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Enzymatic Characterization of Heterologously Expressed Fungal β-Glucosidase BGLII in *Escherichia coli*

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Abstract. Saprophytic filamentous fungi such as Trichoderma sp. are capable of producing three categories of cellulase for cellulose degradation into glucose; endoglucanase, cellobiohydrolase and β-glucosidase. Trichoderma reesei in particular has an extensively studied cellulase enzyme system, establishing it as a model organism for enzymatic production. Studies on β-glucosidases produced by Trichoderma sp. have elucidated two variants, which have been classified into glycosyl hydrolase families 1 and 3, both including retaining enzymes capable of substrate hydrolysis via double-displacement method with retention of anomeric carbon. Extensive studies on BglI enzyme isolated has been compared and found to have sequence similarity to other known glycosyl hydrolase family 3 β-glucosidases, an enzyme that is cell wall bound or expressed extracellularly, affects rate of reducing sugar formation from cellulase and involved in synthesis of soluble inducer of cellulolytic enzymes. Comparatively, enzyme characterization of the second β-glucosidase, BglII, has been relatively recent. Nevertheless, it has been identified to be expressed intracellularly, has high affinity and catalyzes transglycosylation reactions on cellobiose. Of particular interest, BglII has shown lower sensitivity to glucose inhibition, an appealing character for bioprocess development for efficient lignocellulosic biomass saccharification. As such, using local Trichoderma sp., this study has sought to characterize the BglII gene isolated and following heterologous expression in Escherichia coli, characterize the recombinant enzyme for enzyme activity, glucose tolerance and product synthesis.

Keywords: β-glucosidase, glycosyl hydrolase family 1, glucose tolerance, heterologous expression