



***SEROPREVALENCE OF LEPTOSPIROSIS
AMONG MILITARY DOGS
FROM JOHOR, MALAYSIA***

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**SEROPREVALENCE OF LEPTOSPIROSIS
AMONG MILITARY DOGS
FROM JOHOR,
MALAYSIA**

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II

It is hereby certified that we have read this project paper entitled “Seroprevalence of Leptospirosis among Military Dogs from Johor, Malaysia” by Byron Chong Yeu Liang and in our opinion it is satisfactory in terms of scope, quality, and presentation as partial fulfillment of the requirement for the course VPD 4999 – Final Year Project

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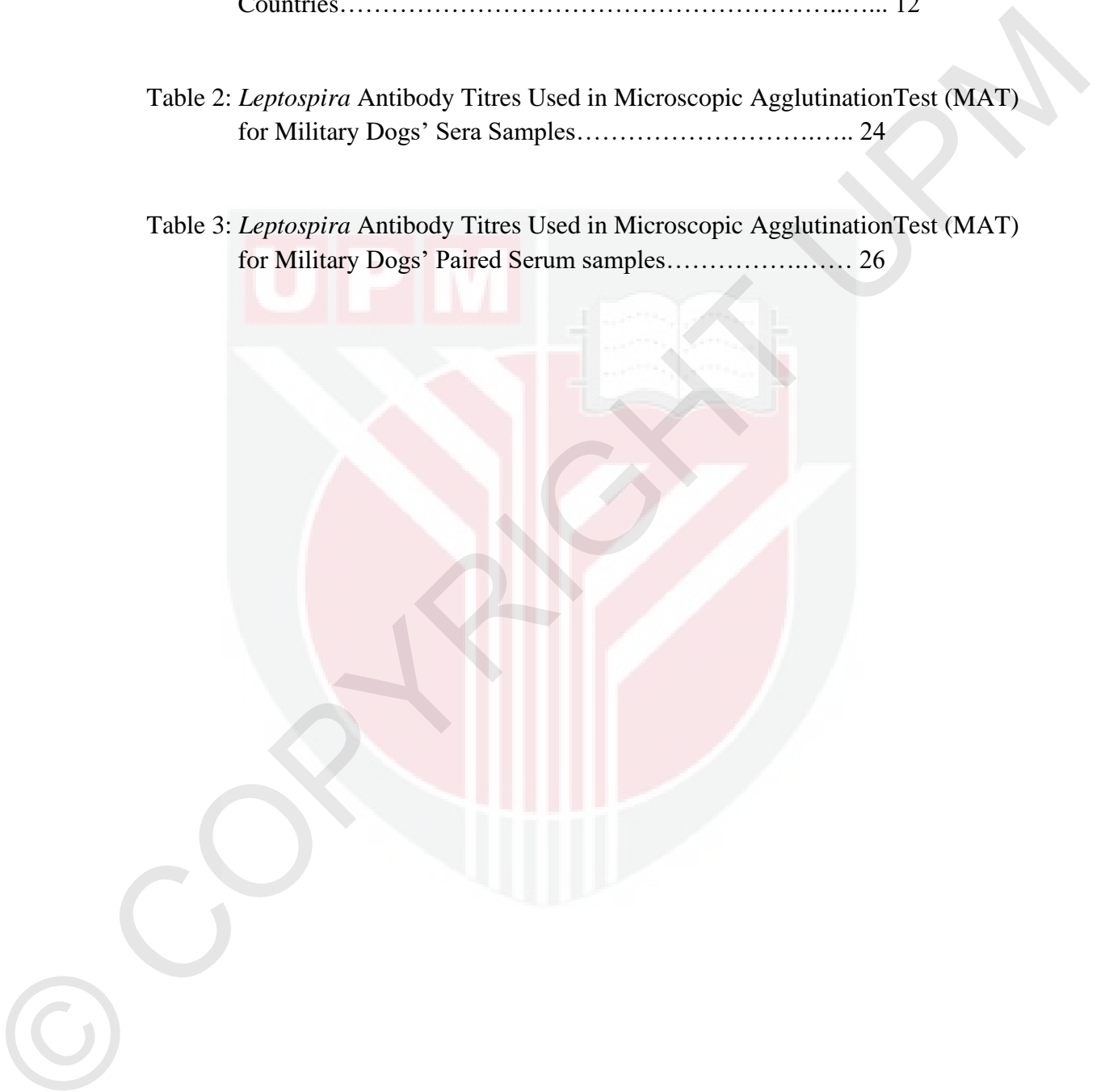
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LIST OF ABBREVIATIONS

