## A PROBLEM ON TROPICAL DISEASES AMONG IMMIGRANT WORKERS IN SAMUT SAKHON HOSPITAL

### MUHAMMAD LUTHFI AL MANFALUTHI

A THEMATIC PAPER SUBMITTED IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR
THE DEGREE OF MASTER OF CLINICAL TROPICAL MEDICINE
FACULTY OF GRADUATE STUDIES
MAHIDOL UNIVERSITY
2013

**COPYRIGHT OF MAHIDOL UNIVERSITY** 

# Thematic paper entitled

# A PROBLEM ON TROPICAL DISEASES AMONG IMMIGRANT WORKERS IN SAMUT SAKHON HOSPITAL

	Mr. Muhammad Luthfi Al Manfaluthi Candidate
	Lect. Prakaykaew Charunwatthana, M.D., Ph.D. (Clinical Tropical Medicine) Major advisor
	Assist. Prof. Dorn Watthanakulpanich, M.D., Ph.D in Biomolecular Sciences Co-advisor
Assoc. Prof. Paron Dekumyoy, B.Sc. (Biol.), M.Sc. (Trop.Med.), Ph.D. (Trop.Med.) Co-advisor	Lect. Viravarn Luvira, M.D., Dip. Thai Board of Infectious Disease Co-advisor
Mr. Ekaporn Bhunnachet, M.D., Dip. Thai Board of Pediatircs Co-advisor	Lect. Saranath Lawpoolsri Niyom, M.D., Ph.D. in Epidemiology Co-advisor
Prof. Banchong Mahaisavariya, M.D., Dip. Thai Board of Orthopedics Ph.D. Dean Faculty of Graduate Studies Mahidol University	Dr. Wirongrong Chierakul, M.D., Dip. Thai Board of Internal Medicine,  Program Director Master of Clinical Tropical Medicine Faculty of Tropical Medicine Mahidol University

# Thematic paper entitled

# A PROBLEM ON TROPICAL DISEASES AMONG IMMIGRANT WORKERS IN SAMUT SAKHON HOSPITAL

was submitted to the Faculty of Graduate Studies, Mahidol University for the Degree of Master of Clinical Tropical Medicine

November 4, 2013

	Mr. Muhammad Luthfi Al Manfaluthi Candidate
	Mr. Molee Wanichsuwan, M.D., Dip. Thai Board of Dermatologist Chair
Assoc. Prof. Paron Dekumyoy, B.Sc. (Biol.), M.Sc. (Trop.Med.), Ph.D. (Trop.Med.) Member	Lect. Prakaykaew Charunwatthana, M.D. Ph.D. (Clinical Tropical Medicine) Member
Lect. Saranath Lawpoolsri Niyom, M.D., Ph.D. in Epidemiology Member	Assist. Prof. Dorn Watthanakulpanich, M.D., Ph.D in Biomolecular Sciences Member
Mr. Ekaporn Bhunnachet, M.D., Dip. Thai Board of Pediatircs Member	Lect. Viravarn Luvira, M.D., Dip. Thai Board of Infectious Disease Member
Prof. Banchong Mahaisavariya, M.D., Dip. Thai Board of Orthopedics Dean Faculty of Graduate Studies Mahidol University	Assoc. Prof. Yaowalark Sukthana, M.D., D.V.M., D.T.M.&H., M.C.T.M., Dip. Thai Board of Oto-Rhino-Laryngology Dean Faculty of Tropical Medicine Mahidol University

#### **ACKNOWLEDGEMENTS**

I would like to thank my major advisor, Lect. Prakaykaew Charunwatthana, and my co-advisors, Lect. Viravarn Luvira, Asst. Prof. Dorn Watthanakulpanich, Lect. Saranath Lawpoolsri, Assoc. Prof. Paron Dekumyoy, Dr. Molee Wanichsuwan as external examiner for their valuable advice and guidance in this study.

I wish to thank Dr. Molee Wanichsuwan, Dr. Ekaporn Bhunnachet and staff of immigrant clinic at Samut Sakhon Hospital for supporting me during data collection process.

I wish to extend my gratitude to all the lecturers for sharing with us their valuable knowledge and experience and to all staff of the Department of Tropical Medicine for their kind assistance.

Finally, I am very grateful to my family, especially my parents, for their endless love and care, my lovely little sisters and brother. I dedicate this thematic paper to "Persyarikatan Muhammadiyah".

Muhammad Luthfi Almanfaluthi

A PROBLEM ON TROPICAL DISEASES AMONG IMMIGRANT WORKERS IN SAMUT SAKHON HOSPITAL

MUHAMMAD LUTHFI AL MANFALUTHI 5538664 TMCT/M

M.C.T.M.

