



UNIVERSITI PUTRA MALAYSIA

**RETROSPECTIVE OBSERVATION ON FILARIASIS
IN DOGS AND CATS PRESENTED TO UNIVERSITY VETERINARY
HOSPITAL AND VETERINARY LABORATORY SERVICE UNIT
OF UNIVERSITY PUTRA MALAYSIA FROM JANUARY 2016 TO AUGUST
2022**

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RETROSPECTIVE OBSERVATION ON FILARIASIS

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AND VETERINARY LABORATORY SERVICE UNIT

OF UNIVERSITY PUTRA MALAYSIA

FROM JANUARY 2016 TO AUGUST 2022

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It is hereby certificated that we have read this project paper entitled “Retrospective Observation on Filariasis in Dogs and Cats Presented to University Veterinary Hospital and Veterinary Laboratory Service Unit of University Putra Malaysia from January 2016 to August 2022”, by Michelle Yew Shyh-Xiao and in our opinion it is satisfactory in terms of scope, quality, and presentation as partial fulfilment on the requirement for the course VPD4999 – Final Year Project.

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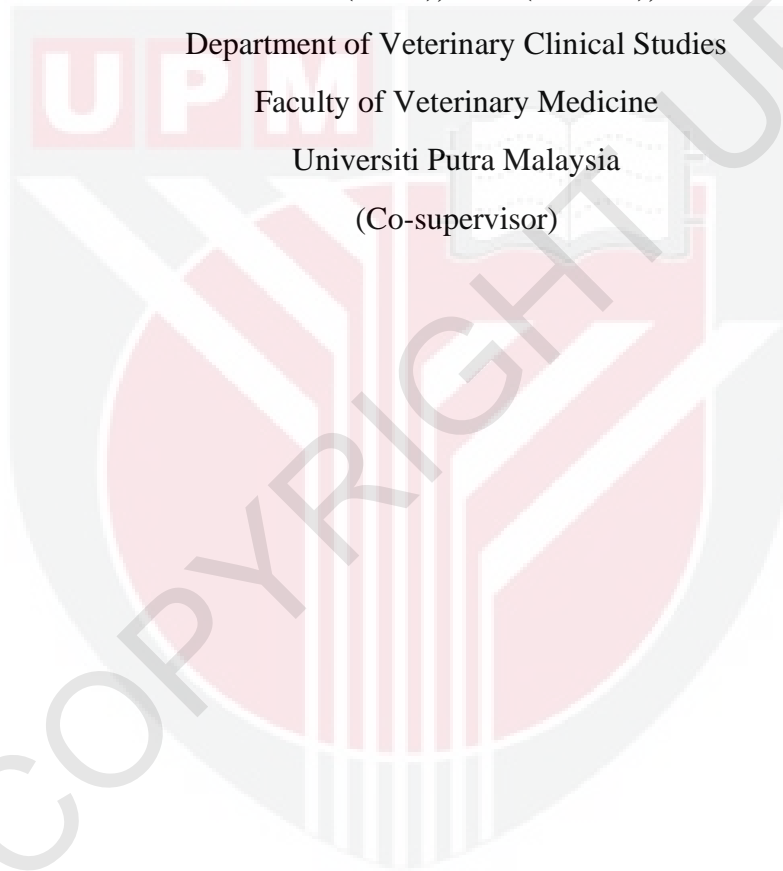
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ABSTRAK

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

**PERMERHATIAN RETROSPEKTIF FILARIASIS
DALAM ANJING DAN KUCING YANG DIKEMUKAKAN
KE HOSPITAL VETERINAR UNIVERSITI DAN
UNIT PERKHIDMATAN MAKMAL VETERINAR
UNIVERSITI PUTRA MALAYSIA DARI JANUARI 2016 HINGGA OGOS**

2022

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2022

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Filariasis disebabkan oleh parasit filaria patogenik yang mengakibatkan penyakit seperti penyakit cacing jantung dan filariasis limfa. Pendekatan yang betul perlu diambil untuk diagnosis dan merawat penyakit ini dengan tepat. Tujuan kajian ini adalah untuk melaporkan kejadian kes filariasis yang dikemukakan ke Hospital Veterinar Universiti, Universiti Putra Malaysia (HVU, UPM). Rekod dari Makmal Patologi Klinikal dan HVU dari Januari 2016 hingga Ogos 2022 diperolehi. Sebanyak 107 kes filariasis positif mikrofilaria ditemui, daripadanya 102 ekor anjing dan lima

ekor kucing. Spesies jangkitan filaria, maklumat pesakit, pengurusan, status pencegahan cacing jantung, tanda klinikal filariasis dan perubahan klinikopatologi diperoleh daripada rekod-rekod berikut. Keputusan kajian mendedahkan kejadian filariasis dan species yang ditemui adalah *Dirofilaria immitis* dan *Brugia spp.* Anjing dewasa pertengahan umur (4-9 tahun; 47.1%), baka tempatan (37.2%) dan anjing yang diurus di luar (43.7%) biasanya terjejas. Antara sampel anjing, 47.9% tidak diberi ubat cacing jantung. Tiada kucing menunjukkan sebarang tanda klinikal filariasis manakala 19.7% anjing menunjukkan tanda klinikal cacing jantung. Penemuan hematologi adalah anemia (36.3%) untuk anjing. Neutrofilia sederhana dengan anjakan ke kiri diperhatikan dalam 17.0% anjing manakala hanya seekor kucing mempunyai neutrofilia ringan dengan anjakan ke kiri, yang menunjukkan jangkitan atau keradangan. Hiperglobulinemia ditemui di kalangan 66.3% anjing dan tiga ekor kucing. Azotemia pra-renal, yang berkaitan dengan kemungkinan masalah jantung dilihat pada 34.0% anjing dan dua kucing. Kesimpulannya, maklumat ini berguna supaya doktor veterinar dapat menasihati pelanggan tentang kepentingan penjagaan pencegahan haiwan peliharaan.

