



UNIVERSITI PUTRA MALAYSIA

***MYCOBACTERIUM AVIUM SUBSPECIES PARATUBERCULOSIS
DETECTION IN BEEF CATTLE IN TAMAN PERTANIAN
UNIVERSITI, UPM***

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MYCOBACTERIUM AVIUM* SUBSPECIES *PARATUBERCULOSIS
DETECTION IN BEEF CATTLE IN TAMAN PERTANIAN UNIVERSITI,
UPM

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A project paper submitted to the
Faculty of Veterinary Medicine, Universiti Putra Malaysia
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DEGREE OF DOCTOR OF VETERINARY MEDICINE

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It is hereby certified that I have read this project paper entitled “*Mycobacterium avium* subspecies *paratuberculosis* detection in beef cattle in Taman Pertanian Universiti, UPM”, by Nur Farah Athirahbinti Ismail. In my opinion, it is satisfactory in terms of scope, quality and presentation as partial fulfilment of the requirement for the course VPD 4999- Project.

The logo of Universiti Putra Malaysia (UPM) is a shield-shaped emblem. It features a red and white striped background. At the top left, the letters 'UPM' are written in white on a red rectangular background. In the center, there is a white book with a red cover. Below the book, there are two white arrows pointing outwards. The entire logo is overlaid with a large, semi-transparent watermark that reads 'COPYRIGHT UPM'.

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DEDICATIONS

To the love of my life.....

Abah and Mama,

Mama, for giving me a life to live, and love me unconditionally.

Abah, for giving me support and make me tougher from inside.

I am somebody now.

Love you both.



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Thank you,

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ABSTRAK

Abstrakdaripadakertaskerja yang dikemukakan kepada Fakulti Perubatan Veterinar untuk memenuhi sebahagian daripada keperluan kursus VPD 4999 – Projek.

Jangkitan *Mycobacterium avium* subspesies *paratuberculosis* pada lembu pedaging di Taman Pertanian Universiti, Universiti Putra Malaysia

oleh

Nur Farah Athirah binti Ismail

2016

Penyelia : Profesor Dr. Abdul Aziz Saharee

Mycobacterium avium subspesies *paratuberculosis* (MAP)

adalah agen etiologi penyakit Johne yang dilihat sebagai penyakit bakteria usus kronik, menular dalam spesies ruminan. Penyakit yang

menyebabkan kerugian besar kepada penternak ini dicirikan oleh cirit-birit kronik, auselang-

seli, pengurangan berat badan progresif, dan penurunan pengeluaran.

Kajian ini dijalankan untuk menentukan kewujudan antigen dan antibodi MAP, masing-masing dalam tinjadan serum lembu. Sejumlah 213 sampel tinjadan 71 serum dikumpul daripada 71 ekor lembu di Taman Pertanian Universiti (TPU), Universiti Putra Malaysia (UPM). Sampel ini diuji menggunakan kaedah pewarna Ziehl-

Neelsenkekalasiduntukpengesanan antigen dankaedahujianpengikatankomplemen (CFT) untukantibodi. PewarnaanZiehl-Neelsenmenunjukkan60 (28.2%) daripada 213 sampelpositifuntuk antigen *M. avium*, manakala, CFT menunjukkan 3 (4.2%) daripada 71 sampelpositifuntukantibodi. Walaubagaimanapun, MAPbukanlahsatu-satunyaorganismakekalasid; justeru, penentuanspesiesorganismaperludisokongdengankulturtinja. Kesimpulannya, jangkitanMAPwujuddalamgerompoklembupedaging di TPU, UPM denganwujudnyaagenpenyebabdanantibodi, masing-masingdalamsampeltinjadan serum lembu.

Kata kunci :*Mycobacterium avium*subspesies*paratuberculosis*, jangkitan, cirit-biritselang-seli, CFT, pewarnaZiehl-Neelsenkekalasid.

ABSTRACT

An abstract of the project paper presented to the Faculty of Veterinary Medicine in partial fulfilment of the course VPD 4999 – Project.