**PAPER** ADVISORY THEMATIC COMMITTEE: **PRAKAYKAEW** CHARUNWATTHANA, M.D., Ph.D. (CLINICAL TROPICAL MEDICINE), VIRAVARN LUVIRA, M.D., DIP. THAI BOARD OF INFECTIOUS DISEASE, WATTHANAKULPANICH, **DORN** M.D., Ph.D. (BIOMOLECULAR SCIENCES), SARANATH LAWPOOLSRI, M.D., Ph.D. (EPIDEMIOLOGY), PARON DEKUMYOY, B.Sc. (Biol.)., M.Sc. (TROPICAL MEDICINE), Ph.D. (TROPICAL MEDICINE), EKAPORN BHUNNACHET, M.D., DIP. THAI BOARD OF PEDIATRICS.

#### **ABSTRACT**

The objective of this prospective study was to investigate the prevalence of intestinal parasitic infections, including helminthic-protozoa and other tropical diseases (tuberculosis, malaria, filariasis, and syphilis), among adult male and female immigrant workers visiting the immigrant clinic in Samut Sakhon Hospital, Thailand. 198 participants fulfilled our study criteria and took part in the study. Our findings revealed that the prevalence of intestinal parasitic diseases among this group was 4.5%. Of these, 55% were male, 78% belonged to the younger group, 89% had been in Thailand for a longer working period, and 100% had single parasitic infections. The majority of infections (67%) were protozoan. These results suggest that regular screening and appropriate medication for the infected, combined with mass-education and prevention-promotion programs are the best ways to control the transmission of disease from immigrant workers to the Thai population.

KEY WORDS: TROPICAL DISEASES/IMMIGRANT/PREVALENCE

57 pages

# **CONTENTS**

	Page
ACKNOWLEDGEMENTS	iii
ABSTRACT	iv
LIST OF TABLES	vii
LIST OF FIGURES	viii
LIST OF ABBREVIATIONS	ix
CHAPTER I INTRODUCTION	1
CHAPTER II OBJECTIVES	2
CHAPTER III LITERATURE REVIEW	3
3.1 Migration and development in Thailand	3
3.2 Samut Sakhon Province	5
3.3 Tropical Diseases and Tropical Medicine	8
3.4 Infectious diseases in immigrant workers in Thailand	15
CHAPTER IV MATERIALS AND METHODS	20
4.1 Design, period and place of study	20
4.2 Study population	20
4.3 Inclusion criteria	20
4.4 Exclusion criteria: None	20
4.5 Methods	21
4.6 Data analyses	23
4.7 Sample size calculation	24
CHAPTER V RESULTS	25
5.1 Immigrant workers demographic data	25
5.2 Physical examination data	25
5.3 Laboratory examination data	26
5.4 Intestinal parasites and Others Tropical diseases	27

# **CONTENTS** (cont.)

	Page
CHAPTER VI DISCUSSION	35
CHAPTER VII CONCLUSION	41
REFERENCES	42
APPENDICES	48
Appendix A Definitions	49
Appendix B Thick Blood Smears For Malaria Infection And Filariasis	50
Appendix C Rectal Swab Examination For Parasite Infection	52
Appendix D Case Record Form	55
BIOGRAPHY	57

# LIST OF TABLES

Table		Page
3.1	Estimated foreign population residing and working in Thailand,	4
	approximately end of 2010	
3.2	Total population compared to number of immigrant workers in 2004-	7
	2007 Health Screening	
3.3	Prevalence of intestinal parasites found in 215 Myanmar workers in	16
	Bangkok and Samut Sakhon	
3.4	Prevalence of Plasmodium infections among foreign migrant workers	17
5.1	Vital sign result	30
5.2	Physical exam result	31
5.3	Laboratory examination result	32
5.4a	Prevalence if intestinal parasites found in immigrant workers (1)	33
5.4b	Prevalence if intestinal parasites found in immigrant workers (2)	33

# LIST OF FIGURES

Figure		Page
3.1	Location site of study, Bangkok is the capital city of Thailand.	6
	Samut Sakhon is about 30 km southwest of Bangkok	
3.2	Investigation Report Form for registered immigrant workers in	8
	migrant clinic Samut Sakhon hospital	
3.3	malaria education campaign proposed by Sir Patrick Manson	14
3.4	Age-related seroprevalence of hepatitis A virus among Thai	18
	population residing near Myanmar border (v), general Thai	
	population ( $\lambda$ ), and Myanmar immigrant workers ( $\blacktriangle$ )	
3.5	The microfilaria rates of Myanmar migrants classified by age group	19
5.1	Enrolment of patient	28
5.2	Participant's gender group	29
5.3	Participant's age group (bar chart)	29
5.4	Participant's duration of working	30
5.5	BMI for gender group (Box Plot)	31
5.6	Intestinal parasitic infection between gender and BMI group	34
5.7	Age distribution among intestinal parasitic infection immigrant	34