Kata kunci: *filariasis; mikrofilaria; Dirofilaria immitis; Brugia spp.*

ABSTRACT

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

RETROSPECTIVE OBSERVATION ON FILARIASIS IN DOGS AND CATS**PRESENTED TO UNIVERSITY VETERINARY HOSPITAL AND****VETERINARY LABORATORY SERVICE UNIT OF****UNIVERSITI PUTRA MALAYSIA FROM JANUARY 2016 TO AUGUST****2022****by****MICHELLE YEW SHYH-XIAO****2022****Supervisor: Dr. Azalea Hani Othman****Co-Supervisor: Dr. Nor Azlina Abdul Aziz**

Filariasis is caused by pathogenic filarial parasite that results in diseases like heartworm disease and lymphatic filariasis. It is important to take the right approach to accurately diagnose and treat these diseases. The purpose of this study was to report the occurrence of filariasis cases presented at University Veterinary Hospital, Universiti Putra Malaysia (UVH, UPM). The records from Clinical Pathology Laboratory and UVH from January 2016 to August 2022 were obtained. A total of 107 microfilaria-positive filariasis cases were found, comprising of 102 dogs and five cats. The species of filarial infection, signalment, management, heartworm prevention status, filariasis related clinical signs and clinicopathological changes were obtained

from these records. Results revealed occurrence of filariasis, with the common species being *Dirofilaria immitis* and *Brugia* spp. Middle aged adult (4-9 years old; 47.1%), local breed (37.2%) and outdoor-managed dogs (43.7%) were commonly affected. Among dogs, 47.9% was not given any heartworm preventative. None of the cats showed any filariasis clinical signs while 19.7% of dogs showed common heartworm diseases clinical signs. The common haematological finding was anaemia (36.3%) for dogs. Moderate neutrophilia with left shift was observed in 17.0% of dogs while only one cat had mild neutrophilia with left shift, which indicates infection or inflammation. Hyperglobulinaemia was observed among 66.3% of dogs and three cats. Pre-renal azotaemia, relating to possible heart problem was seen in 34.0% of dogs and two cats. In conclusion, the information gathered would be useful for clinicians to advise clients on the importance of pet preventative care.

Keywords: *filariasis; microfilaria; Dirofilaria immitis; Brugia* spp.

1.0 INTRODUCTION

1.1 Background

Filariasis is the infection caused by the filarial nematode parasite. The two main genera of veterinary importance in Malaysia are *Dirofilaria immitis* and *Brugia* spp. which causes heartworm disease and lymphatic filariasis respectively. It is also important to note that these parasites have zoonotic potential. These parasites are transmitted to a susceptible animal host with the aid of the mosquito vector. The vector of these parasites favours the hot and humid tropical countries like Malaysia which aids in completing the lifecycle of the parasites (Jitsamai *et al.*, 2021). It is difficult for filariasis to be eradicated in Malaysia due to the persistence of the mosquito vectors and the consistent animal reservoirs, be it from the stray population or in pet animals that are not given filarial worm preventatives.

The adult filarial worms will produce microfilaria, which will be released into the bloodstream of the infected animal. The microfilaria circulates in the blood stream until a mosquito vector takes a blood meal from the infected animals. Then, the microfilaria will mature into its infective larvae, L3 form. When the infective mosquito bites a susceptible animal host, it releases the infective L3 into the bloodstream. The larvae will then be able to complete its life cycle and grow into adult worms to produce more microfilaria (Bowman, 2014).

The most common way for the detection of *Dirofilaria immitis* is via antigen detection using rapid test kits that are widely available in the market. However, these test kits come with some limitations such as only able to detect for antigen of adult female heartworms. The test kit is unable to test for adult male heartworms nor circulating microfilariae. Therefore, detection of heartworm disease using test kit

should be done adjunct to the screening of microfilaria in blood smears (Vezzani *et al.*, 2008). *Brugia spp.* on the other hand is diagnosed based on clinical signs such as lymph node enlargement as well as limb oedema and also detection of microfilaria in blood smear.

At present, there are data limited only to the prevalence of filariasis in the stray cat population in Malaysia. The *Brugia spp.* commonly infect cats however dogs may be affected too. Hence, a more accurate detection of filaria can aid in giving the proper advice to pet owners on the preventative measure to be taken, for both the pet plus the pet's environment and treatment, if ever warranted.

