thr) 14:00 - 17:00 / Hall D]

The Screening of Toxicity from the Processed Aconiti Tuber in vitro and in vivo

Kim Kiyoung1, Ma Jinyeul2, Im Mikyoung3 Park Jinsea2, Kim Ho-Cheol4, Kim Jin Sook2*

Dept. of Pathology Medical school & Professional Grad. School of Oriental Med.1, Dept.Conservative