Preparation of a recombinant *Armadillidium vulgare* androgenic gland hormone with a glycan moiety

Sayaka Kotaka[1], Kazuki Hirabayashi[1], Yumi Sakamoto[1], Hirotaka Haibara[2], Shinji Nagata[2], Hiromichi Nagasawa[2], Susumu Izumi[1], Tsuyoshi Ohira[1]

[1]Department of Biological Sciences, Kanagawa University, Japan, [2]Department of Applied Biological Chemistry, The University of Tokyo, Japan

The sex differentiation in crustaceans is known to be controlled by a peptide hormone called androgenic gland hormone (AGH). AGH has been purified from the male specific androgenic gland (AG) of the terrestrial isopod *Armadillidium vulgare*. In 1999, the primary structure of *A. vulgare* AGH (Arv-AGH) was finally determined to be a heterodimeric glycoprotein. An *N*-linked glycan moiety in the mature Arv-AGH was found to be essential for biological activity. Therefore, in this study, a recombinant Arv-AGH with a glycan moiety was produced using a baculovirus expression system.

Insect Sf9 cells were infected with a recombinant baculovirus expression vector containing an Arv-AGH cDNA insert and subsequently recombinant Arv-AGH was expressed. Cell pellet and culture supernatant were subjected to Western blot analysis using an anti-Arv-AGH antibody. Two immunoreactive bands were detected at 17.5 and 22.5 kDa in cell pellet, while no bands were detectable in the culture supernatant. Since the calculated molecular mass of the recombinant Arv-AGH without a glycan moiety is 17,610, the former and the latter bands are thought to be non-glycosylated and glycosylated molecules of recombinant Arv-AGH, respectively. Now, we are attempting to purify the glycosylated recombinant Arv-AGHs from the cell pellet using reverse-phase HPLC.