1.2 Objectives

The objectives of this study are to determine the occurrence of filariasis in dogs and cats diagnosed in Faculty of Veterinary Medicine (FVM), Universiti Putra Malaysia (UPM) and to determine filarial species of the microfilaria in blood smear of dogs and cats diagnosed with filariasis in FVM, UPM.

1.2.1 Justification

Currently there has not been any study done on the occurrence of filariasis and the common species of filarial worm that are found in dogs and cats in University Veterinary Hospital (UVH), UPM. Filariasis cannot always be diagnosed solely by looking at clinical signs and using rapid antigen test kit. There are limitations to the test kit for *Dirofilaria immitis* which only detects female worm antigen and not male worm antigen, nor microfilaria. Hence, further blood smear testing of the infected animal would be required to confirm the diagnosis of filariasis. However, even with

the presence of microfilaria in the blood sample, the species must be determined whether it is heartworm disease or lymphatic filariasis. This is because the treatments differ between both diseases. Heartworm disease requires systemic antiparasitic whereas lymphatic filariasis requires only topical antiparasitic treatment. Pet owners should also be educated on the importance of giving filariasis prevention to ensure that their pets do not get infected and consequently reduce and eradicate these diseases from the population.

1.3 Hypothesis

1. Occurrence of filariasis in dogs and cats diagnosed in UVH and VLSU, UPM.

H₀: There is no occurrence of filariasis in dogs and cats.

H_a: There is occurrence of filariasis in dogs and cats.

2. Filarial species of the microfilaria in blood smear of dogs and cats diagnosed with filariasis in UVH and VLSU, UPM.

H₀: *Dirofilaria immitis* and *Brugia* spp. are not the common species of filarial worm of dogs and cats diagnosed with filariasis respectively.

H_a: *Dirofilaria immitis* and *Brugia* spp. are the common species of filarial worm of dogs and cats diagnosed with filariasis respectively.

2.0 LITERATURE REVIEW

2.1 Introduction of Disease/Species

2.1.1 Heartworm Disease (*Dirofilaria immitis*)

An endoparasite called heartworm or also known as *Dirofilaria immitis* is found in canines and felines which causes pathological alterations in multiple organs such as the heart mainly the pulmonary arteries and the lungs (Hays *et al.*, 2020). The transmission of this disease is based solely on the presence of vectors, which in this case are mosquitoes to complete their lifecycle. These vectors would require optimum weather and environmental conditions to be able to survive and reproduce for the continual spread of this disease. This disease is more prevalent in countries that are hot and humid such as the tropics and subtropical areas.

When the mosquito vector takes a blood meal from an infected animal with circulating microfilaria, the microfilaria will mature into the infective L3 larvae stage in the Malpighian tubules of the mosquito itself. The mosquito will then take a blood meal from a susceptible animal and then deposit the L3 larvae simultaneously. Within a few months, the larvae in the newly infected animal will mature into sexually mature filarial worms in the right ventricles and pulmonary arteries (Morchón *et al.*, 2012). The matured filarial worms will then release microfilaria which will circulate in the bloodstream and when a mosquito takes a blood meal the whole cycle repeat all over again (Hays *et al.*, 2020).

2.1.2 Lymphatic filariasis (*Brugia* spp.)

Filarial worm of the genus *Brugia* causes the commonly overlooked tropical illness, lymphatic filariasis in cats, dogs and also humans (Jitsamai *et al.*, 2021). Dogs and mostly cats are the reservoir for lymphatic filariasis. However, when they are

infected, clinical signs are rarely observed (Tropical Council for Companion Animal Parasites, 2019). The L3 larvae is inseminated into a susceptible host when an infected mosquito takes a blood meal. It will travel to the lymph nodes through the lymphatic vessels where the filarial worms will mature into adults and produce microfilaria. Once the lymphatic vessels are obstructed the pathogenesis of the disease will progress. As the host's lymphatic system is continually invaded, so as the severity of the disease progression (Junhom *et al.*, 2006).

2.1.3 Subcutaneous filariasis (*Acanthocheilonema reconditum*)

A nonpathogenic canine filarial parasite that resides in the subcutaneous tissues is known as *Acanthocheilonema reconditum* and this parasite is found in locations that are endemic for *Dirofilaria immitis* or heartworm (Marchiondo *et al.*, 2019). These sexually matured adult female filarial worms produce numerous microfilaria in the subcutaneous layer of the infected dog which will then circulate around the bloodstream until it is taken up by the vector of this parasite. The growing or infective L3 larvae stage has been found in both fleas and lice. The vector playing the main role in the transmission of *Acanthocheilonema reconditum* is the cat flea, *Ctenocephalides felis*. However, it is still uncertain whether this parasite is transmitted via fleabite or direct ingestion parasite infested flea (Napoli *et al.*, 2014).

2.2 Clinical Signs

2.2.1 Heartworm disease

Majority of dogs with heartworm disease are asymptomatic meaning they do not show any clinical signs whether the parasite burden or infection duration is accounted for. Those dogs who show presenting clinical signs of heartworm disease

are usually those inflicted with immensely high worm burden or have existing complications due to heartworm. The duration of the infection and the severity of the disease plays a role in the clinical signs shown which normally coincides with the effect of the worms in the lungs and heart primarily the pulmonary arteries. The patients may come in with history of coughing, dyspnea, syncope, exercise intolerance, weight loss, lethargy, cyanosis, haemoptysis and ascites. Abnormal heart and lung sound such as cardiac murmur and crackles respectively can be heard upon physical examination of the dog (Hoch and Strickland, 2008). Caval syndrome can occur when a dog is heavily infected with heartworms and presents itself acutely. This syndrome can progress rapidly and is also fatal if prompt intervention is not carried out. Caval syndrome is a condition that occurs when a load of heartworm becomes lodge in the right atrium, right ventricles and also the vena cava which obstruct the normal blood flow in the heart (Jones, 2015).

