

## **Development of molecular-based kits for rapid diagnosis of leptospirosis and other important economic diseases.**

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### **Introduction**

The technologies available today make it possible to raise the level of understanding of epidemiology and pathogenesis of diseases for more effective diagnosis and control strategies. It is now possible to bring sophisticated diagnostic technologies out of research laboratories to the field and hospitals where effective control requires rapid diagnosis and early treatment. It is important to further develop technologies to improve the diagnosis and prevention of diseases, both in animals and humans. Very often the source of infection for humans is the animals which maintain the infectious disease. In the diagnosis of diseases, there are many times when the clinical signs and symptoms of the diseases are not pathognomonic and clinicians quite often considered them as pyrexia of unknown origin (PUOs). The prevalence of leptospirosis in animals and humans is very high and warrants control strategies to bring it down to an acceptable level. It is a disease of great economic and public health interest.

### **Materials and Methods**

**Bacterial strains and growth conditions:** The leptospiral serovars used will be from our collection obtained through earlier surveys and reference strains and isolates collected from laboratories. Culture conditions for the leptospires will essentially be those described by Johnson and Harris (1967).

**DNA extraction:** Leptospires will be grown in JS medium. The preparation and extraction of leptospiral genomic DNAs will be as in the standard procedures (Pacciarinni et al., 1992).

**PCR assay and hybridization experiments:** Two sets of primers for PCR will be used and they will be synthesized through the Applied Biosystems DNA synthesizer.

**Detection and characterization of PCR products:** Amplified products will be separated by electrophoresis and subsequently Southern blotted to a nylon membrane. To detect the PCR fragments generated by the primers, two oligonucleotides tailed with digoxigenin will be used as probes. Restriction endonuclease analysis will be done to characterize the PCR-amplified products.

### **Results and Discussion**

New technique in the detection and identification of leptospirosis and similar diseases will be developed. Production of a simple, rapid and cheap diagnostic kit for definitive diagnosis, surveys and monitoring of the disease for prevention and control programmes would be available. It will contribute to the human resource development by providing research facilities and training for scientists and students. The Ministry of Agriculture Malaysia specifically the Department of Veterinary Services would benefit tremendously with availability of the rapid diagnostic kit and the disease will now be diagnosed quickly and conclusively. With the diagnostic kit and technologies developed, animal farmers would benefit greatly as their animals could be examined, carriers of the infection identified and control and prevention programmes could be quickly be implemented. Subsequently, the animals will be in better health and more productive. Farmers and related workers involved with livestock and the environment like butchers, joggers, loggers, housewives, picnickers would not be exposed to one of the most infectious diseases in Malaysia.

### **Conclusions**

The diagnostic kit developed would speed up diagnosis and proper treatment could then be instituted. Control and preventive programmes could then be formulated to reduce the high prevalence of leptospirosis in animals and humans in the country.

### **Benefits from the study**

The diagnostic techniques developed could be made into test kits which would then speed up diagnosis and early treatment could then be instituted. Control and preventive programmes could be formulated to help reduce the high prevalence of infection in animals and humans. Public health would be improved leading to reduced loss of manpower and mortalities. Living standard would be enhanced and the public will be free to lead a safe and healthy lifestyle.

### **Project Publications in Refereed Journals**

Khairani-Bejo, S. and Bahaman, A.R. 2000: *Leptospira interrogans* serovar *hardjo* infection in cattle in Malaysia:

Serological and bacteriological prevalence. *Journal of Veterinary Association Malaysia* 12: 15-17.

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- Ali, A.S.; Mohd-Azmi, M.L.; Aini, I.; Sheikh-Omar, A.R. and Bahaman, A.R. (2001): Effects of cyclophosphamide and corticosteroids on virus growth and antibody response to non-pathogenic pseudorabies virus. Veterinary Research Communications (submitted).
- Ali, A.S.; Mohd-Azmi, M.L.; Aini, I.; Sheikh-Omar, A.R. and Bahaman, A.R. (2001): Serologic and antigenic relationship between Malaysian pseudorabies virus isolates. Journal of the Veterinary Association Malaysia (submitted).

<i>Expertise Development</i>			
<b>Name of Graduate</b>	<b>Degree Awarded</b>	<b>Field of Expertise</b>	<b>Graduation Year</b>
<b>El-Jahii, I.M.</b>	<b>Ph.D</b>	<b>Veterinary Microbiology</b>	<b>2000</b>
<b>Thongted Punbunchorn</b>	<b>M.Sc</b>	<b>Veterinary Microbiology</b>	<b>2001</b>
<b>Siti Khairani Bejo</b>	<b>Ph.D</b>	<b>Veterinary Microbiology</b>	<b>2001</b>

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