

P-type Ca^{2+} channels of the cell body using conventional two-electrode voltage clamp techniques over a 4-year period. The Ca^{2+} in the bath was replaced with Ba^{2+} to reduce the inhibitory effect of high extracellular Ca^{2+} on o-agatoxin IVA. The activation voltage and kinetics of the P-type Ca^{2+} channel were constant during all seasons. However, the density of P-type Ba^{2+} current was the smallest during the summer (15.6 ± 2.2 nA/nF, n=63) and the greatest during the winter (23.7 ± 3.7 nA/nF, n=29), whereas the P-type Ba^{2+} current densities were intermediate during the spring and the fall. These results indicate that there were seasonal changes in the number of functional P-type Ca^{2+} channel, not in the channel properties. These seasonal changes in the inactivation of P-type Ca^{2+} current are presumably due to seasonal differences in neuronal impulse activity since the animal activity is generally the greatest during the summer. Thus, activity-dependent inactivation of P-type Ca^{2+} channel may play a role in transmitter release from crayfish motoneuron in which inactivation of P-type Ca^{2+} channel appeared to be responsible for activity-dependent long-term depression in transmitter release.

E114

Effects of NAD^+ on the Stability of Soluble Proteins in the Pectoral Muscle of Neurotoxin 6-aminonicotinamide Treated Quail against the Proteolytic Digestion

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The effects of neurotoxin 6-aminonicotinamide (6-AN) on the stability of soluble proteins of quail pectoral muscle towards trypsin treatment were studied. SDS-polyacrylamide electrophoresis showed

that in the control group the soluble proteins with molecular masses corresponding to 130, 109, 96, 62 and 47 kDa were decreased whereas those with molecular masses 35.3, 34, 30, 28, 27, 21.7 and 19 kDa were increased. In 6-AN treated group the soluble proteins with molecular masses corresponding to 130, 96, 67, 60, 57, 50, 47, 36 and 32 kDa were decreased whereas those with molecular masses 30, 27, 26 and 18 kDa were increased. In the pair-fed group, soluble proteins with molecular masses 130, 80, 76, 69, 59, 47, 35 and 28 kDa were decreased whereas those with molecular masses 28, 26 and 25 kDa were increased. In the presence of 3 mM NAD^+ , the soluble proteins with molecular masses in the control group corresponding to 58.5, 55, 49, 43 and 26 kDa were reinforced. In 6-AN treated group, the soluble proteins with molecular masses 58, 49, 43 and 32 kDa were reinforced. In the pair-fed group, the proteins with molecular masses 93.5, 58, 50, 49, 44 and 40 kDa were reinforced. The results suggest that NAD^+ exerts nonspecific effects on the stabilization of soluble proteins of varying size of masses against trypsin digestion.

E115

Effects of Cadmium (Cd) on Total Lipid Content in Developmental Stages of the Greater Wax Moth, *Galleria mellonella*

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The beekeeping pest insect *Galleria mellonella* larvae (greater wax moth) were reared on an artificial diet contaminated independently with cadmium chloride at three different concentrations (5.0, 20.0, 40.0 $\mu\text{g/g}$ food fresh weight). Larvae were contaminated up to pupation, either from hatching or from pupae 2 days. Results of

these studies suggested that Cd exposure of *G. mellonella* may influence its whole body lipid contents. We decided, therefore, to analyze lipid content of *G. mellonella* exposed to different concentrations of Cd. Lipid concentrations were measured photometrically by phosphovanillin method. Significant decrease in the total lipid content was found in Cd-contaminated larvae and pupae.

E116

Purification and Characterization of the Lipid Transfer Particle (LTP) for the Larvae of Wax Moth, *Galleria mellonella*

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A lipid transfer particle (LTP) was partially purified from the hemolymph of wax moth, *Galleria mellonella* by 2 step KBr density gradient ultracentrifugation, anion exchange chromatography (resource Q) and gel permeation chromatography (superose 6) using fast performance liquid chromatography (FPLC) system. LTP of *Galleria mellonella* is composed of 3 subunits (Apo-LTP I, Apo-LTP II, Apo-LTP III). Molecular masses of each subunit were determined. The confirmation of LTP was determined by western blotting with polyclonal antibody for *Bombyx mori* LTP because LTP is non species-specific. Apo-LTP I and Apo-LTP III of *Galleria mellonella* react intensively with that of *B. mori* LTP whereas Apo-LTP II react weakly with that of *B. mori* LTP. Localization of LTP was also performed with polyclonal antibody for *B. mori* LTP. We will perform further works including purification of LTP, lipid transfer assay with radiolabeled HDLp, human LDL and determination of N-terminal sequencing and amino acid composition of each subunit.

E117

Cloning of the Bilin Binding Protein (BBP) cDNA in Fall Webworm, *Hyphantria cunea*

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The bilin-binding protein is a blue pigment protein binding heme-related compound in insect. The amino acid sequence from the bilin binding protein (BBP) of the fall webworm has been determined. The apoprotein shows a molecular mass of around 20 kDa. This cDNA has high homology with human apolipoprotein D, insecticyanin, and human retinol binding protein. Computer searches of data banks yielded in a new member of this superfamily, the alpha 2-microglobulin superfamily whose other members transport small hydrophobic ligands in a wide variety of biological contexts.

E118

Two Vitellogenic Carboxypeptidases (VCPs) Purified from Ovary of Mosquito, *Aedes aegypti*

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In response to a blood meal, the fat body of the female mosquito, *Aedes aegypti*, begins massive production of several yolk proteins which are subsequently stored in the developing oocytes by the receptor-mediated