Cats with heartworms are usually asymptomatic or only show symptoms for a short while. Feline heartworm disease clinical signs are usually just barely noticeable malaise or manifest in the gastrointestinal, respiratory or sometimes neurological systems. Cats can face per acute syndrome of this disease which shows respiratory signs, incoordination, seizure, collapse, haemoptysis and occasionally sudden death (American Heartworm Society, 2014).

2.2.2. Lymphatic filariasis

Cats are usually asymptomatic and usually handle the infection of *Brugia* spp. quite well. There have been little information of lymphedema nor lymphadenopathy occurring in infected cats (Tropical Council for Companion Animal Parasites, 2019).

Dogs are rarely infected with *Brugia* spp. and even if infected would mostly be asymptomatic. Genetics of dogs play a role in determining the infected dog's clinical outcome (Tropical Council for Companion Animal Parasites, 2019). However, in experimentally inoculated dogs, they displayed clinical signs similar to that of human such as limb edema, lymphangitis, episodic lymphadenopathy and lymph node enlargement. (Jitsamai *et al.*, 2021).

2.3 Diagnosis

2.3.1 Heartworm disease

The diagnosis of heartworm disease can be done by finding for *Dirofilaria immitis* microfilariae in blood samples by using various techniques such as direct wet mount, thin blood smear and Knott's concentration test. The species of the microfilaria is then further identified based on its morphology for locations with multiple known filarial nematodes (European Society of Dirofilariosis and Angiostrongylosis, 2017). However, this method is not very reliable as it can produce false-negative results due to amicrofilaraemia or occult infection, low number of circulating microfilaria and not sufficient examination of blood samples (Hoch and Strickland, 2008).

Another highly sensitive diagnostic technique to detect for heartworm is the detection of circulating heartworm antigen (European Society of Dirofilariosis and Angiostrongylosis, 2017). The antigens tests currently available in the market only test for antigen secreted by the adult female heartworms, regardless of other heartworm stages also producing certain amount of antigen (Little *et al.*, 2018). The antigen testing should only be carried out after seven months of age as it only detects female heartworm that have matured into the adult phase. It is highly advised to carry

out both microfilariae detection and antigen testing to obtain maximum diagnostic results (European Society of Dirofilariosis and Angiostrongylosis, 2017). The results of the above-mentioned tests should also be interpreted together with patient history, presenting clinical signs and other diagnostic methods (Rizzo and Ware, 1989). Other diagnostic methods include radiography, echocardiography and electrocardiography (Hoch and Strickland, 2008).

2.3.2. Lymphatic filariasis

Viewing for microfilariae in thin blood smear and wet blood mount using a light microscope is one of the ways to diagnose for *Brugia* spp. infection. By using the Knott's Concentration Technique, *Brugia* spp. is detected upon finding a sheathed microfilariae under a microscope, as compared to *Dirofilaria immitis* which do not possess a sheath (Figures A1 and A2). Enzyme-linked immunosorbent assay (ELISA) can also be used to confirm the diagnosis of lymphatic filariasis by the detection of its antigen or antibody produced by an infected host. Polymerase Chain reaction (PCR) can be useful to determine the species of filarial worm and also determine the filarial infection during low parasitaemia phase (Tropical Council of Companion Animals Parasites, 2019).

2.4 Prevention

2.4.1 Heartworm disease

Heartworm disease is very much preventable and is a much safer and effective approach than treatment (Rizzo and Ware, 1989). Clients should be exposed to the knowledge of the lifecycle and seasonality of the heartworm, relevant clinical signs and testing limitations. This would educate the client and show the importance of

heartworm prevention for their pets and treatment if ever warranted (Hoch and Strickland, 2008). Puppies should be started on heartworm prophylaxis before eight weeks of age and should be tested six months after the first dose, and then annually or biannually after that. In puppies and dogs older than seven months of age, they should carry out an antigen test and check for microfilaria before the initial heartworm prevention dose. This is to not disrupt the subclinical detection of heartworm infection detection and make as though the preventative do not work as intended (American Heartworm Society, 2018). Heartworm prophylaxis should be given once or twice a year depending on manufacturer's instructions and route of administration (Rizzo & Ware, 1989; American Heartworm Society, 2018).

Controlling of access of vector to susceptible dog also plays a role in the prevention of heartworms. Owners can use insect repellents or ectoparasiticides to deter mosquitoes from going near their pets. Limiting of pet's outdoor access during peak mosquito feeding time can also reduce the incidence of meeting with mosquito vectors (American Heartworm Society, 2018).

2.4.2 Lymphatic filariasis

Monthly heartworm preventatives such as moxidectin or selamectin spot-on has likely shown to be effective against lymphatic filariasis. Together with the usage of topical repellents and insecticides to keep the mosquito vectors away (Tropical Council of Companion Animals Parasites, 2019).

