

### E133

#### Immunological Study of Lysozyme I and II Purified from Haemolymph of Immunized Larvae of Cabbage Butterfly, *Artogeia rapae*

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Lysozyme (EC 3.2.1.17) specifically hydrolyzes the glycosidic bonds of peptidoglycan present in the cell walls of bacteria and causes bacterial cell lysis. Lysozyme is one of the antibacterial proteins that are produced by the cabbage butterfly, *Artogeia rapae* in response to bacterial infection. We have determined about 40 amino acids at the N-terminus of two lysozymes, lysozyme I (LI) and II (LII) purified from the haemolymph of immunized larvae of *A. rapae*. LI and LII were confirmed to exist in normal haemolymph by immunodiffusion test using anti-LI and anti-LII, respectively. Distribution and correlation of LI and LII were investigated through immunodiffusion test and western blot analysis.

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#### DNA sequence encoding the antibacterial protein from *Artogeia rapae*

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The partial DNA sequence, possibly encoding the cecropin-like antibacterial protein, was obtained from the analysis of the DNA fragment which is amplified by use of RT-PCR and subcloned into the T-vector. Three different kinds of the degenerated primers were used in this study: two of them are located on the presequence and the remaining is on the 5'-end of the structural gene of the antibacterial gene. The new DNA sequence shows 52-76 % homology to the cecropins, based on the amino acid sequence deduced from the DNA. Especially, the highly conserved amino acid residues through the antibacterial proteins from the moth is also well conserved in this sequence. However, it does not match with any of the antibacterial proteins purified from *Artogeia rapae* even if it shows higher amino acid homology to the hinavin I, the antibacterial protein purified from the *Artogeia rapae*. The NH<sub>2</sub>-terminal domain seems to be more conserved than the COOH-terminal domain which is believed to confer the spectrum of the antibacterial activities.