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Expression and use of Truncated Recombinant Flagellin Protein (FlaB) in ELISA for Diagnosis of Leptospirosis

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Leptospirosis, caused by Leptospira spp. is a zoonotic infection which exhibits a broad spectrum of clinical manifestations, ranging in severity from acute to chronic with multiorgan syndrome to fetal.

The diagnosis of Leptospirosis depends either on the detection of antibodies in the sera or the presence of the organism in the tissues or body fluids. Since the isolation of Leptospires is difficult and laborious, serological diagnosis is extensively used. The group specific microscopic agglutination test (MAT) is widely used as a standard reference test.

The purpose of this study was to investigate the application of recombinant FlaB in the diagnosis of Leptospirosis, and to make the simple and rapid diagnostic system.

A periplasmic flagellin gene, *flaB*, of *Leptospira interrogans* Icterrohaemorragiae, *lai* (strain HY10) was expressed in *E. coli* BL21. The flaB truncated structure gene, which was previously cloned pGEMT. Under the employed, the *flaB* was expressed as inclusion bodies. Recombinant FlaB was about 29 kDa and allowed for the purification of protein by Ni-chelate affinity chromatography. Western blot analysis demonstrated that purified FlaB was reacted by MAT positive leptospirosis patient sera, but not reacted other diseases patients sera, Malaria, HBV, HFRS, Tsutsugamushi, and Syphilis.

A population of 30 MAT-positive and 30 MAT-negative serum samples was tested by ELISA using purified the truncated recombinant FlaB as antigen. The ELISA cutoff was explained in mean and 99% confidence interval. Compare to the results of MAT, the positive predictive value of ELISA 80.3% and negative predictive value was 71.6%. These results suggested that the truncated recombinant FlaB is one of the favorable candidate antigen for diagnosis on Leptospirosis.