3.0 METHODOLOGY

3.1 Data Retrieval

Records of patients with confirmed filariasis were obtained from the Clinical Pathology Laboratory, VLSU, UPM from January 2016 to August 2022. Patients' information including lab reference and case number, patient ID, species, breed, sex, age were recorded. Among the records, cases with microfilaria were searched using the keywords "microfilaria" from comments of the findings from the blood result by the clinical pathologist on duty. The total number of confirmed filariasis by the detection of microfilaria in the blood smear obtained were 107 cases.

Based on the Clinical Pathology Laboratory, VLSU, UPM lab reference number, the blood smear archive slides were retrieved and observed under a microscope to identify the species of microfilaria based on the microfilaria's morphology. The findings of the requested haemogram and serum biochemistry tests were also recorded.

The case number obtained were then used to find for any records of these patients in the Parasitology Laboratory. The test methods done which comprises of direct wet mount and thin blood film with Giemsa stain and the findings of whether there was presence of microfilaria and their species were recorded.

The same case number was used to retrieve patient files from UVH archive file room. Additional patient's information such as management, heartworm prevention, relevant history and clinicals signs relating to heartworm disease or lymphatic filariasis were recorded. The total number of patient's files found from UVH archive were a total of 76 files and majority had incomplete patient's record.

3.2 Statistical Analysis

Data was recorded using Microsoft Excel and were subjected to descriptive analysis.

4.0 RESULTS

4.1 Filariasis Cases

The results illustrate that the number of filariasis cases from January 2016 to August 2022 were a total of 107 cases with 5 (4.7%) which were cats and 102 (95.3%) dogs (Table 1).

Table 1. Distribution of filariasis cases in dogs and cats.

	Total no. of cases	Percentage (%)
Cats	5	4.7
Dogs	102	95.3
Total	107	100

4.1.1 Filariasis Yearly Cases Trend

Table 2 shows the number of filariasis cases according to year with 2016 had the highest number of positive cases, while 2022 had the least number.

Table 2. Yearly trend of filariasis cases in dogs and cats

Year	No. of cases
2016	25
2017	23
2018	16
2019	22
2020	9
2021	10
2022	2
Total	107

4.2 Genus of Microfilaria Identified

The results showed that all five feline filariasis cases were infected with *Brugia* spp whereas for the canine filariasis cases, majority of the dogs were infected with *Dirofilaria immitis* while only 1% was co-infected with both *Brugia* spp. and *Dirofilaria immitis* (Table 3).

Table 3. Genus of microfilaria identified in dogs and cats.

Genus of microfilaria identified	No. of cases	
	Dogs	Cats
<i>Brugia</i> spp.	0	5 (100%)
<i>Dirofilaria immitis</i>	101 (99%)	0
Co-infection (<i>Brugia</i> spp. + <i>Dirofilaria immitis</i>)	1 (1%)	0
Total	102	6

4.3 Age

The age group that was the most affected with filariasis were four to nine years old, whereas the age group that was the least affected was one to three years old (Table 4).

For the feline filariasis, the age group that was affected most was the age group termed “adult” which means the age of the cat is not exactly known but they are within the adult feline age. (Table 4).

Table 4. Age distribution of filariasis in dogs and cats.

Age Group	No. of dogs	No. of cats
1-3 years old	7 (6.9%)	0
4-9 years old	48 (47.0%)	1 (20%)
>9 years old	42 (41.2%)	1 (20%)
Adult	5 (4.9%)	3 (60%)
Total	102	5

4.4 Breed

4.4.1 Dogs

The most affected dog breed with canine filariasis were local followed by cross-bred, Shih Tzu, Rottweiler, Doberman and German Shepherd (Table 5).

Table 5. Breed distribution of filariasis dogs.

Breed	No. of Dogs
Local	38 (37.2%)
Cross-bred	13 (12.7%)
Rottweiler	9 (8.8%)
Shih Tzu	9 (8.8%)
Doberman	5 (4.9%)
German Shepherd	4 (3.9%)
Miniature Pinscher	3 (2.9%)
Husky	3 (2.9%)
Terrier	3 (2.9%)
Bull Terrier	2 (2.0%)
Dachshund	2 (2.0%)
Dalmatian	2 (2.0%)
Pomeranian	2 (2.0%)
Beagle	1 (1.0%)
Cavalier King Charles Spaniel	1 (1.0%)
Cocker Spaniel	1 (1.0%)
Golden Retriever	1 (1.0%)
Labrador Retriever	1 (1.0%)
Pug	1 (1.0%)
Yorkshire Terrier	1 (1.0%)
Total	102

4.4.2 Cats

Among the five cat filariasis cases, one was cross-bred and two each were Domestic Longhair and Domestic Shorthair respectively.

4.5 Management

For canine filariasis, outdoor management were most affected whereas the least affected management is indoor-outdoor. Indoor-outdoor management means that the dog can walk in and out of the house as it pleases (Table 6).

Table 6. Management of filariasis dogs and cats.

