

Thesis Title Study of Chitinase and β -(1,3)-Glucanase Enzymes in the
 Rubber Latex

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ABSTRACT

The chitinase and β -(1,3)-glucanase enzyme were detected and found in B-serum prepared from luteoid particles of centrifuged rubber latex. Both enzyme in B-serum were characterized in this study. Colloidal chitin prepared from prawn shell was used as substrate in the chitinase assay. The chitinase enzyme had a narrow range of pH optimum between 4 - 5.5 with the maximum activity at pH 5.0. The temperature optimum was found between 45 - 55 ° C with maximum activity at 50 ° C. The enzyme was found to be stable in the pH range of 4 - 10. The chitinase was stable to the heat treatment up to 70 ° C, indicating this was a heat stable enzyme. The β -(1,3)-glucanase enzyme was also found to be moderately heat stable protein. The thermal stability of the glucanase was up to 60 ° C heat treatment. It also had a narrow range of pH optimum between 5 - 6 with the maximum activity at pH 5.5. Temperature optimum for the β -(1,3)-glucanase enzyme activity was found as a wide range between 40 - 60 ° C. Kinetics parameters of Km values of chitinase was 25 mM chitin for the B-serum enzyme and 17 mM chitin for the fractionated purified chitinase. The Km value of 40 mM laminarin was found for the B-serum β -(1,3)-glucanase enzyme.

Fractionation and purification of the enzymes were carried out using CM-cellulose chromatography. The chitinase was found to consist of 3 different isoenzymes by CM-cellulose chromatography while the β -(1,3)-glucanase showed only a single enzyme activity peak. The three chitinases, designated as CM I, CM II and CM III, were fractionated and characterized. All the three chitinase isozymes had the same molecular weight of 26 kD. The pI values for chitinase isozymes were 9.3, 9.7 and 9.8 for CM I, CM II and CM III, respectively.

Antifungal activities of the enzymes and protein in B-serum were carried out in this study. The B-serum containing both the chitinase and β -(1,3)-glucanase activities was effective on growth inhibition in the antifungal activity assays. All the six tested fungi were inhibited by the B-serum. Fractionated purified chitinase and C-serum showed growth inhibition on three of fungi tested. Chitosan which was well known as fungal growth inhibitor was used as a reference to this antifungal study. The chitosan solution showed growth inhibition on all the tested fungi except *Sporotrichum pulverulentum*. These studies suggested an opportunity for potential application of the latex B-serum on many different aspects.