# LIST OF ABBREVIATIONS

Ab	breviation of Symbol	Term
	AFB	Acid fast bacilli
	BCG	Bacillus Calmette-guerrin
	ВРНРР	Bureau of Public Health Policy and Plan
	CNS	Central nervous system
	CSF	Cerebro spinal fluid
	CRF	Case record form
	EDTA	Ethylenediaminetetraacetic acid
	EIA	Enzym immune assay
	ELISA	Enzyme linked immunosorbent assay
	FTA-abs	Treponema pallidum antigen – flourosences
		treponemal antibody absorbsion test
	GDP	Gross Domestic Product
	GNI	Gross National Income
	IFA	Immuno fluorescence assay
	Ig	Immunoglobulin
	TB	Tuberculosis
	MOI	Ministry of Interior
	MOL	Ministry of Labor
	O&P	Ova and Parasites
	ORS	Oral rehydration solution
	PCR	Polimerase chain reaction
	RBC	Red blood cell
	RDT	Rapid diagnostic test
	STD	Sexual transmitted disease

# LIST OF ABBREVIATIONS (cont.)

Abbreviation of Symbol	Term
QBC	Quantitative buffy coat
TB	Tuberculosis
ТРНА	Treponemal palidum haemaglutination
TST	Tuberculin skin test
VDRL	Venereal disease research laboratory
WB	Western blot
WBC	White blood cell
WHO	World Health Organization

# CHAPTER I INTRODUCTION

The economic growth of Thailand has attracted a large number of people from the neighboring countries for seeking better life. Most immigrants are exposed to poor working and living conditions as they are hired in unskilled labor sectors, which tend to be dirty, difficult and dangerous. Socioeconomic and political factors tend to limit their accessibility to health services, making them vulnerable to poor health.

According to Wong et al (2003) immigrants played a role in disease transmission. There had been a number of tropical diseases associated with immigrant workers including TB, leprosy, syphilis, typhoid, filariasis, malaria and intestinal parasitic infections abundant in many urban centres. (Lam et al., 1998).

Samut Sakhon province is one of the crowded destinations for Myanmar immigrants. Authorized local health has made regulation to screening every immigrants before giving working permit in order to maintain the policy of Thai public health disease control program. In 2008, Samut Sakhon was declared as high risk transmission area for parasitic diseases with infectious prevalence as 62.3% reported by Nuchprayoon et al (2009). The latest study was conducted by Ngrenngarmlert et al (2012) indicated declining trends as 13.6%.

As there is scanty information about underlying diseases among immigrant workers. There is a need to obtain the prevalence of diseases in order to better understand the health status related with tropical diseases in that area. Rectal swab examination was performed and added test to evaluate the intestinal parasitic infections out of the routine screening test at the immigrant clinic. (Lam et al., 1998).

# CHAPTER II OBJECTIVES

## 2.1 Primary Objective

To obtain the prevalence of intestinal parasitic infections among adult immigrant workers at immigrant clinic in Samut Sakhon hospital

## 2.2 Secondary Objectives

To obtain the prevalence of tropical diseases among adult immigrant workers at immigrant clinic in Samut Sakhon hospital, such as: tuberculosis, syphilis, malaria and filariasis

To assess the association between intestinal parasitic infections and other tropical diseases.

# CHAPTER III LITERATURE REVIEW

## 3.1 Migration and development in Thailand

The economic growth of Thailand has resulted in marked improvements in a number of economic and social indicators. In 2009, the Gross National Income (GNI) Per-capita equaled USD 3,760 in current US dollars (WHO, 2011). With a GDP that accounts for 91% of the entire Mekong sub-region and its social stability has attracted a large number of people from the neighboring countries such as Laos, Myanmar, and Cambodia for seeking job opportunities and better income. Currently, it is estimated that more than 2 million immigrants are living in Thailand while only 30% of them entered with an official permission (Martin, 2004).

Thailand has been trying to manage labor immigrants from the three neighboring countries since more than ten years. Immense progress has been made. However, still a lot of tasks are left to be tackled due to the ongoing economic disparities with its neighbors, the rapid change of migration patterns and trends and the readiness of the origin countries in sending their labors on the regular mode.