Management	No. of cases	
	Dogs	Cats
Indoor	17 (23.9%)	1 (20%)
Outdoor	31 (43.7%)	2 (40%)
Indoor-outdoor	4 (5.6%)	2 (40%)
Not stated	19 (26.8%)	0
Total	71	5

4.6 Dog Heartworm Prevention Status

For canine filariasis, the heartworm prevention was mostly not given or not stated in the records (Table 7).

Table 7. Heartworm prevention status of filariasis dogs

Heartworm Prevention Status	No. of Dogs
Yes	0
Not Up to Date	3 (4.2%)
No	34 (47.9%)
Not stated	34 (47.9%)
Total	71

4.7 Filariasis Clinical Signs

For filariasis cases with filariasis clinical signs such as coughing, exercise intolerance, dyspnea, cyanosis, syncope, lethargy and ascites, there were 14 of dogs affected, while 57 cases of dogs and all the cats were asymptomatic or show no visible clinical signs (Table 8).

Table 8. Filariasis dogs and cats presented with clinical signs

Clinical Signs Present	No. of Dogs	No. of cats
Yes	14 (19.7%)	0
No	57 (80.3%)	5 (100%)
Total	71	5

4.8 Haemogram

4.8.1 Dogs

All 102 canine filariasis cases had haemogram requested. From those cases, 37 dogs were anaemic with 17, 11 and 9 having mild, moderate and marked anaemia respectively (Table 9).

Table 9. Haemogram of filariasis dogs

Anaemia	Regenerative	Non-regenerative	No Reticulocyte count	Total
Mild	8 (7.8%)	9 (8.8%)	0	17 (16.7%)
Moderate	4 (3.9%)	6 (5.9%)	1 (1.0%)	11 (10.8%)
Marked	7 (6.9%)	2 (2.0%)	0	9 (8.8%)
				37 (36.3%)

RBC= 5.5-8.5x10¹²L; PCV=0.35-0.55 L/L

4.8.2 Cats

For feline filariasis, all 5 cases had haemogram requested. From those cases, 1 (20%) cat was moderately anaemic and it was non-regenerative (Table 10).

Table 10. Hemogram of filariasis cats

Anaemia	Regenerative	Non-regenerative	Total
Mild	0	0	0
Moderate	0	1	1
Marked	0	0	0
Total	0	1 (20%)	1 (20%)

RBC= 5-10x10¹²L; PCV=0.24-0.45 L/L

4.9 Differential WBC Count

4.9.1 Dogs

Among the positive filariasis cases, 100 of them had requested for differential white blood cells (WBC) count (98%). From those cases, 36 had leucocytosis, 33 had monocytosis, 24 had eosinophilia and 25 had basophilia. Thirty-nine cases had increased in neutrophil count with moderate neutrophilia was the highest find (Table 11).

Table 11. Differential white blood cell counts of filariasis dogs

Differential WBC Count		Haematological Changes	
Total WBC (6.0-17.0x10⁹L)		Leucocytosis	
		36 (35.3%)	
Monocyte (0.2-1.4x10⁹L)		Monocytosis	
		33 (33.0%)	
Eosinophil (0.1-1.3x10⁹L)		Eosinophilia	
		24 (24.0%)	
Basophil (Rare)		Basophilia	
		25 (25.0%)	
<hr/>			
Neutrophil (3.0-11.5x10⁹L)			
Neutrophilia	No left shift	With left shift	Total
Mild	3 (3.0%)	8 (8.0%)	11 (11.0%)
Moderate	0	17 (17.0%)	17 (17.0%)
Marked	0	11 (11.0%)	11 (11.0%)
			39 (39.0%)

4.9.2 Cats

For feline filariasis cases, all had requested for differential WBC count. From those cases, one cat had leucocytosis, monocytosis and mild neutrophilia with left shift. Eosinophilia affected three out of five cats.

4.10 Serum Biochemistry

4.10.1 Dogs

For canine filariasis case, 97 out of 102 dogs tested for renal function parameters urea and creatinine (Table 12). Among them, 20.6% were severe azotaemia. The increased of creatinine only affected nine dogs with IRIS Stage 2 creatinine being the highest.

Table 12. Renal parameters of filariasis dogs.

Parameter	Biochemical Changes	Severity/IRIS Staging		
		Mild	Moderate	Severe
Urea (3.0-7.5 mmol/L)	Increased 33/97 (34.0%)	9 (9.3%)	4 (4.1%)	20 (20.6%)
Parameter	Biochemical Changes	Stage 2	Stage 3	Stage 4
Creatinine (88-176 µmol/L)	Increased 12/97 (12.4%)	8 (8.2%)	3 (3.1%)	1 (1.0%)

For liver and muscle enzyme parameters, 97 (95.1%), 70 (68.6), 20 (19.6%), 15 (14.7%) and 4 (3.9%) out of 102 canine filariasis cases requested for ALT, ALP, GGT, AST and CK respectively (Table 13). Majority of the cases had marked elevation of ALT and ALP, GGT and AST.

Table 13. Liver enzyme parameters of filariasis dogs.

