

Molecular Characterization of Silkworm β -Glucosidase

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A digestive β -glucosidase cDNA was cloned from the silkworm, *Bombyx mori*. The *B. mori* β -glucosidase cDNA contains an open reading frame of 1,473 bp encoding 491 amino acid residues. The *B. mori* β -glucosidase possesses conserved amino acid residues involved in catalysis and substrate binding in glycosyl hydrolase family 1. Southern blot analysis of genomic DNA suggested the *B. mori* β -glucosidase was a single gene. Northern blot analysis and enzyme activity assay of *B. mori* β -glucosidase confirmed midgut-specific expression. The *B. mori* β -glucosidase synthesis in larval midgut was disappeared from the spinning stage and its expression level was significantly decreased during the starvation. The *B. mori* β -glucosidase cDNA was expressed as a 67-kDa polypeptide in baculovirus-infected insect Sf9 cells and the recombinant β -glucosidase was active on cellobiose, lactose, and salicin, indicating that the *B. mori* β -glucosidase is a Class 1 enzyme. Furthermore, N-glycosylation of recombinant *B. mori* β -glucosidase was revealed by tunicamycin to the recombinant virus-infected Sf9 cells, indicating that the carbohydrate moieties in *B. mori* β -glucosidase activity assay appear to be required for enzyme activity.