**Table 3.1:** estimated foreign population residing and working in Thailand, approximately end of 2010

Category	Stay	Stay and work
Professional, skilled and semi-skilled workers**		
<ul> <li>Foreigners with work permits</li> </ul>		100,338
Diplomats and officials		6,148
Subtotal		106,486
Other temporary stay <sup>a</sup>		
Stay with Thais	14,946	
Stay with Thai wife	11,381	
Stay with resident families	1,098	
Retirement	28,509	1
<ul> <li>Others (including medical treatment and study)</li> </ul>	65,175	
Subtotal	121,109	
Tourist and transit visa extension and visa changes*	92,014	
Subtotal	92,014	
Students	- 2	
Higher education (2010)	19,052	
Subtotal	19,052	
Other regulars		
Residents awaiting nationality		233,811
Born in Thailand to non-national parents		69,799
Previously undocumented persons		210,182
Subtotal		513,792
Undocumented expatriates		
<ul> <li>Persons overstaying visas, 2007<sup>a</sup></li> </ul>		65,558
Subtotal		65,558
Refugees and asylum-seekers*		
Registered in official camps (December 2010)	95,330	
Unregistered and other categories	45,746	
Subtotal	141,076	
Migrants from Cambodia, Lao PDR and Myanmar		
Regular new entrants under MOU (end 2010) <sup>y</sup>		78,686
<ul> <li>Entered or completed NV process (end 2010)<sup>r</sup></li> </ul>		932,255
Unregistered and family members <sup>c</sup>		1,444,803
Subtotal		2,455,744
Total	373,251	3,141,580
Overall total	3,51	4,831

PUNESCO, table 18, International flows of mobile students at the tertiary level, <a href="http://stats.uis.unesco.org/Table/liewer.aspx">http://stats.uis.unesco.org/Table/liewer.aspx</a>, accessed on 8 August 2022

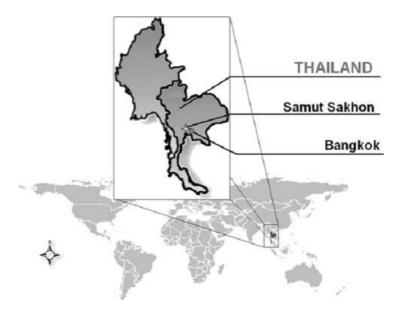
The policy to regularize immigrants, which capturing the highest number of immigrants than ever, was put into practice in 2010. All immigrants from the three countries were encouraged to register with the Ministry of Interior (MOI) prior to applying for a work permit with the Ministry of Labor (MOL). From subtotal 2,455,744 immigrants from Cambodia, Lao PDR and Myanmar, it is noted 1,444,803 unregistered.

Most immigrants are exposed to poor working and daily living conditions as they are hired in unskilled labor sectors which tend to be dirty, difficult and dangerous. Socioeconomic and political factors tend to limit their accessibility to health services; making them vulnerable to poor health, thus ensuring access to health service among immigrants is required to minimize the negative health impact on individuals and communities as a whole.

Immigrants play a role in disease transmission (Wong et al., 2003). There have been a number of disease associated with immigrant There have been a number of tropical disease associated with immigrant workers including TB, leprosy, syphilis, typhoid, filariasis, malaria are abundant in many of these urban centre (Lam, 1998).

#### 3.2 Samut Sakhon Province

Samut Sakhon is the third highest income/per capita province of Thailand 524,956 THB/capita after Rayong Province and Chonburi Province as high as 1,225,700 THB/capita and 544,160 THB/capita, respectively in 2013. It is located at the lower part of the Central Region of Thailand and approximately 30 kilometres south of Bangkok. The province occupies a total area of 872 square kilometers and is administratively divided into three districts: Muang, Krathum Baen and Ban Phaeo.



**Figure 3.1:** Location site of study, Bangkok is the capital city of Thailand. Samut Sakhon is about 30 km southwest of Bangkok

Having more than 5,000 factories related to fishery with various sizes in place, Samut sakhon has attracted a large number of immigrants. In 2007, Ministry of Inferior's Citizen Registration records from total population in Samut Sakhon Province 442,687, total registered migrant in 2007 is 74,531 (report from Ministry of Labour).

