



***IN VITRO ASSESSMENTS OF BETEL LEAVES (*Piper betle*) AQUEOUS
AND ETHANOL EXTRACTS ON BROWN DOG TICK
(*Rhipicephalus sanguineus*)***

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***IN VITRO* ACARICIDAL ASSESSMENT OF BETEL LEAVES (*Piper betle*)**

AQUEOUS AND ETHANOL EXTRACTSON

BROWN DOG TICK (*Rhipicephalus sanguineus*)

MUHAMMAD HAFIZ BIN SHAMSI

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Faculty of Veterinary Medicine, Universiti Putra Malaysia
in partial fulfilment of the requirement for the
DEGREE OF DOCTOR OF VETERINARY MEDICINE

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CERTIFICATION

It is hereby certified that we have read this project paper entitled “*IN VITRO* ACARICIDAL ASSESSMENT OF BETEL LEAVES, *Piper betle* AQUEOUS AND ETHANOL EXTRACTS ON BROWN DOG TICK *Rhipicephalus sanguineus*” by Muhammad Hafiz bin Shamsi 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 – Project.

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DEDICATION

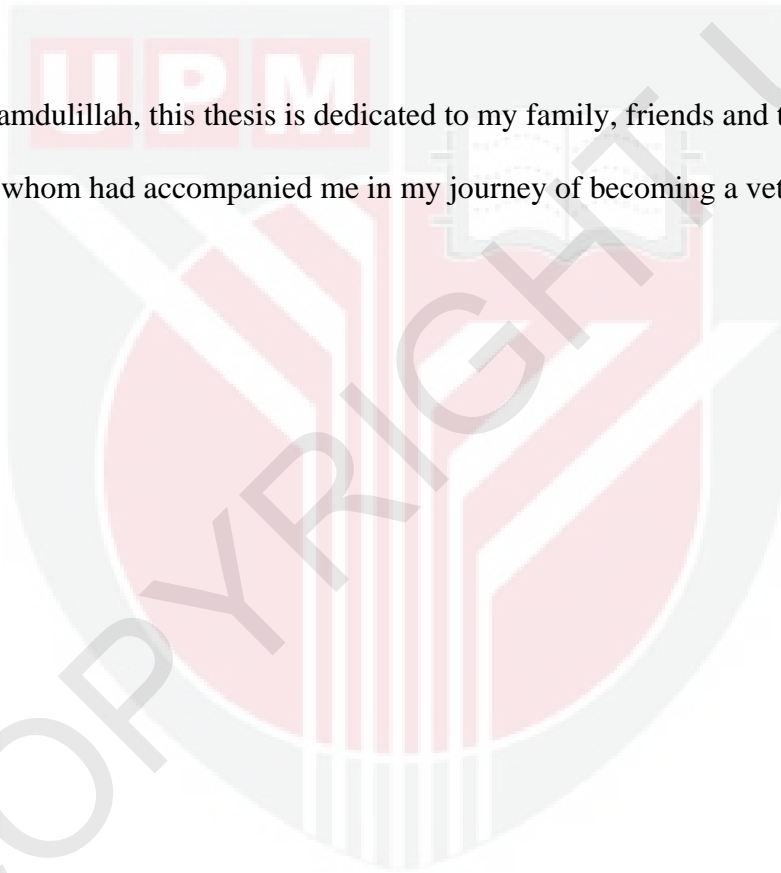
“Behold in the creation of the heavens and earth,

And the alternation of night and day there are

Indeed signs for men of understanding”

Surah ‘Ali ‘Imran 3:190

Alhamdulillah, this thesis is dedicated to my family, friends and the one I love,
whom had accompanied me in my journey of becoming a veterinarian.