Parameter	Biochemical Changes	Severity		
		Mild	Moderate	Severe
ALT (5-90 U/L)	Elevated 34/97 (35.1%)	8 (8.2%)	3 (3.1%)	23 (23.7%)
ALP (40-100 U/L)	Elevated 26/70 (37.1%)	3 (4.3%)	3 (4.3%)	20 (28.6%)
GGT (<6.0 U/L)	Elevated 10/20 (50.0%)	1 (5.0%)	1 (5.0%)	8 (40.0%)
AST (<60 U/L)	Elevated 6/15 (75.0%)	1 (6.7%)	1 (6.7%)	4 (26.7%)

In filariasis dogs with hyperproteinaemia, majority are mild response with hypoalbuminaemia and hyperglobulinaemia (Table 14).

Table 14. Total protein parameters of filariasis dogs

Parameter	Biochemical Changes	Total no. of cases		
		Mild	Moderate	Severe
Albumin (25-40 g/L)	Hypoalbuminaemia	31/95 (32.6%)		
	Hyperalbuminaemia	3/95 (3.2%)		
Globulin (25-45 g/L)	Hyperglobulinaemia 63/95 (66.3%)	35 (36.8%)	16 (16.8%)	12 (12.6%)

4.10.2 Cats

For feline filariasis case, four out of five cats tested for renal function parameters urea and creatinine. Two out of four that requested for urea measurement had moderate azotaemia. The IRIS Stage 2 creatinine only affected 1 cat (Table 15).

Table 15. Kidney parameters of filariasis cats.

Parameter	Biochemical Changes	Severity/IRIS Staging		
		Mild	Moderate	Severe
Urea (3.0-10.0 mmol/L)	Azotaemia 2/4 (50%)	0	2 (50%)	0
Parameter	Biochemical Changes	Stage 2	Stage 3	Stage 4
Creatinine (60-193 µmol/L)	Increased 1/4 (25%)	1 (25%)	0	0

For liver enzyme parameters, all five cats requested for ALT while four requested for ALP (Table 16). All cats had elevation in ALT with moderate increase being the highest at 40%.

Table 16. Liver enzyme parameters of filariasis cats.

Parameter	Biochemical Changes	Severity		
		Mild	Moderate	Severe
ALT (5-90 U/L)	Elevated 4/5 (80.0%)	1 (20%)	2 (40%)	1 (20%)

Hyperproteinaemia was a common finding in these cats with 3 out of 4 being affected (Table 17). Only one out of four cats had mild hypoalbuminaemia. Another common finding in cats with feline filariasis was hypoalbuminaemia with the majority being mildly affected.

Table 17. Total protein parameters of filariasis cats

Parameter	Biochemical Changes	Total no. of cases		
Albumin (25-40 g/L)	Hypoalbuminaemia	1 (25%)		
	Hyperalbuminaemia	0 (0%)		
Globulin (25-45 g/L)	Hyperglobulinaemia 3/4 (75%)	Mild	Moderate	Severe
		2 (50%)	1 (25%)	0 (0.0%)

5.0 DISCUSSION

In the present study, it was shown that all cats were infected with *Brugia* spp. In previous study, it was found that 35% (40/170) of cats were infected with *Brugia pahangi* from blood samples obtained in selected area of Selangor, Malaysia (Al-Abd et al., 2015). However, in dogs, it was shown that they were commonly infected with both *Dirofilaria immitis* and *Brugia* spp. In a study done in Chiang Mai, Thailand, it was shown that *Dirofilaria immitis* infection was found in 12.17% (41/337) of the community dogs and 8.31% (28/337) were *Brugia pahangi* infection (Kaikuntod et al., 2020). This supports the finding of this study that *Dirofilaria immitis* and *Brugia* spp. are the common filarial parasite found in both dogs and cats.

It was found that the highest percentage of dogs infected with filariasis were from the age of four to nine years old. Similar findings were also found by Vieira et al. (2014), in which dogs the same age group had a significant risk factor at 37.8% for heartworm. This shows that this age group have a higher risk of getting infected with heartworm.

Overall, the breed of dog that was found to be infected by heartworm the most were local breed. However, a study done previously showed that there was no significance of dog breed with the incidence of heartworm infection (Vieira et al., 2014). In present study, there may have been an over representation of local breed dogs presented to UVH and Clinical Pathology Laboratory, UPM.

The most common management of dogs with heartworm infection that were diagnosed was outdoor. In the study by Vieira et al. (2014), dogs living outdoors (37.7%) was one of the risk factors of developing heartworm infection.

Based on this retrospective study, the dogs with canine filariasis were mostly not given heartworm prevention. A previous study by Atkins (2014) states that 80.7% of dogs had gap in heartworm prevention. Another study by Ku (2016) illustrated that 30% of heartworm positive patients had no history of heartworm prevention. Heartworm prevention status is important to ensure protection of pets from heartworm disease.

Majority of the dogs did not display any clinical signs of dirofilariasis. This finding was supported by the study conducted by Vieira et al. (2014), that stated majority of the dogs infected with *Dirofilaria immitis* were asymptomatic. This shows that majority of the microfilariae found in blood smear were incidental findings during routine blood screening. Therefore, this highlights the importance of giving filariasis preventative care as without it whether the animal harbours this parasitic infestation unless tested.

Based on the haemogram results, dirofilariasis dogs experienced mild to moderate anaemia. Niwetpathomwat et al. (2007) showed similar findings where the microfilaraemic dogs had mild to moderate anaemia. This signifies that dirofilariasis causes anaemia to a certain degree.

