A PEPTIDE ELISA TO DETECT ANTIBODIES AGAINST PYTHIUM INSIDIOSUM BASED ON PREDICTED ANTIGENIC DETERMINANTS OF EXO-1,3-β-GLUCANASE

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Abstract. Human pythiosis is a life-threatening infectious disease caused by the oomycete Pythium insidiosum. Diagnosis of pythiosis relies on culture identification, serodiagnosis, and molecular-based assay. Preparation of a serodiagnostic test requires culture filtrate antigen (CFA) extracted from the live pathogen. A 74-kDa immunoreactive protein of P. insidiosum, is encoded by the exo-1,3-β-glucanase gene (PinsEXO1). PinsEXO1 protein is recognized by sera from pythiosis patients but not by sera from uninfected patients; therefore, this protein could be used to detect anti-P. insidiosum antibodies. In this study we aimed to: identify, synthesize, and evaluate an antigenic determinant (epitope) of PinsEXO1 to be used to serodiagnose pythiosis based on peptide ELISA, and to compare the diagnostic performance of that test with the current CFA-based ELISA. Two antigenic determinants of PinsEXO1 (Peptide-A and -B) were predicted using the PREDITOP program. The sera from 34 pythiosis patients and 92 control subjects were evaluated. Peptide-A, Peptide-B, and CFA-based ELISAs all had a specificity of 100%. Peptide-B ELISA had a sensitivity of 91% and an accuracy of 98% and both Peptide-A and CFA-based ELISAs had a sensitivity of 100% and an accuracy of 100%. Peptide-A is a more efficient epitope than Peptide-B, and can be used as an alternative antigen to develop a serodiagnostic assay for pythiosis.

Keywords: Pythium insidiosum, pythiosis, diagnosis, ELISA, exo-1,3-β-glucanase

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