

# Identification of chitin binding proteins and characterization of two chitinase isoforms from *Vibrio alginolyticus* 283

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## Abstract

This study reports the isolation of chitin binding proteins secreted from a highly chitinase producing bacterium, *Vibrio alginolyticus* strain 283. Tryptic peptide mass analysis by HPLC–ESI/MS identified four proteins that bound specifically to chitin. Submission of mass fingerprinting data for database search identified the 90, 65, and 47 kDa proteins as chitinases. On the other hand, the 38 kDa protein was compatible with a sugar-inducible porin. The 90 and 65 kDa proteins, later designated Chi-90 and Chi-65 respectively, were further co-purified using Sephacryl 200 HR gel filtration chromatography and their enzymatic properties relatively studied. Kinetically, Chi-65 displayed 2.3 folds greater catalytic efficiency ( $k_{\text{cat}}/K_{\text{m}}$ ) towards *p*NP-diNAG than Chi-90. Investigation of the chitinase activity as a function of pH revealed that both enzymes worked best at pH of 6.5. At this pH, Chi-65 revealed almost seven folds higher activity than Chi-90. The similarity in peptide mass fingerprinting data, together with the equivalence of the optimal pH value and the resemblance in their hydrolytic patterns towards soluble chitooligosaccharides and insoluble chitin suggested that Chi-65 may be derived from the Chi-90 precursor.

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