**Table 3.2:** Total population compared to number of immigrant workers in 2004 - 2007 Health Screening

		Total	Number of migrants <sup>2</sup>			
Province	Year	Total population <sup>1</sup>	Registered	Demand for migrant workers		
Samut Sakhon	2004	442,687	78,794	137,080		
	2005	452,017	73,896	184,960		
	2006	462,510	50,713	201,497		
	2007	469,934	74,531	250,000 <sup>3</sup>		
Kanchanaburi	2004	810,265	14,743	39,554		
-	2005	826,169	12,226	37,172		
	2006	834,447	9,141	37,172		
112	2007	835,282	7,551	110,296 <sup>3</sup>		
Tak	2004	515,877	52,768	96,874		
	2005	522,197	41,242	107,264		
	2006	527,677	27,248	143,012		
	2007	530,928	26,912	300,000 <sup>3</sup>		
Ranong	2004	176,372	31,962	49,704		
3.3866323 <del>5</del> 67	2005	178,122	23,312	45,690		
	2006	179,850	19,099	48,517		
	2007	180,787	17,809	68,199 <sup>3</sup>		

So urce:

- 1 Total population figures sourced from the annual Ministry of Interior's Citizen Registration.
- 2 Total numbers of registered migrants in 2004 2007 sourced from the Ministry of Labour

All the immigrant workers were passed the process of health screening program that including history taking, physical examination (Figure 3) and investigations (i.e., blood examination, chest X- ray, rectal swab culture, etc.). The list of diseases that need to be investigated for registered immigrant workers at immigrant clinic, Samut Sakorn hospital are

- 1) Malaria
- 2) Syphilis
- 3) Filariasis
- 4) Tuberculosis
- 5) Leprosy
- 6) Drug abuse
- 7) Pregnancy test for female
- 8) Rectal Swab Culture (RSC): only marine food preparation company

<sup>3</sup> Total number of migrants - estimated from surveys, or approximate figures calculated by the studied province from Provinical Employment Office

The results of diseases from screening program at immigrant clinic in Samut Sakhon hospital since 2005-2012 are showing in Table 3.2

				สุขภาพแร ท 2 และ1				
			(김 254	8 — 긥 255	55)			
โรคที่พบบ่อย	2648	2549	2550	2661	2662	2663	2554	2666
โรคซิฟิลิส	18 (0.07%)	106 (0.37%)	31 (0.15%)	23 (0.09%)	80 (0.80%)	4 (0.01%)	37 (0.08%)	18 (0.07%)
โรคพยาธิ์ เท้าข้าง	2 (0.01%)	12 (0.04%)	1	2 (0.01%)	14 (0.03%)	2	34 (0.07%)	(0.0%)
วัณใรคปลด	12 (0.05%)	65 (0.23%)	60 (0.28%)	17 (0.07%)	64 (0.12%)	25 (0.05%)	66 (0.15%)	13 (0.05%)
โรคเรื่อน	-	-	-	-	-	-	-	-
สารเสพติด	-	-	-	-	-	-	-	-
การตั้งครรภ์	283 (1.11%)	471 (1.65%)	512 (2.45%)	517 (1.99%)	610 (3.01%)	740 (1.54%)	1051 (3.28%)	655 (2.68%)

**Figure 3.2**: Investigation Report Form for registered immigrant workers in migrant clinic Samut Sakhon hospital

## 3.3 Tropical Diseases and Tropical Medicine

Tropical medicine began in the 19<sup>th</sup>century when doctors diagnosed infectious diseases in colonists and soldiers of war who had lived in tropical areas. In 1877, Scottish scientist, Sir Patrick Manson proved that mosquitoes spread elephantiasis to humans and later theorized that the same was true in malaria. (Cook et al., 2008)



**Figure 3.3:** malaria education campaign proposed by Sir Patrick Manson (image taken from Wikipedia online data based collection)

The field of tropical medicine consists of the study, treatment, and prevention of tropical diseases. Tropical diseases can be defined as those that are mainly of parasitic origin and are common in tropical or subtropical areas. (Binford et al., 1976) The climate of the tropics (between the latitudinal lines of the Tropic of Cancer (23.5N) and the Tropic of Capricorn (23.5S)) is a particularly fertile climate for these diseases (Cook et al., 2008), because of the insects that exist there. Insects such as mosquitoes, flies, fleas, lice, and ticks are the most effective way in which these diseases are transmitted between humans, from animals to humans, and between animals. In addition to these vectors, tropical diseases that are bacterial or viral are transmitted through soil and water (Hotez et al., 2007). Poor hygiene, crowded conditions and lack of education can add to the spread of disease in the tropics.

#### 3.3.1 Tropical Intestinal Parasitic Diseases

At least one-third of the world's population is infected with intestinal parasites which is making as common infections of human beings and one of common conditions detected in immigrant workers (CDC, 2012). Although these infections are asymptomatic and unnoticed, some have the potential to be chronic and lead to serious consequences.

