Molecular biological analysis of parasitic strategies of endoparasitoid wasps

Generalistic endoparasitoid wasp *Asobara japonica* utilizes *Drosophila melanogaster* and the other broadly related *Drosophila* species as habitual hosts by using a notable feature of parasitism strategy. *A. japonica* females possess highly toxic venom and they inject the venom components with a lethal dose for the host *Drosophila simulans* at parasitism. A few seconds after the venom injection, they introduce eggs together with the lateral oviduct components that neutralize the detrimental effects of the venom toxin. In the present study, I demonstrated that the venom components impaired the cellular immune responses, spreading and phagocytosis, of the host hemocytes but did not affect the humoral immune responses such as expression of antimicrobial peptide genes. Furthermore, injection of the venom components activated a serine protease-like enzyme activity approximately 100 times in the plasma 4 h after the injection. The neutralizing factor in the oviduct blocked the protease-like enzyme activation, while it was unable to neutralize the venom-induced detriment of the host hemocytes. I also demonstrated that the venom components of *A. japonica* with a broad host range are much more toxic than those of *A. rossica* with a limited host range. Therefore, it is reasonable to propose that strong toxicity of *A. japonica* venom has contributed to exploiting wider ranges of host insect species during the evolutionary process. The subsequent characterization of the venom toxic factor revealed that this factor is a previously unidentified virus whose structural protein partly shares a similarity with that of known Iridovirus

In the study of *Cotesia kariyai* polydnavirus (CkPDV), I analyzed the surface structure of CkPDV particles by focusing on immunoevasive protein (IEP), which is previously identified as a mediator of immunoevasion by the wasp from the encapsulation reaction of the host insect's hemocytes, because it has been demonstrated to be present on the surface of *Cotesia kariyai* PDV (CkPDV) particles. Western blotting and immunoelectronmicroscopic analyses revealed that CkPDV particles are coated with the previously unidentified thin surface layer that composes IEP. This thin surface layer is essential for the virus infection because when the layer was removed from the CkPDV particles by shaking, they drastically lost their infectivity against the host tissues. Furthermore, IEP homolog was found to be expressed in the venom reservoirs of *C. kariyai* wasps. Although direct experimental evidence remains limited, it is believed that PDVs and venom proteins have co-evolved in endoparasitoid systems. The fact that IEP family proteins express both in the lateral oviducts and the venom reservoirs of *C. kariyai* wasps strongly confirms this expectation. All of the results in the present study strongly suggest that cooperative contribution of both ovaries and venom reservoirs of parasitoid wasps is essential to the successful parasitism, strongly implying their co-evolution in endoparasitoid system.