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## **STRUCTURAL BIOLOGY AND PROTEOMICS**

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### IMMUNODIAGNOSTIC TEST KITS FOR RAPID DETECTION OF FASCIOLOSIS AND PARAMPHISTOMOSIS

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Statement of the Problem: Tropical fasciolosis caused by *Fasciola gigantica* infection and paramphistomosis caused by paramohistomes are the major diseases infecting ruminants and humans in the tropical regions of Africa and Asia including Thailand. Parasitological diagnosis of fasciolosis is often unreliable and possesses low sensitivity. Therefore, the detection of circulating parasite antigens is thought to be a better alternative for diagnosis of fasciolosis, as it reflects the real parasite burden.

**Methodology & Theoretical Orientation:** In this study, we have produced a monoclonal antibody (moAb) against native and recombinant antigens and developed both sandwich enzyme-linked immunosorbent assay (sandwich ELISA) and immunochromatographic (IC) test for rapid detection of circulating antigens in the sera or stool from mice experimentally and cattle naturally infected with *Fasciola gigantica* or paramphistomes.

**Findings:** The lower detection limits of sandwich ELISA and IC test were 3 pg/ ml and 0.256 ng/ml, respectively. Sandwich ELISA and IC test could detect *F. gigantica* infection from day 1 to 35 post infection. In experimental mice, the sensitivity, specificity and accuracy were 95%, 100% and 98.6% (for sandwich ELISA), and 93%, 100% and 98.2% (for IC test), while in natural cattle they were 98.3%, 100% and 99.5% (for sandwich ELISA) and 96.7%, 100% and 99.1% (for IC test).

**Conclusion & Significance**: These two assay methods showed high efficiencies and precisions for diagnosis of fasciolosis and paramphistomosis.



Figure.1: An immunochromatographic (IC) strip test is developed for diagnosis of fasciolosis by *F. gigantica*: experiment trial. (A) A schematic diagram of the immunochromatographic (IC) strip test showing several

components: a sample pad, a conjugate pad, an immobilized nitrocellulose membrane (control and test antibody) and an absorbent pad. (B) The samples of the IC strip test for deciding the results: a positive result shows two red dots at the test and control regions, while a negative result exhibits only one red dot in the control region. The strip tests are invalid when there is no red dot at the control region.

#### **Recent Publications**

- Anuracpreeda P, Kullanid Tepsupornkul, Chawengkirttikul R (2017). Immunodiagnosis of paramphistomosis using monoclonal antibody-based sandwich ELISA for detection of *Paramphistomum gracile* circulating 16 kDa antigen. Parasitology. 144: 899-903.
- 2. Anuracpreeda P, Amaya Watthanadirek, Chawengkirttikul R, Sobhon P (2017). Production and characterization of a monoclonal antibody specific to 16 kDa antigen of *Paramphistomum gracile*. Parasitol Res. 116: 167–175.
- Anuracpreeda P, Chawengkirttikul R, Sobhon P (2016). Immunodiagnostic monoclonal antibody-based sandwich ELISA of fasciolosis by detection of *Fasciola gigantica* circulating fatty acid binding protein. Parasitology. 143: 1369-1381.
- Anuracpreeda P, Chawengkirttikul R, Sobhon P (2016) Antigenic profile, isolation and characterization of whole body extract of *Paramphistomum gracile*. Parasite Immunol. 38: 431-438.

### BIOGRAPHY

Panat Anuracpreeda is an Associate Professor of Mahidol University and belongs to Molecular Medical Biosciences Cluster. He is associated with Institute of Molecular Biosciences. He has his research interests in parasite immuno-molecular biology, advance hybridoma technology, advance immuno-molecular diagnostic assays and advance immuno-molecular therapy.

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