Intestinal parasites that common make health problems classified as either single-celled protozoa or multicellular helminths. The helminths consist of two phyla: the hermaphroditic *Platyhelminths* (flatworms) and the Nematoda (nematodes or roundworms), each having separate male and female worms. The *Platyhelminths* are further subdivided into two classes: the trematodes (flukes) and cestodes (tapeworms) (CDC, 2012).

The pathogenic protozoa and coccidia most likely to affect immigrant include the ameba *Entamoeba histolytica/dispar* and the flagellate *Giardia intestinalis* (also known as *G. lamblia* or *G. duodenalis*). Two protozoa which potential to cause illness and associated with asymptomatic infection are *Blastocystis hominis* and *Blastocystis hominis* (Miller et al., 2000).

Nematodes or roundworms, are common parasites infecting humans and associated with the term "parasite" infection, due to the appearance in the body. Intestinal nematodes are transmitted by: soil contaminated ingestion with infective eggs (e.g., *Ascaris lumbricoides, Trichuris trichiura*) or skin penetration with infective larvae (e.g., hookworm, *Strongyloides stercoralis*). Generally, *Ascaris, Trichuris*, and hookworm may be associated with eosinophilia while hookworm with anemia, nutritional status (Hall et al., 2008), these organisms are rarely disease associated with immigrants.

Trematodes or flukes are a one of parasites that need snails as intermediate hosts. They cause chronic infection, with numerous important long-term consequences. All trematodes tend to cause eosinophilia due to the tissue-invasive character of chronic infection in specific population depending dietary of certain types of crustaceans, raw fish, or aquatic plants contaminated with the infective form.

The cestodes or tapeworms infect humans can be differentiated by adult characteristics divided into segments or proglottids. The mode of transmission by undercooked meats consumption. When humans eat undercooked pork, beef, or encysted larvi, fish, they can be infected with the adult form of *Taenia saginata*, *Taenia solium*, and *Diphyllobothrium latum*. The most serious morbidity and mortality is T. *solium* (CDC, 2012).

### Laboratory diagnosis for tropical intestinal parasitic diseases

The methods for identifying intestinal parasites are by the macroscopic and microscopic examinations of stool specimens for detection the ova and parasites. These methods are performed both protozoan and helminth infections and may be performed in a variety of laboratory settings, including the field laboratory. Although most intestinal parasites can be readily detected through by presence of ova or parasites, some require alternative or additional techniques for optimal detection such as *Strongyloides stercoralis*, *Cryptosporidium* spp.

In general, serology is the most useful method in detecting parasites with a disseminated or invasive stage, such as amebiasis (ie, liver abscess) (Singh et al., 2009), schistosomiasis, strongyloidiasis, and trichinosis. In contrast, serology is not useful for the detection of infections without a significant invasive component (eg, ascariasis, trichuriasis, and giardiasis) because these parasites do not generate a detectable immune response. For infections in which serology is used, cross-reactivity between helminth infections is common (Ishida et al., 2003).

#### 3.3.2 Tuberculosis

One third of the world's population is considered to have been infected with TB, with occurring at a rate of about 1 per second. In 2007, there were an estimated 13.7 million chronic active cases (WHO, 2011). In 2010, the incidence was estimated 8.8 million and 1.5 million associated deaths, mostly in developing countries.

Tuberculosis are infectious disease caused by various strains of mycobacteria, usually *Mycobacterium tuberculosis*. It attacks the lungs, but also affects other parts of the body. It is spread the air when people who have an active TB cough, sneeze, or otherwise transmit saliva through the air.

#### Laboratory diagnosis for tuberculosis

Active TB diagnosis relies on chest X-rays radiology, microscopic examination and body fluids microbiological culture. Latent TB diagnosis relies on the tuberculin skin test (TST) and/or blood tests. To overcome the resistance of drug treatment requires multiple administration antibiotics over a period of time. Social

contacts are also screened and treated. Prevention relies on screening and vaccination with the Bacillus Calmette–Guérin vaccine (BCG)

Definitive diagnosis of TB can be made by culturing *tuberculosis* from any specimen taken from the patient (most often sputum, but may also include pus, CSF, biopsied tissue, etc). The diagnostic methods performed out of the culture are classified as "probable" or "presumed" methods. If the patient is producing sputum, smears and cultures should be done for acid-fast bacilli and M.tuberculosis appearance, respectively.

#### 3.3.3 Syphilis

Syphilis is a sexually transmitted disease (STD) caused by spirochete *Treponema pallidum*, known as the "great imitator," with numerous and complex manifestations. It results in devastating cardiovascular and neurologic diseases.

