Biochemical and Biological Characterization of a Recombinant Glycoside Hydrolase Family 18 (GH18) Chitinase from *Chromobacterium violaceum*

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**INTRODUCTION:** *Chromobacterium violaceum* is a Gram-negative, saprophytic, non-pathogenic and free living bacterium. The complete sequencing of its genome has revealed many genes encoding products of biotechnological interest, like chitinases. They are hydrolytic enzymes involved in the degradation of chitin, an insoluble polymer of β(1,4)-N-acetyl-D-glucosamine. These enzymes are of great biotechnological interest because they can be used to convert chitin in useful derivatives as well as they can be exploited as biocontrol agents against pests and pathogens. This study aimed to perform the biochemical/biological characterization of a recombinant chitinase, belonging to family GH18, from *C. violaceum* ATCC 12472. **MATERIALS AND METHODS:** The ORF CV2935 was amplified by polymerase chain reaction (PCR), cloned into the vectors pET303/CT-His and pPICZαA, aiming the heterologous expression in *Escherichia coli* and *Pichia pastoris*, respectively. In both systems, the enzyme was secreted into the culture medium, in its soluble and functional form. The secreted chitinase was purified to homogeneity by affinity chromatography on a chitin matrix followed by size-exclusion chromatography. **RESULTS AND DISCUSSION:** The recombinant chitinase produced in both microorganisms showed optimal activity at pH 3.0 and it was active after treated at temperatures up to 60 °C for 30 min. The enzyme produced in *P. pastoris* (45 kDa) was N-glycosylated as revealed by Schiff’s reagent and digestion with N-glycosidase F. Moreover, the glycosylated chitinase was found to be slightly more thermostable than the enzyme produced in *E. coli* (43 kDa). The rCV2935 showed hydrolytic activity against colloidal chitin, crab shell, synthetic substrates containing p-nitrophenol and glycol-chitin. The recombinant chitinase reduced the growth of the phytopathogenic fungus *Rhizoctonia solani*, and feeding bioassays with the cowpea weevil (*Callosobruchus maculatus*) showed that the rCV2935 caused a decrease in the weight of adults. **CONCLUSIONS:** The biochemical and biological studies of rCV2935 may be useful for biotechnological application of this enzyme.

Key-words: chitinase, *Chromobacterium violaceum*, recombinant

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