

Here, we reported MBL purified by using PG affinity column, and its biochemical characterization. MBL might be engaged in activation of complement-MBL mediate pathway.

[PC1-37] [04/18/2002 (Thr) 14:00 - 17:00 / Hall E]

Studies on Inhibitory Effect of Melanoma Cell Invasion by Human Placenta Histone H1

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Histone H1 was purified from human placenta and the inhibitory effect on the invasion of melanoma cell was studied. The histone subtraction H1.5 was purified from acid-extract of human placenta. It did not affect the melanoma cell proliferation, and slightly increase the cell motility on gelatin coated membrane. It was detected that histone H1 inhibited the invasion of melanoma cell on Matrigel coated membrane in dose-dependent manner. But, the inhibitory effect of histone H1 on the invasion of melanoma cells were not related to the early expression of MMP gene. These results suggest that the inhibitory action of histone H1 on the invasion of melanoma cells and will provide the possibility of development on anti-invasive agents.

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Purification of Paraoxonase 1 from Human Plasma

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Human paraoxonase 1 (PON1), an enzyme associated with high density lipoprotein, is responsible for the hydrolysis of organophosphates as well as aryl esters. Especially, the enzyme possesses the ability to protect low density lipoprotein against Cu^{+2} catalyzed oxidation. Here, PON1 is purified from human plasma by using Cibacron Blue 3GA chromatography, DEAE sephacell chromatography, Sephacryl HR-200 chromatography and Concannavalin A chromatography. Finally, FP-HPLC is utilized to remove other contaminating proteins. The purified paraoxonase, showing a specific activity of 585 $\mu\text{mole}/\text{min}/\text{mg}$ protein (195 fold) was relatively pure on SDS-PAGE analysis. The enzyme, expressing a K_m value of 0.87 mmole, was inactivated irreversibly by p-hydroxymercury benzoic acid and acrolein, indicating that cystein residue exist in the active site. In the addition, the susceptibility of PON1 to Cu^{+2} bound 'OH may support the involvement of histidine residue in active site. Based on the results, human plasma is identical to that from human serum in various physicochemical properties.

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The novel peptidoglycan detection system by using prophenoloxidase cascade reaction

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The prophenoloxidase(proPO)-activating system(proPO-AS) is an efficient non-self recognition system in invertebrates that can recognize and respond to micrograms of lipopolysaccharides(LPS) or peptidoglycans (PG) from bacteria and B-1,3-glucans from fungi. To obtain the solution showing specific phenoloxidase(PO) activity against PG, I have prepared PG-specific solution from *Galleria mellonella* larvae by using the first and the second Sephadex G-100 gel filtration

column. The second Sephadex G-100 solution was not shown PO activity by microgram order of LPS and B-1,3-glucan. Nanogram quantity of soluble PG was specifically quantified by using this G-100 solution in vitro. Also, I purified and characterized specific PG recognition proteins from G-100 solution by using Dextran sulfate CL-6B column and Butyl-Toyopearl FPLC.

[PC1-40] [04/18/2002 (Thr) 14:00 - 17:00 / Hall E]

Down-regulation mechanism of Senescence Marker Protein 30 by ROS during aging

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The senescent changes in the expression of functional proteins affect multiple deteriorative factors for various cellular activities and homeostasis. As the cause of deterioration during aging, reactive oxygen species (ROS) are well-known factors. Senescence marker protein 30 (SMP-30) plays an important role as a calcium binding protein that is known to be identical to regucalcin. The expression of SMP-30 that is preferentially exhibited in hepatocytes and renal tubular epithelia significantly declined during aging. It has been demonstrated that SMP-30 rescues cell death by enhancing plasma membrane Ca^{2+} -pumping activity. However, recently, there is no information on the SMP-30 modulation by the anti-aging action of calorie restriction (CR). To characterize the status of SMP-30, the study explored the effect of aging on SMP-30 modulation by CR. The kidney and liver were isolated from Fischer 344 rats at 6, 12, 18, and 24 months of age fed ad libitum (AL) and CR rats. Results showed that SMP-30 expression markedly decreased during aging, whereas this decreased expression was clearly blunted by CR, showing a comparable level of 6 month-old AL rats. To investigate an aspect that age-induced ROS are related with SMP-30 gene expression, it was examined whether LPS-induced ROS affect gene expression of SMP-30 and DNA binding activity for nuclear protein. These results suggest that down-regulation of SMP-30 is reconciled with both age-related ROS and experimentally LPS-induced ROS.

[PC1-41] [04/18/2002 (Thr) 14:00 - 17:00 / Hall E]

Regulation of Redox-sensitive Transcription Factors in aging process

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Oxidative stress is considered to be the major cause of aging and many age-related diseases. Calorie restriction (CR) is known to retard the aging, and age-related deleterious processes. Recent studies documented that CR retards the aging process with its anti-oxidative ability by regulating the intracellular redox balance. Among key cellular components exquisitely sensitive to the redox status are transcriptions factors such as nuclear factor kappa B (NF- κ B), activator protein-1 (AP-1), and hypoxia inducible factor-1 (HIF-1). Currently, there is a limited information available on the age-related and dietary modulation on these factors. In this review, major focus was placed on whether age affects the regulation of NF- κ B, AP-1 and HIF-1, and further to delineate how the age-related changes are modulated by CR. It is concluded that the age-related increases in redox-sensitive NF- κ B, AP-1, and HIF-1 binding activities are associated with increased ROS, and further that CR modulates their activations by suppressing oxidative stress. Data on molecular regulation provides better molecular insights into the mechanisms underlying the cellular redox maintenance, which may reveals the cross-talk between the aging and age-associated pathogenic processes.

[PC1-42] [04/18/2002 (Thr) 14:00 - 17:00 / Hall E]

Participation of Protein Disulfide Isomerase in Molecular Fate of Thyroglobulin and its Regulation by Endogenous Oxidants and Reductants