***Mycobacterium avium* subspecies *paratuberculosis*(MAP) infection in beef cattle
in Taman PertanianUniversiti (TPU), UPM**

by

Nur Farah Athirahbinti Ismail

2016

Supervisor :Prof.Dr. Abdul Aziz bin Saharee

Mycobacterium avium subspecies *paratuberculosis*(MAP) is the aetiology agent causes Johne's disease which viewed as a chronic, contagious bacterial of intestinal tract in ruminant species. The infection that caused substantial losses to the farmer was characterized by chronic or intermittent diarrhoea, progressive weight loss, and decreased production. This study was conducted to determine the presence of MAP antigen and antibodies in the faeces and serum, respectively, in the cattle. A total of 213 faecal samples and 71 serum samples were collected from 71 cattle at Taman PertanianUniversiti (TPU), Universiti Putra Malaysia (UPM). These samples

were tested using Ziehl-Neelsen acid-fast stain method for antigen detection and complement fixation test (CFT) for antibodies detection. Ziehl-Neelsen staining revealed that 60 (28.2%) of 213 samples were positive for *M. avium* antigen while, CFT revealed 3 (4.2%) of 71 serum samples were positive for the antibody. However, MAP is not the only acid fast organism; thus the determination of species of the organisms should be supported by faecal culture. In conclusion, this study showed that there is MAP infection in the cattle herd at TPU, UPM, with the presence of antigen causing agent and antibodies in faecal and serum samples, respectively, of cattle.

Keywords : *Mycobacterium avium* subspecies *paratuberculosis*, bacterial disease, intermittent diarrhoea, CFT, Ziehl-Neelsen acid-fast stain

1.0 INTRODUCTION

Paratuberculosis or Johne's disease is a chronic contagious bacterial disease caused by causative organism called *Mycobacterium avium* subspecies *paratuberculosis* (MAP) which commonly affects domestic ruminants (cattle, sheep, goats and buffaloes) as well as wild ruminants (cervids) (Mercier, 2014). It was first observed by Drs Heinrich Albert Johne and Langdom Frothingham at Veterinary Pathology Unit, Dresden in 1895 (Manning and Collins, 2010). This organism causes granulomatous intestinal lesion and usually the disease is characterized by chronic or intermittent diarrhoea (Stabel, 1998), progressive weight loss, decreased production (milk and meat) and cause substantial losses (Hayton, 2007).

MAP can be transmitted and spreads by both horizontal and vertical means. It spreads through ingestion of MAP from contaminated environment (Mercier, 2014) most likely through faecal oral route, either by direct ingestion of faecal from infected animals or indirect ingestion of faecal contaminated colostrum, milk, water and feed (Manning and Collins, 2010). In addition, it can transmitted vertically from infected dam to foetus as conferred by Larson and Kopecky, 1970 and infection in calves is primarily due to ingestion of milk from infected dam or faecal contaminated milk (Mercier, 2014).

Once the MAP ingested, it survives and replicates within the macrophages in the intestine wall and the regional draining lymph nodes. Although subsequently phagocytised by macrophages, it will replicate slowly and stimulate inflammatory and cellular response as it is resistant to intracellular degradation (Hayton, 2007). The incubation period is long, up to 5 years. After this period, the animals will start to

shed these organisms in the faeces from low numbers and gradually will increase until the time of clinical onset although some will show intermittent shedding from the early course of the disease (Hayton, 2007), particularly the subclinical infection when the animals shed the organisms in the faeces while apparently looking normal with no clinical signs shown.

Clinical signs shown include intermittent to chronic diarrhoea, cachexia despite normal appetite while in advanced cases, they will show emaciation, lethargy, oedema and anaemia (Hayton, 2007). As the lesions start at the wall of intestine which will gradually develop to chronic granulomatous lesions thus causing protein leak and protein malabsorption syndrome leading to muscle wasting (Mercier, 2014). The economic impact of the disease in beef production is devastating particularly due to loss in production and treatment cost.

There are various types of detection methods available for diagnosing Johne's disease, either by detection of the antigen in the faecal or tissue or serologically. Common serological methods used include complement fixation test (CFT), absorbed enzyme-linked immunosorbent assay (ELISA) and agar gel immunodiffusion (AGID) although their sensitivity and specificity is often based on the result of faecal culture. All tests lack accuracy and have difficulty to detect the *MAP* in subclinical infected animal.

1.1 Rationale of study

This disease is important as it can cause serious chronic disease which can spread to other animals within the same farm for the bacteria will be shed in the environment by the affected animals although not showing any clinical signs. Besides, there is a lack of information regarding this disease in Malaysia. But there are several cases of Johne's disease reported intermittently in Selangor. The results and knowledge obtained from this study may serve as a future reference in knowing the prevalence and improving the health protocol in the prevention of Johne's disease at TPU.

1.2 Hypothesis and objectives of the study

There were presence of the *Mycobacterium avium* subspecies *paratuberculosis* (MAP) agent and antibody in faecal and serum tested respectively.

The prevalence of MAP infection in beef cattle at TPU is unknown, thus the objectives of this study are :

1. To determine the presence of *Mycobacterium avium subsp. paratuberculosis* (MAP) in the faecal of beef cattle at TPU, UPM.
2. To determine the presence of antibody against *Mycobacterium avium subsp. paratuberculosis* (MAP) in the serum of beef cattle at TPU, UPM.

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