

3-4-4. Molecular Cloning, Expression, and Characterization of the Chitinase Gene of the Spider, *Araneus ventricosus*

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Chitinase cDNA was cloned from the spider, *Araneus ventricosus*. The cDNA encoding the chitinase of *A. ventricosus* is 1293 base pairs long with an open reading frame of 431 amino acid residues. The deduced amino acid sequence of the chitinase gene of *A. ventricosus* showed 52.2% identity with *Glossina morsitans morsitans* chitinase and 32.3%~50.2% with other invertebrate chitinases. Phylogenetic analysis further confirmed that the deduced amino acid sequences of the *A. ventricosus* chitinase gene belonged to the invertebrate group. Southern blot analysis of genomic DNA suggested the presence of the *A. ventricosus* chitinase gene as a single copy and Northern blot analysis confirmed fat body-specific expression at the transcriptional level. The cDNA encoding the chitinase of *A. ventricosus* was expressed as a 61-kDa band in the baculovirus-infected insect cells and the extracts of the recombinant baculovirus-infected cells showed activity in the chitinase enzyme assay using 0.1% glycol chitin as a substrate. Furthermore, the Northern blot hybridization and Western blot analysis assay exhibited expression in fat body tissue, suggesting the fat body is a site where expression of chitinase is synthesized for facilitating the molting process.