Primary syphilis manifests as a solitary, painless chancre at the site of infection with average approximately 3 weeks after infection. Without treatment, *T. pallidum* spreads over blood to the next several weeks to months and results in secondary syphilis, with numerous clinical manifestations. The common features are fever, lymphadenopathy, diffuse rash (maculopapular), and genital or perineal condyloma latum (Kent et al., 2008). During the latent stage, skin lesions resolve, patients are in asymptomatic condition. However, serologic are positive. Tertiary or late syphilis develops years after the first and involve any organ system. The most dangerous complications are the involvement of the aortic valve and neurosyphilis.

#### Laboratory diagnosis for syphilis

Dark-field microscopy is the specific technique for diagnosing when active chancre or *Condyloma latum* is present. However, the accuracy is limited by the operator experience, the number of lesion *Treponemes* living, and the presence of non-pathologic treponemes in anal or oral lesions (Eccleston et al., 2008).

Syphilis leads the production of non-specific antibodies responding to cardiolipin. This reaction is the basis of conventional nontreponemal tests such as the VDRL test and rapid plasma reagin test (RPRT) (Fischbach et al., 2008). Qualitative nontreponemal tests are used for screening in the infected cases. However, it is limited

in sensitivity in early primary and during late phase, about one third of untreated patients diagnosesd as nonreactive result (Eccleston et al., 2008).

Treponemal specific test detect antibody (Ab) to antigen (Ag) components of *T. pallidum* which is used to confirm the diagnosis in a reactive non-treponemal test. However, the enzyme immunoassay (EIA) test for antitreponemal IgG also can be used for screening infected cases (Egglestone et al., 2000). Treponemal-specific tests are EIA for anti-treponemal IgG, the *T. pallidum* hemagglutination (TPHA) test, the microhemagglutination test with *T. pallidum* antigen, the fluorescent treponemal antibody-absorption test (FTA-abs), and the enzyme-linked immunosorbent assay. Treponemal tests have both sensitivities and specificities higher than those for nontreponemal tests. However, it is difficult and expensive to perform, which limits the usefulness as screening test (Taylor et al., 1988)

#### 3.3.4 Malaria

Malaria is a mosquito-borne infectious disease of human and other animals caused by protista of the genus *Plasmodium*. It starts with an infected female mosquito biting, which introduces the sporozoit via its saliva into the circulatory system, and straight to the liver where they reproduce and mature. The disease causes fever and headache, which in severe condition progress to coma and death. Malaria is widespread in tropical and subtropical regions in a broad band around the equator (Wyler et al., 1990). WHO has estimated that there were 216 million documented cases of malaria (2010) who were dead between 655,000 and 1.2 million people from the disease (roughly 2,000–3,000/day)

### Laboratory diagnosis of Malaria

The standard method for malaria diagnostic test is microscopic examination of Giemsa stained thin and thick blood films. Wright, Wright-Giemsa, or a rapid stain, such as Field, also is used as long as white blood cells (WBC - stained the same as parasites) are stained well. Although this method requires a minimum amount of resources (staining materials and high-quality microscopes), but still well-trained experienced technologists and must be available. Blood film examinations ideally should be examined within 2 to 3 hours. In addition, if blood is collected in

EDTA tubes, parasite morphology may be altered by delays in preparation of the slides (Versalovic, 2011).

The thick blood film is a lysed concentrate technique of red blood cells (RBCs) and the most sensitive preparation for the microscopic identification of *Plasmodium* (Versalovic, 2011). Thick films do not need definitive identification of the infecting species. However, the thin film can be used to specify and determine the degree of parasitemia. Parasitemia is expressed as a percentage of RBCs parasitized for direct initial therapy utilization or to follow the response to therapy.

Rapid antigen diagnostic tests (RDTs) are available, the use of RDTs has been shown to be preferable to routine microscopy in some studies (Kahama et al., 2011). Positive RDTs should be confirmed by blood film examination, which are necessary to determine the species. This RDT is less sensitive than a thick blood film and may be false negative in cases with very low degrees of parasitemia (Stauffer et al. 2006).

Rapid real time PCR (polymerase chain reaction) assays for malaria diagnosis have also been developed. PCR is used to confirm malaria diagnosis in cases empirically treated without prior laboratory confirmation through remnant nucleic acid detection. PCR tests remain persistent positive with all forms of *Plasmodium* seen in the peripheral blood. Finally, Immunofluorescence antibody testing (IFA) has been a reliable serologic test for malaria in recent decades. It is available but it is not generally useful for acute disease diagnosis. Although IFA is time-consuming and subjective, it is highly sensitive and specific (She et al., 2010).