DNA	Deoxyribonucleic acid
EDTA	Ethylenediaminetetraacetic acid
ELISA	Enzyme-linked Immunosorbent Assay
EMJH	Ellinhausen-McCullough-Johnson-Harris
IACUC	Institutional Animal Care and Use Committee
Ig	Immunoglobulin
IHA	Indirect Hemagglutination Assay
LFA	Lateral Flow Assay
MAT	Microscopic Agglutination Test
mL	milliliter
°C	degree Celsius
OMP	Outer Membrane Proteins
PBS	Phosphate Buffer Saline
PCR	Polymerase Chain Reaction
Rpm	Revolutions per minute
RSAT	Rapid Slide Agglutination Test
μL	microliter

ABSTRACT

An abstract of the project paper presented to the Faculty of Veterinary Medicine in
partial fulfilment of the Course VPD 4999 – Final Year Project

**SEROPREVALENCE OF CANINE LEPTOSPIROSIS AMONG MILITARY
DOGS FROM JOHOR, MALAYSIA**

By

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2017

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Canine leptospirosis is a well-known bacteria zoonotic disease with worldwide distribution. A study on seroprevalence of canine leptospirosis was carried out in a dog population (n=40) consisted of 13 searcher dogs, 15 tracker dogs, and 12 guard dogs in Military Camp, Johor, Malaysia. A total of forty blood sera were obtained and examined by Microscopic Agglutination Test (MAT) against ten *Leptospira* serovar (*Canicola*, *Pomona*, *Icterohaemorrhagiae*, *Grippityphosa*, *Lai*, *Australis*, *Pyrogenes*, *Javanica*, *Bataviae*, and *Celledoni*) with titres $\geq 1:80$ were recorded as positive results. A seroprevalence rate of 37.5% was observed in this study. *Leptospira interrogans* serovar *Icterohaemorrhagiae* 15% (n=6/40) was found to be most prevalent serovar followed by *Grippityphosa* 12.5% (n=5/40), *Pomona* 5% (n=2/40), and *Canicola* 5% (n=2/40). All seropositive dogs were searchers (n=9/40) and trackers (n=6/40), and guard dogs were all seronegative. Paired-serum samples were obtained, but there is no detection of four-fold increase in MAT. In this study, working environment, and food contamination were believed as the sources of infection in this population of military dogs. To prevent and control of canine leptospirosis in Malaysia, commercial vaccines against the four major serovars of *Canicola*, *Pomona*, *Icterohaemorrhagiae*, and *Grippityphosa* in all military dogs is highly recommended.

Keywords: Canine Leptospirosis, Seroprevalence, Military Dogs, Microscopic Agglutination Test

ABSTRAK

Abstrak daripada kertas projek yang dikemukakan kepada Fakulti Perubatan Veterinar untuk memenuhi sebahagian daripada keperluan kursus VPD 4999- Projek Ilmiah Tahun Akhir

PENGESANAN LEPTOSPIROSIS DALAM KALANGAN ANJING PERANG DARI**JOHOR, MALAYSIA**

Oleh

Byron Chong Yeu Liang

2017

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Leptospirosis (Penyakit Kencing Tikus) adalah bakteria zoonotik yang dilaporkan di seluruh dunia. Kajian yang dijalankan adalah untuk prevalen mengesan leptospirosis dan serovars yang terlibat dalam Kem Tentera, Johor, Malaysia. Sampel darah diperoleh daripada 40 anjing terdiri daripada 13 pencari, 15 pengesan, dan 12 pengawal anjing. Ujian serologi Agglutinas Mikroskopik (Microscopic Agglutination Test) telah digunakan untuk mengesan antibodi anti-leptospiral dalam sampel serum kepada 10 serovars (Canicola, Pomona, Icterohaemorrhagiae, Grippityphosa, Lai, Australis, Pyrogenes, Javanica, Bataviae, dan Celledoni) dengan rekod positif titer antibody penentuan adalah 1:80. Kelaziman

