E105 Immunological analysis of ferritin in wax moth, *Galleria mellonella* L.

Beom Su Kim*, Chang Seok Lee, Chi Young Yun[1], Woo Kap Kim and Hak Ryul Kim
Dept. of Biology, Korea Univ, [1]Dept. of Biology, Daejon University

The distribution of ferritin in various organs were observed by electron microscopic immunogold labeling, using the antibody against ferritin. We showed immunological relationship of other species ferritins with *Galleria mellonella* anti-ferritin and concentration of ferritn in each stage of *G. mellonella*. The antibody from 26kDa subunit was able to recognize 26kDa but was not reactive with other subunits from western blot. The ciruclar dichroism spectra of ferritin were obtained between 195 and 280nm on a Jasco J-715 sepctropolarimeter. Carbohydrate analysis showed positive reaction with 26kDa, 30kDa, 32kDa, respectively. Fifth instar *G. mellonella* larvae were feeded with 50mM of heavy metal ion($FeCl_2$, $CdCl_2$, $CuSO_4$, $Pb(NO_3)_2$, HgO, $ZnCl_2$) dissolved in DW. After 24h, There are distinct band patterns between normal hemolymph and heavy metal ion feeded hemolymph.

E106 Pepstatin-Insensitive Carboxyl Proteinase: A Biochemical Marker for Late Lysosomes in *Amoeba proteus*

Hyun Jung Kim, Hae Kyung Kwon, Dae Hui Park, and Tae In Ahn
Department of Biology Education, Seoul National University

By using a monoclonal antibody (mAb) against late lysosomes in *Amoeba proteus* as a probe, we cloned two cDNAs, a 1.3-kb cDNA in pBSK-lys45 and a 1.6-kb cDNA in pBSK-lys60, encoding proteins homologous to pepstatin-insensitive carboxyl proteinases (PICPs). *E. coli* transformed with pBSK-lys45 produced a 49-kDa immunopositive protein and the cDNA in 1274 bases encoded a 44,733-Da protein (Lys45) of 420 amino acids containing one site for a core oligosaccharide. On the other hand, *E. coli* transformed with pBSK-lys60 produced four polypeptides (64, 54, 47, 41, and 37 kDa) reacting with the mAb. The cDNA contained 1629 bases and encoded a 59,231-Da protein (Lys60) of 530 amino acids containing two sites for asparagine-linked core oligosaccharides. These two cDNAs showed identities of 60.3% in nucleotide sequences, and 23.6% in amino acid sequences, respectively. Lys45 and Lys60 appeared to share XXEFQK as a common antigenic domain. The amino-acid sequence of the Lys45 protein showed a 17.4% identity and 40.9% similarity with that of PICP from *Pseudomonas* sp. 101. On the other hand, Lys60 showed a 24.3% identity and 51.9% similarity with human lysosomal PICP in the amino-acid sequence. A putative active center for serine protease, GTS*XXXTXEXG, was found to be all conserved among PICP homologues. The two PICPs are the first reported enzymatic markers for late lysosomes in *A. proteus*.