

## Bioconversion of chitinous compounds using chitin deacetylase

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Chitin deacetylases (EC 3.5.1.41) are the enzymes which hydrolyze amide-bonds of N-acetyl groups in chitin. The reaction pattern of an extracellular chitin deacetylase from a Deuteromycete, *Colletotrichum lindemuthianum* ATCC 56676 was investigated using chitooligosaccharides [(GlcNAc)<sub>n</sub>, n=3-6] as substrates. When 0.5% of (GlcNAc)<sub>n</sub> were deacetylated, the corresponding mono-deacetylated products were initially detected without any processivity, suggesting the involvement of a multiple-chain mechanism for the deacetylation reaction. The analysis of the structure of these first-step products indicated that the chitin deacetylase strongly recognizes a sequence of four N-acetyl-D-glucosamine (GlcNAc) residues of the substrate (defining the subsites for the four GlcNAc residues as -2, -1, 0, +1, respectively, from the non-reducing end to the reducing end), and the N-acetyl group in the GlcNAc residue positioned at the subsite 0 is exclusively deacetylated. When substrates of a low concentration (100 μM) were deacetylated, the initial deacetylation rate for (GlcNAc)<sub>4</sub> was comparable to that of (GlcNAc)<sub>5</sub>, while deacetylation of (GlcNAc)<sub>3</sub> could not be detected. Also, we found that the chitin deacetylase could acetylate free amino sugar residues into N-acetylated forms in the presence of 3.0 M sodium acetate. These studies offer novel methods for enzymatic control of N-acetyl groups in chitinous compounds, which could lead to better ways for utilization of chitinous biomass.