

Silkworm β -Glucosidase: cDNA Cloning, Expression and Characterization

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A cDNA encoding the digestive β -glucosidase from the midgut of the silkworm, *Bombyx mori*, was cloned and characterized. The *B. mori* β -glucosidase (EC. 3.2.1.21) cDNA contains an open reading frame of 1,473 bp encoding 491 amino acid residues. The *B. mori* β -glucosidase belongs to the insect β -glucosidase group and possesses the amino acid residues involved in catalysis and substrate binding common to insect β -glucosidase. Southern blot analysis of genomic DNA suggested the presence of *B. mori* β -glucosidase gene as a single copy and Northern blot analysis confirmed midgut-and larval stage-specific expression. The *B. mori* β -glucosidase synthesis in midgut was decreased during the starvation. The *B. mori* β -glucosidase cDNA was expressed as a 67-kDa polypeptide in the baculovirus-infected insect Sf9 cells and *N*-glycosylation of the recombinant β -glucosidase was revealed by tunicamycin to the recombinant virus-infected Sf9 cells, demonstrating that the silkworm β -glucosidase is glycosylated. The enzyme activity of the recombinant β -glucosidase was analyzed by Congo-Red assay and cellobiose zymography.