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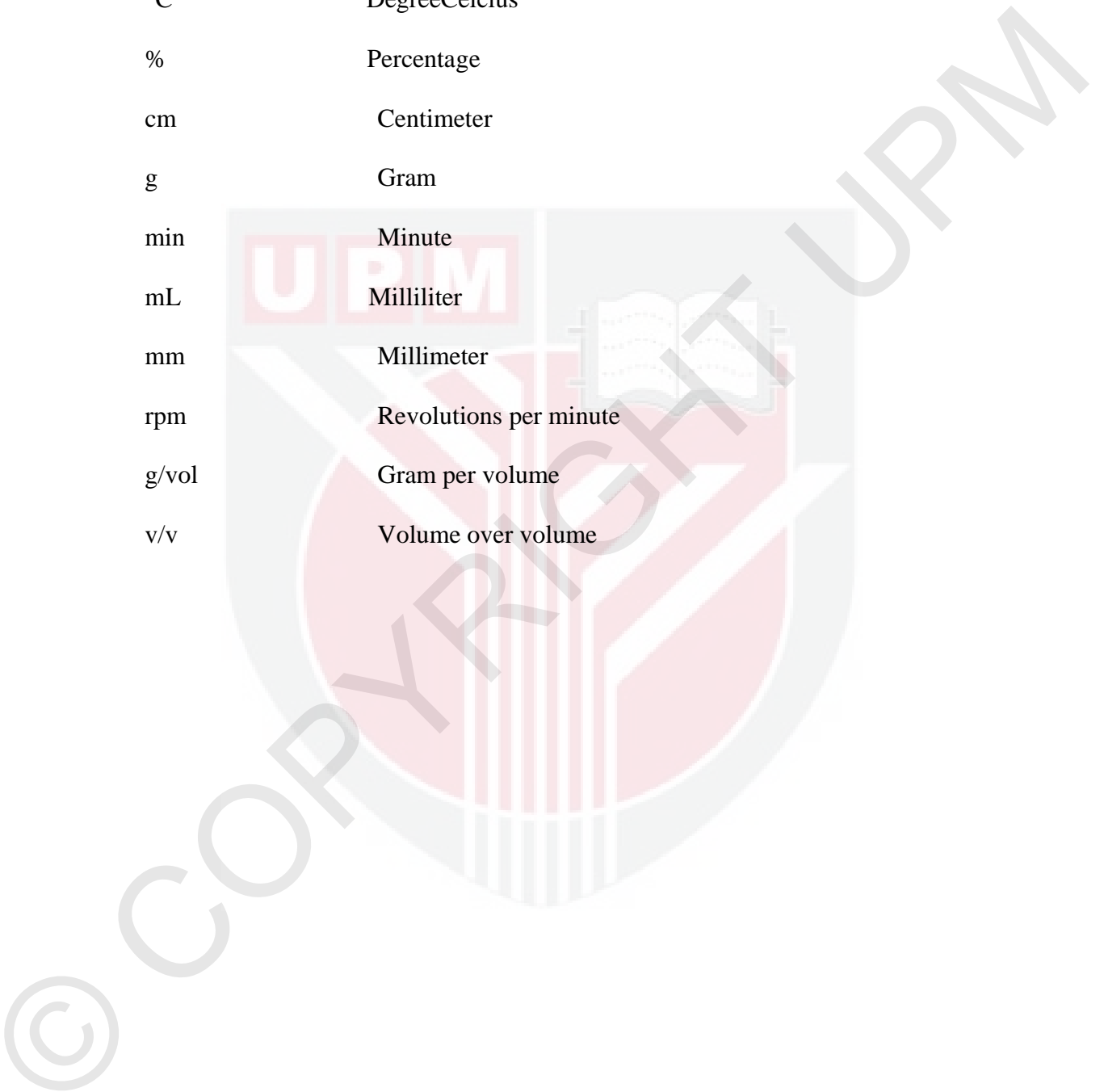


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LIST OF ABBREVIATIONS

°C	DegreeCelcius
%	Percentage
cm	Centimeter
g	Gram
min	Minute
mL	Milliliter
mm	Millimeter
rpm	Revolutions per minute
g/vol	Gram per volume
v/v	Volume over volume



ABSTRAK

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

PENILAIAN *IN VITRO* KESAN AKARI EKSTRAK AKUADAN ETANOL**DAUN SIRIH (*Piper betle*) TERHADAP KUTU ANJING****(*Rhipicephalus sanguineus*)****Oleh****MUHAMMAD HAFIZ BIN SHAMSI****2017****Penyelia: Profesor Madya Dr Hassan Hj. Mohd Daud****Penyelia Bersama: Dr. Nur Mahiza Md Isa**

Kutu anjing kelabu, *Rhipicephalus sanguineus*, boleh dijumpai seluruh dunia menghisap darah anjing dan malahan juga pada mamalia lain dan manusia, sambil pada masa yang sama menyebarkan pelbagai jenis penyakit. Terdapat pelbagai jenis kimia sintetik bagi menangani masalah jangkitan kutu ini. Oleh itu matlamat utama kajian ini dijalankan adalah untuk mengkaji cara semulajadi untuk membunuh kutu ini. Untuk itu daun sireh (*Piper betle*) telah dipilih kerana diketahui mengandungi pelbagai jenis fitokimia yang bioaktif. Kutu anjing telah dikutip dari anjing

geladangan di Unit Kawalan Vektor, Dewan Bandaraya Kuala Lumpur menggunakan penyepit dan disimpan didalam bekas khas yang telah diubahsuai. Daun sireh yang segar dipetik dari Taman Pertanian Universiti, Universiti Putra Malaysia. Daun dikeringkan menggunakan ketuhar udara panas dan kemudian di hancurkan menggunakan pengisar dapur. Daun yang telah dikisar telah direndam dalam larutan ethanol dan air, ditapis dan dikeringkan menggunakan penyejat putar untuk mendapatkan pati ekstrak herba. Dua jenis eksperimen berbeza telah dijalankan bagi menguji kesan ekstrak terhadap kutu yang dilarutkan dari 5^0 ke 5^{-4} . Keputusan menunjukkan bahawa kedua-dua jenis eksperimen bagi larutan etanol mempunyai kesan ketara sebagai racun akari dengan kadar dos maut dari kadar cairan satu ke lima kali. Larutan ekstrak etanol mempunyai kadar kematian yang lebih tinggi di mana 26 dari 50 ekor kutu berjaya dibunuh (52%), manakala, bagi larutan ekstrak akua hanya mampu membunuh 8 ekor dari 50 ekor kutu (16%). Sebagai penutup, ekstrak daun sireh menunjukkan potensi untuk dijadikan racun akari bagi *R. sanguineus*.

