extracellular glycosylphosphatidylinositol-specific phospholipase C in porcine renal proximal tubules

PARK SW[O], PARK HS

Laboratory of Biochemistry, College of Pharmacy, Chonnam National University, Kwangju 500-757, Korea

It was previously reported that human urinary dipeptidase was the released form of renal dipeptidase (RDPase) (EC 3.4.13.19), a glycosylphosphatidylinositol (GPI)-anchored glycoprotein to apical membrane of proximal tubules. This *in vivo* release was accomplished by GPI-PLC.
In *in vitro* model system of porcine proximal tubules, RDPase was also released by not only bacterial PI-PLC but also endogenous GPI-PLC, which did not follow the intracellular PLC signaling. In this study we assayed the activity of released RDPase using a direct NO donor, sodium nitroprusside (SNP), a nitric oxide synthase (NOS) substrate, L-arginine (Arg), an NOS inhibitor, Nω-Nitro-L-arginine-methyl ester (NAME) in order to investigate the relationship between NO and GPI-PLC activity. Addition of 5 mM Arg to porcine proximal tubules showed the increase of NO production and the decrease of RDPase release simultaneously in the time- and concentration-dependent manner. Treatment with 10 mM NAME inhibited the Arg effect on NO production and RDPase release. A concentration-dependent treatment with SNP also showed the inhibition of RDPase release and 0.5 mM SNP induced an immediate and continuous decrease in GPI-PLC activity. This effect was confirmed quantitatively by western blot using polyclonal antibody raised against porcine RDPase. These results suggest that NO down-regulates the RDPase release by inhibition of GPI-PLC, but not by direct inhibition.

[PC1-16] [04/21/2000 (Fri) 14:50 - 15:50 / [1st Fl, Bldg 3]]

Age-related up-regulation of NF-kappaB and modulation by calorie restriction

Kim HJ[O], Yu BP, Chung HY

College of Pharmacy, Pusan National University, Pusan 609-735, Korea

Recent data strongly suggested that the calorie restriction (CR) may retard aging by its anti-oxidative action on the regulation of the intracellular redox status. Regulation of the redox-sensitive NF-κB during aging is important because oxidative stress increases during aging, and CR is an effective modulator against oxidative stress. In this present study, we investigated whether age affects the regulation of NF-κB, and how the age effect is modulated by CR. The kidney isolated from Fischer 344 rats at 6, 12, 18, and 24 months of age fed ad *libitum* (AL) and CR rats were used. Results show that the aging process strongly enhanced NF-κB DNA-binding activity that was in parallel with an increased generation of reactive oxygen species (ROS). Accompanied with changes in NF-κB was the decreased inhibitory IκBα protein in cytosol. At the same time, it was found that the nuclear p65 protein increased with age, affirming the increased translocation of NF-κB into nucleus. However, CR reversed the age-related activation and translocation of NF-κB. Our results further revealed that CR effectively blocked increased activation of NF-κB by suppressing the IκBα degradation. Based on these data, we concluded that the age-related increase in redox-sensitive NF-κB binding activity is associated with increased ROS, and CR modulates the NF-κB activation by suppressing oxidative stress.

[PC1-17] [04/21/2000 (Fri) 14:50 - 15:50 / [1st Fl, Bldg 3]]

A 40 kDa Ca2+-dependent Cytoslic Phospholipase A2 Is Implicated in A23187-induced Release of Arachidonic Acid from Mammalian Red Blood Cells

Shin HS[O], Chin M-R, Jung J-H[1], Ryu CK[2], and Kim DK