E101 Tissue specific Juvenile Hormone Epoxide Hydrolase during the Last stadium of the Cabbage Looper, *Trichoplusia ni*

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Juvenile hormone epoxide hydrolase(JHEH), which may play a pivotal role in regulating insect juvenile hormone(JH) titer along with JH esterase, was identified in cabbage looper, *Trichoplusia ni.* A partition assay was developed to measure insect JH III metabolism in biological samples containing both JH esterase and JHEH activity. The assay utilizes commercially available radiochain [^{3}H]-labeled JH as substrate and the selective JH esterase inhibitor 3-octylthio-1,1,1,-trifluoro-2-propanone(OTFP). JH partitions into an isooctane phase and the metabolites JH acid, JH diol and JH diol-acid into aqueous methanol after incubation of JH substrate with inhibited and uninhibited sample. *In vitro* metabolism of JH III(5×10^{-4}M) was measured in fat body, midgut and integument homoginate. And also that was measured in different subcellular fraction(cytosol, nuclear, mitochondria, microsome). Microsomal protein was co-purified 168.5-fold with the JHEH activity through DEAE-sephacel, sephacryl S-200 and hydroxylapatite columns.

E102 Molecular Cloning and Expression of Calsequestrin-like protein in *C.elegans*

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Calsequestrin is one of the major calcium binding proteins localized in sarcoplasmic reticulum (SR) of skeletal and cardiac muscles. It is believed that calsequestrin buffers intraluminal calcium concentration in SR. However, we do not fully understand its role during E-C coupling and its interactions with other molecules. We have obtained a calsequestrin-like gene sequence from the genome sequencing project database. This gene encodes a highly charged protein of 417 amino acids, which exhibits 50% sequence similarity with its mammalian counterparts. In order to get the cDNA, we prepared a set of primers which covers the entire open reading frame of the gene and we could successfully amplify out a DNA fragment of the expected size (1.3 kb) from the *C. elegans* mixed-stage cDNA library. Sequence of the DNA matched perfectly to the previously reported sequence with several exceptions which could be polymorphysms. This work also verified the exon-intron boundaries of the gene. In order to obtain antibody of the gene product, the C-terminal half was expressed as a glutathionine S-transferase fusion protein and subsequently purified from *E.coli* using Glutathionine Sepharose 4B affinity column. We recently began to immunize rabbits with the fusion protein. Once we get the antibody, we plan to perform immunostaining and immunoblotting experiments to examine the spatial and temporal expression of the gene. It will be interesting to see if the gene expresses in all types of muscle or a small subset of muscle.