keseluruhan leptospirosis adalah 37.5%. *Leptospira interrogans* serovar *Icterohaemorrhagiae* 15% (n=6/40) mencatat prevalen tertinggi berbanding serovar-serovar lain *Grippityphosa* 12.5% (5/40), *Pomona* 5% (2/40), dan *Canicola* 5% (2/40). Semua anjing sero-positif terdiri daripada kalangan pencari (n=9/40), dan pengesan (n=6/40), manakala pengawal anjing adalah semua sero-negatif. Ujian kali kedua juga dijalankan ke atas semua sampel darah tetapi keputusan yang diperoleh tidak menunjukkan peningkatan empat kali ganda dalam MAT. Dalam kajian ini, tempat latihan dan pencemaran makanan adalah sumber yang menyebabkan penyebaran leptospirosis dalam anjing perang. Untuk pencegahan dan kawalan leptospirosis di Malaysia, vaksin komersial menentang empat serovars *Canicola*, *Pomona*, *Icterohaemorrhagiae*, dan *Grippityphosa* yang digunakan bagi semua anjing perang adalah digalakkan.

Kata kunci: Leptospirosis, serologi, anjing perang, Ujian Agglutinas Mikroskopik

1.0 INTRODUCTION

Leptospirosis is an emerging or re-emerging infectious zoonotic disease occurs worldwide. It is caused by spirochetes of genus of *Leptospira*, which belong to the family Leptospiraceae, order Spirochaetales, which is characterized as highly motile, thin, filamentous spirals with hook-shaped at the both ends. Leptospirosis is commonly occurred in tropical and sub-tropical regions with relating to the climate, and environmental condition (WHO, 2003; Goldstein, 2013).

In human, it is known as Weil's disease. The first leptospire was isolated in Kyushu, Japan in 1914. The first case of leptospirosis in Malaysia was isolated in human by Fletcher in 1925. In addition, Fletcher also success in isolating leptospire from streams and ponds in Malaysia (Bahaman *et al.*, 1988). The recent study reported that a seroprevalence of human leptospirosis in Malaysia was 9.77% (n=829) (Samsi *et al.*, 2013).

In dog, before 1960, *L. interrogans* serovars Icterohaemorrhagiae and Canicola were believed responsible for most of the clinical manifestation of canine leptospirosis. However, to date, the most common serovars reported are *L. kirschneri* serovar Grippotyphosa, *L. interrogans* serovar Pomona, and *L. interrogans* serovar Bratislava. Hence, new vaccines are developed that include four serovars namely, Pomona, Canicola, Grippotyphosa, and Pomona (Goldstein, 2010). To date, dogs have become source of outbreak that transmit Leptospirosis to human being (Guernier *et al.*, 2016). There were studies shown that serovars (serogroup) Canicola (Canicola), Fortbragg (Autumnalis), Icterohaemorrhagiae (Icterohaemorrhagiae), and Bim (Australis) were isolated from different places with outbreak of human leptospirosis associated with pet dogs and guard dogs (Haunz & Cardy, 1952; Barkin

& Glosser, 1973; Fraser *et al.*, 1973; Feigin *et al.*, 1973; Everard *et al.*, 1987). Hence, higher seroprevalence of leptospirosis in dogs may contribute to public health issue due to close contact and relationship namely farmers, veterinarians, laboratory staff, pet owners, abattoir worker, and etc (WHO, 2013).

In Malaysia, thirty-seven *Leptospira* serovars had been isolated from both human and animals (Bahaman *et al.*, 1988). According to Fletcher (1928), the first canine leptospirosis in Malaysia was serovar Hebdomadis. The recent studies show that the seroprevalence of canine leptospirosis in pet dogs from selected area of Klang Valley was 7% (n=57) (Lau *et al.*, 2016). In additional, another study in a selected dog shelter in Selangor reported a seroprevalence of 3.8% (n=80) (Khor *et al.*, 2016). Furthermore, the latest studies show in working dog in Malaysia reported a seroprevalence of 3.1% (n=96) (Wong, 2016).

The result of this preliminary study can be used to provide a general overview of the current status of canine leptospirosis in selected areas in Malaysia. This information can provide the veterinarians with the estimation of the spatial distribution and trend of this infectious disease among the dog populations.

The objectives of this study were:

1. To survey the seroprevalence of canine leptospirosis among military dogs in military camp, Johor, Malaysia.

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