Katakunci: Ekstrak, *Piper betle*, racun akari, *Rhipicephalus sanguineus*

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.

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AQUEOUS AND ETHANOL EXTRACTS ON BROWN DOG TICK
(*Rhipicephalus sanguineus*)**

By

MUHAMMAD HAFIZ BIN SHAMSI

2017

Supervisor: Assoc. Professor Dr Hassan Hj. Mohd Daud

Co-Supervisor: Dr. Nur Mahiza Md Isa

The brown dog tick, *Rhipicephalus sanguineus*, is found worldwide with high tendency to feed on dogs and other mammals and human while spreading various types of pathogens. Various synthetic chemicals are available to treat the infestation. The main objective of this study was to find a natural alternative as a potential anti-tick compound. Naturally, betel leaves was chosen as it has many ascribed phytochemicals. The brown dog ticks were collected from stray dogs compounded at Vector Control Unit, Kuala Lumpur City Municipal, using forceps and kept alive in

modified specimen containers. Fresh *P. betle* leaves were collected from the University's Agriculture Park, Universiti Putra Malaysia. The leaves were dried in hot-air oven and grounded using bench-top grinder. Later the powder was soaked in ethanol and water dilutions, filtered and dried using the rotary evaporator to obtain the crude extract. Two different set of experiments were set-up, whereby ticks were exposed to both of the extracts prepared in dilutions ranging from 5^0 to 5^{-4} . Results from both experiments showed significant effect of the betel leaves as acaricide with the lethal dose from one to five dilutions of the pure extract. The ethanol extract have higher efficacy with 26 out of 50 ticks died (52%), meanwhile for the aqueousextract only 8 ticks died out of 50 (16%). In conclusion, the betel leaves ethanolic extract showed the potential to be used as acaricides for *R. sanguineus*.

Keywords: Acaricides, extract, *Piper betle*, *Rhipicephalus sanguineus*

1.0 INTRODUCTION

The brown dog tick, *Rhipicephalus sanguineus* (Latreille, 1806) is an arthropod, which make it of great significant in human medical and veterinary aspects. It feeds on blood for survival, and as it feeds it causes direct damage to the host and able to transmit various agents of diseases (Oliver, 1989). Examples of the pathogenic agents are *Babesia vogeli*, *B.gibsoni*, *Hepatozoon canis*, *Rickettsia conorii*, *R. rickettsii*, *Ehrlichia canis*, *Anaplasma platys* (Dantas-Torres, 2008; Fourie et al., 2013; Nicholson, Allen, McQuiston, Breitschwerdt, & Little, 2010).

It can feeds on other mammals including domestic animals and humans, which likely to happen when there is no dog or food source around and is able to complete its entire life cycle indoors (Lord, 2008). Thus, the infestation of the tick could increase drastically with just a few tick presence in the house or in the kennel especially after the walk about in the field (Dantas-Torres, 2008; Dantas-Torres, Figueredo, & Brandão-Filho, 2006; Lord, 2008; Nicholson et al., 2010).

Currently, there are various methods being used to control the tick flare, such as chemical controls using spot-on formulations, impregnated collars, shampoos, sprays, dips and powders, containing various active pesticides compounds e.g. Fipronil, amitraz, carbaryl, and pyrethroids (Dantas-Torres, 2008; Fourie et al., 2013; Williams et al., 2015). However, the increase trend of misuse of the pesticides brings a lot of concern as it could cause environmental pollution and toxicity to humans and other non-target organisms (Dantas-Torres, 2008; Dantas-Torres et al., 2006). Several issues, importantly the abuse in the pesticide usage have

caused significant increase in the pesticide resistance, thus creating new alternative of chemical control is extremely difficult and expensive (World Health Organization, 2006).

Alternatively, the idea is to use the common and readily available tropical herbs in Malaysia as a new source of cheap, effective, and environmental friendly to combat this problem. The betel leaves, *Piperaceae betle* was selected as it has been proven traditionally to have anti-microbial, anti-oxidative, anti-haemolytic and also anti-parasitic properties (Pin *et al.*, 2006; Chakraborty & Shah, 2011; Wendy *et al.*, 2014; Jawale & Society, 2016; Syahidah *et al.*, 2017).

Thus, the main objective of this study is:

To assess the effects of the betel leaves aqueous and ethanol extracts on the *Rhipicephalus sanguineus* as pesticide.

The hypothesis for this study is:

Betel leaves extract has potential effect on the *Rhipicephalus sanguineus* as acaricides.

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