



UNIVERSITI
TEKNOLOGI
MARA

**IDENTIFICATION AND CHARACTERIZATION OF *Enterocytozoon bienersi*
ISOLATES FROM THE PROTO-MALAY AND SENOI TRIBES IN SUNGAI
LEMBING, PAHANG**

By

AZAH ASHIKIN BINTI AZHARI

Thesis Submitted in Partial Fulfillment of the Requirements for

Bachelor of Medical Laboratory Technology (Hons),

Faculty of Health Sciences, Universiti Teknologi MARA

2016

ACKNOWLEDGEMENT

First and above all, I praise God, the Almighty for His blessings for giving me this opportunity and permitting me the ability to precede successfully this study to be very smooth and achieved what was planned. Foremost, I would like to express my sincere gratitude to my supervisor Dr. Tengku Shahrul Anuar bin Tengku Ahmad Basri for his constantly provide a big support and useful guidance to train me in the scientific field. Thank you for all the continuous advice and encouragement throughout the course has been given.

I acknowledge my gratitude to Dr Nazrina Bt Camalxaman as a Final Year Project coordinator in Medical Laboratory Technology Department for giving fully responsible in coordinating, handling and planning all of stuffs related in this project. Besides that, do not forget to all of my lecturers in Medical Laboratory Department who helped directly or indirectly in the success of this study. To all the staff in the Medical Laboratory Department, thank you for your help and support over the last six months.

My very sincere thanks to all the members of my research group: Nabilah Amelia, Juwairiyah, Munirah and Nurul Azmiera who helped starting from preparing of this research until complete of my research. I hope that all of my partners continue to be successful in facing life's journey.

Lastly, I take this opportunity to express the insightful gratitude from bottom of my heart to my beloved parents and my siblings for their love and continuous support-both spiritually and materially throughout my life. They have never questioned whatever crazy adventures I chose to go on.

TABLE OF CONTENTS

	Page
TITLE PAGE	
DECLARATION	ii
INTELLECTUAL PROPERTIES	iii
ACKNOWLEDGEMENTS	vi
TABLE OF CONTENTS	vii
LIST OF TABLES	ix
LIST OF FIGURES	x
LIST OF ABBREVIATIONS	xi
ABSTRACT	xii
CHAPTER	
1 INTRODUCTION	1
1.1 Objective of the Study	3
1.1.1 General Objective	3
1.1.2 Specific Objective	4
1.2 Hypothesis	4
2 LITRATURE REVIEW	5
2.1 Introduction	5
2.2 Classification and Taxonomy	8
2.3 Morphology of genera and Species infecting human	9
2.3.1 <i>Encephalitozoon</i> species	9
2.3.2 <i>Enterocytozoon</i> species	10
2.3.3 <i>Nosema</i> species	11
2.3.4 <i>Pleistophora</i> species	11
2.3.5 <i>Microsporidium</i> species	11
2.4 Biology of microsporidia	12
2.5 Life cycle of microsporidia	15
2.6 Geographical distribution and Prevalence in Human	17
2.7 Sources of human infection	19
2.7.1 Environmental source	19
2.7.2 Animal source	19
2.8 Modes of transmission	20
2.8.1 Inhalation transmission	20
2.8.2 Zoonotic transmission	20
2.8.3 Human (horizontal transmission)	21
2.8.4 Water-borne transmission	22
2.8.5 Food-borne transmission	23
2.8.6 Trans-placental transmission	23
2.8.7 Sexual transmission	23
2.8.8 Vector-borne transmission	24
2.9 Laboratory diagnosis for microsporidia	24
2.9.1 Staining for light microscopy	24
2.9.2 Electron microscopy	25
2.9.3 Molecular method	26
2.9.4 Indirect immunofluorescence	26

ABSTRACT

Enterocytozoon bieneusi is most common of microsporidian species that infects many types of cells including human and animals. These organisms are causing long lasting diarrhea primarily in immunocompromised patients but microsporidiosis is also higher in immunocompetent individuals. The previous study of microsporidial infection is mostly focused on immunodeficiency or immunosuppressive individuals. Information regarding microsporidial infections among immunocompetent individuals is lacking. Hence, this cross sectional study was carried out to determine prevalence of *E. bieneusi* infection and to find out the occurrence of *E. bieneusi* according to age group and gender among the Proto-Malay and Senoi tribes in Sungai Lembing, Pahang, Malaysia. Stool samples were collected from 209 voluntary participants; 96 males and 113 females that aged between one to 85 years old. Demographic data, socioeconomic, environmental, and behavioral information was collected by using pre-tested questionnaire. Overall prevalence of *E. bieneusi* infection was 3.83 % (8/209). From the observation, majority of individual infected with *E. bieneusi* was male (8.33 %) can be observed in those aged ≥ 15 years old. These samples were detected Small Subunit rDNA gene by using Polymerase chain reaction amplification with specific primer EBIEF1 and EBIER1, as demonstrated by an amplicon of 607-bp. This is the first report of *E. bieneusi* in Malaysia. It is hoped that these findings will allow greater emphasis from public health authorities through public education on personal hygiene and the consumption of boiled or filtered water in order to reduce the infection.

CHAPTER 1

INTRODUCTION

Microsporidia are known as single celled (Gill, 2007) which able to infect all major groups of animal kingdoms, including invertebrates, fish, amphibians, reptiles, birds and mammals (Wittner & Weiss, 1999). This organism is obligate intracellular protozoa which poison to many types of cell (Didier *et al.*, 2000, 2004; Weber *et al.*, 2000). A study by Lono *et al* (2010) found microsporidia have been reported in HIV patients, which recognized as opportunistic infection because they are involved as agents of human disease, especially in immunosuppressed individuals such as organ transplantation and chemotherapy recipients. Microsporidiosis is also can found in immunocompromised population and produce systemic and non-systemic disease (Garcia *et al.*, 1994; Kotler & Orenstein, 1998). The HIV positive patient population is more susceptible to intestinal parasitic infection than others patient populations, it has been estimated that 7-50% of chronic diarrhea in AIDS patients is associated with microsporidia. There is a wide spectrum of clinical manifest microsporidiosis including arthritis, prostatic abscess, tongue ulcer, bone infection, cutaneous infection, gastrointestinal and ocular infections (Franzen & Muller, 2001). The common clinical features of microsporidia infection usually produce persistent diarrhea and weight loss which associated with AIDS. In addition, AIDS patient is the main risk of microsporidia, but have another increasingly reported such as in organ transplant recipient, travellers, the chronically ill, the elderly, children and healthy people. Furthermore, this infection can lead to death (Weber *et al.*, 1997).

There are eight genera of microsporidia which is ability to infect human. From eight different genera of these organisms, majority species causes of human infection are either *Enterocytozoon bieneusi* or