

Irrespective of the explanation, it is clear that measurement of the level of HA in biological fluids may provide a useful marker for monitoring the onset and progression of a number of important diseases and disorders.

[PC1-30] [04/18/2002 (Thr) 14:00 - 17:00 / Hall E]

MALDI-TOF MS Approach to Proteomics : Identification of the E6AP-interacting factors in SiHa cervical cancer cells

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Human papillomaviruses have been recognized as the primary cause of cervical cancer. Viral oncoproteins are selectively retained and expressed in carcinoma cells infected with human papillomavirus and cooperated in immortalization and transformation of primary keratinocytes. E6 associated protein (E6AP) is a 100kDa cellular protein which mediates the stable association of the high-risk HPV E6 protein with tumor suppressor protein p53, resulting in the degradation of p53. E6AP was known as E3 ubiquitin-protein ligase, which has been proposed to play a role in defining the substrate specificity of the ubiquitin-proteasome degradation. In order to identify the E6AP-interacting molecules, SiHa cervical carcinoma cells having HPV 16 genome, was used. We have produced his tagged E6AP and E6AP-Ni²⁺-NTA-affinity column was prepared to obtain E6AP-interacting proteins. The E6AP-interacting proteins were resolved in 2D-gel and analysed by matrix-assisted laser desorption/ionization (MALDI/TOF). Among 21 proteins identified in 2D patterns of SiHa cell lysate bound to E6AP protein, there are 2 proteins not yet identified. Desmocollin, NT2RM1000563 protein, GR AF-1 coactivator 3, SMF protein, CD2 binding protein 1, Rab interacting lysosomal protein, pigment epithelium-derived factor, A20-binding inhibitor of NF- κ B activation-2, GTP-binding protein, PACSIN3, novel protein similar to mouse thrombospondin type 1 were bound to E6AP. These results suggest that E6AP can have several functions by interacting with the cell adhesion molecules, immune-regulatory factors, cell cycle regulators and cell signaling regulating factors in SiHa cells (This work was supported from the Molecular Medicine Program, MOST).

[PC1-31] [04/18/2002 (Thr) 14:00 - 17:00 / Hall E]

2-DE/MALDI-TOF MS Analysis of Age-dependent Mitochondrial Proteome in Rat Liver

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Mitochondria is called power plant of the cell because they product biological energy, ATP, using electron transport system and proton pump. But this system generates ROS, which cause mitochondrial damage and cell apoptosis. Furthermore, proteins are damaged by this oxidative stress in aging process. That is one of the most possible factor responsible for the functional destruction in aged tissues. To study the age-dependent proteome profile of mitochondrial proteome, we used 2D-electrophoresis and MALDI-TOF MS technique, in young (6 months of age) and aged (24 months of age) rats. As a result of image analysis and MALDI-TOF/MS of 64 spots from silver-stained 2D gel, we identified 45 spots by peptide mass fingerprint. Among them, 30 spots showed age-dependent expression patterns (increase or decrease) between young and old rat liver mitochondria proteome. But in calorie-restricted old rat, these patterns were reversed. These results suggest that age-related changes of mitochondrial proteome are responsible for functional loss of organ in aging process and further study of the age-dependent interaction mechanism of these proteins can give us the key to elucidate aging process.

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