The current study shows that dogs with filariasis had leucocytosis lymphopaenia, monocytosis, eosinophilia, basophilia and moderate neutrophilia with left shift. Similar findings were also found by Niwetpathomwat et al. (2007) which microfilaraemic dogs had leucocytosis, eosinophilia, monocytosis and moderate to marked neutrophilia was observed. These results shows that infection or inflammation occurs in dogs with circulating microfilariae.

The filariasis positive dogs had commonly displayed severe azotaemia. Similarly, to a study by Niwetpathomwat et al. (2007), there was a significant increase of blood urea nitrogen in the heartworm infected dogs. In this case, the azotaemia was mostly pre-renal which correlates with ongoing heart problems.

The severe elevation of ALT, ALP, GGT and AST liver enzyme were commonly seen among the tested dogs. There was a significant rise in ALT, ALP and AST of dirofilarial infected dogs (Rath et al., 2014). Since ALT is a liver specific enzyme in dogs, this indicated these filarial worms causes liver damage. The other liver enzyme ALP and AST that are not liver specific indicates that liver was not the only organ affected whereas other organs and tissues such as the kidney, muscle and others are affected as well.

There was decrease in albumin level in the current study with hypoalbuminaemia more commonly seen. However, a study done by Rath et al. (2014) shows hyperalbuminaemia instead. The hypoalbuminaemia in this study was likely due to microfilariae damage to both the liver and kidney, as supported by the kidney and liver parameters findings. However, this would require further investigation to confirm.

6.0 CONCLUSION AND RECOMMENDATIONS

The study allowed us to determine the occurrence of filariasis and the species of filarial worms of dogs and cats diagnosed in UVH and VLSU. The species of filarial worms found were *Dirofilaria immitis* and *Brugia* spp. Middle aged adult, local breed and outdoor managed dogs were the most affected. The dogs with heartworm infection were usually not given heartworm prevention or prevention was not up to date. Pets with filariasis were usually asymptomatic. The common haematological changes were mild anaemia, moderate neutrophilia, monocytosis, eosinophilia and basophilia. The common biochemical changes were pre-renal azotaemia, elevated ALT, hypoalbuminaemia and hyperglobulinaemia. With the information gathered, clinicians will be able to advise clients on the importance of preventative care for their beloved pets.

The main limitation in this study was acquiring complete patient medical records that include the preventative care of the patients and their management, whether they are kept indoor or outdoor.

It is recommended that future studies to include complete history mainly preventative prophylaxis and include other risk factors that were not assessed such as weather, temperature and seasonality which can affect the occurrence of filariasis. This would allow for a more accurate picture of the true occurrence of filariasis, identify various affecting risk factors and show the importance of client education.

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APPENDIX

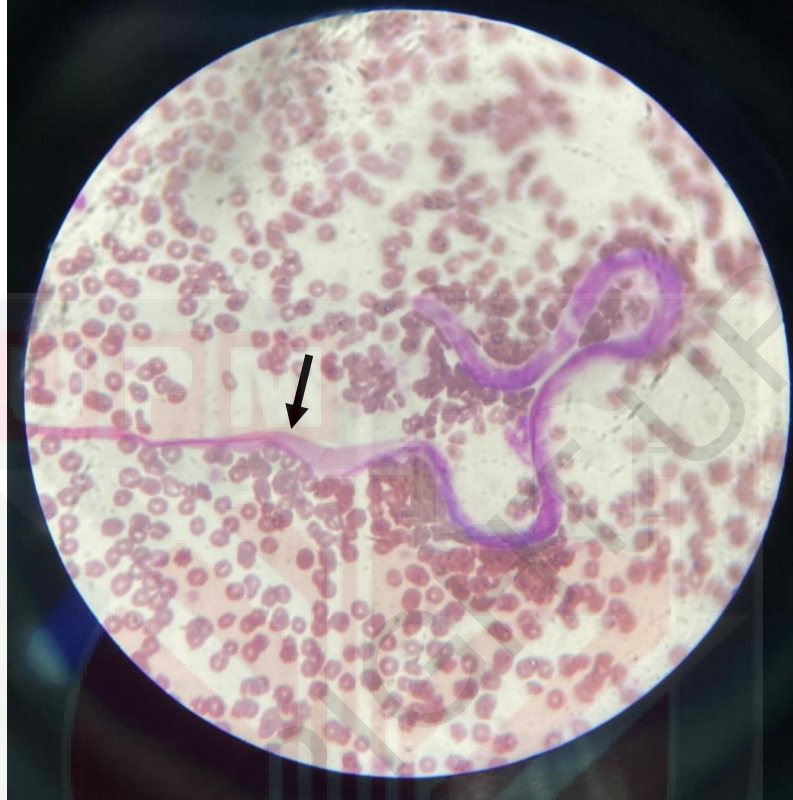


Figure A1. *Brugia* spp. Microfilaria. *Brugia* spp. microfilaria has a sheath at the microfilaria tail-end (black arrow). (1000x magnification).



Figure A2. *Dirofilaria immitis* microfilaria. *Dirofilaria immitis* microfilaria has no sheath at the microfilaria tail-end. (1000x magnification).