#### 3.3.4 Filariasis

Filariasis is an infectious tropical parasitic disease caused by thread-like nematodes (roundworms) in Superfamily Filarioidea, also known as "filariae". The most common is lymphatic filariasis caused by *Wuchereria bancrofti*, *Brugia malayi*, and *Brugia timori*, which occupy lymphatic system (lymph nodes) and in chronic cases lead to elephantiasis. In 2009, approximately 9.5 million individuals have already been infected with the lymphatic filariasis (Hooper et al., 2009).

#### Laboratory diagnosis of Filaria

The standard method for *lymphatic filariasis* diagnosis is microscopic examination of thin and thick blood or buffy coat films stained with Giemsa. The QBC system also be used to enhance sensitivity but it is not widely used. Additional thin films are required to determine the identity of any microfilaria presence. Live motile microfilaria is observed in fresh wet preparations of blood or buffy coat samples. Concentration methods, such as the Knott technique can increase the sensitivity of light microscopic examination.

The identification of adult filarial worm's identification can be performed through either gross examination or microscopic examination of histopathology sections. Filarial serology is also available as it can detect filarial immunoglobulin (Ig) antibodies but the assay is limited to nonspecific reactions and provides only qualitative positive or negative results (Versalovic, 2011)

### 3.4 Infectious diseases in immigrant workers in Thailand

Human migration can result in the transport of pathogens in long distances and infectious diseases can spread rapidly due to the movement of migrant workers. Registered immigrants in Thailand have to pass a routine immigration medical screening every year.

#### 3.4.1 Intestinal parasitic infections

Intestinal parasitic infections are an important public health problem, especially in developing countries. These infections are frequently found in Thailand and Myanmar. According to the growth of labor demand, foreign workers from low wage countries, mainly Myanmar, Cambodia and Laos have migrated to urban areas of Thailand, particularly Bangkok and Samut Sakhon where are industrialized provinces. A cross-sectional survey on intestinal parasitic infection among Myanmar workers in Bangkok and Samut Sakhon was done by Ngrenngarmlert W et.al (2012). 213 stool samples were examined for intestinal parasites by using simple smear and formalinether concentration techniques. The overall prevalence of intestinal parasitic infections

was 13.6%. Helminth and protozoan infection were 10.3% and 8.5% respectively. In this study, soil transmitted helminthes (*A. lumbricoides* and *T. trichiura*) and the fecal oral transmitted parasites (*E. histolytica, G. lamblia and B. hominis*) were commonly found in Myanmar migrant workers. (Nuchprayoon et al., 2009)

**Table 3.3:** Prevalence of intestinal parasites found in 215 Myanmar workers in Bangkok and Samut Sakhon

Parasite class species	Infected cases (N=213)	Prevalence (%)
Helminthes		
A. lumbricoides	7	3.3
T. trichiura	5	2.3
Taenia spp.	5	2.3
Opisthorchis spp.	3	1.4
Hookworm	2	0.9
<b>Subtotal helminths</b>	22	10.3
Protozoa		
E. histolytica	8	3.8
G. lamblia	2	0.9
B. hominis	2	0.9
E. coli	2	0.9
E. nana	4	1.9
Subtotal protozoa	18	8.5

#### 3.4.2 Malaria infection

In Thailand, the increase of immigration from Thailand-Myanmar and Thailand-Cambodia borders where malaria is endemic has been a significant risk in immigrant workers (Ngrenngarmlert et al., 2012). Lack of epidemiological data, especially the prevalence of malaria among foreign immigrant workers, the impact of situation on public health could not be determined. All the registered immigrant workers have to receive a health screening program for communicable diseases and

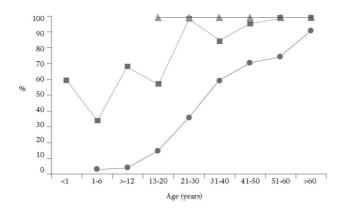
malaria is also on the list in this program. Ngrenngarmlert et al (2012) conducted a cross sectional survey in Bangkok and Samut Sakhon Province. The results from malaria blood film showed that 1.5% of Myanmar immigrant workers were infected with malarial parasites without any symptoms were defined as the asymptomatic *Plasmodium* spp. Infections based on physical examination including clinical interviews.

**Table 3.4:** Prevalence of Plasmodium infections among foreign immigrant workers

	Parameters	No. of examined	No. of positive (% prevalence)
Nationality	Myanmar	265	4(1.5)
	Laos	10	0(0.00)
	Cambodia	19	0(0.00)
Resident	Bangkok	174	2(1.10)
province	Samut sakhon	120	2(1.70)
Type of blood	Thin blood smear	294	1(0.34)
smears	Thick blood smear (single)	294	3(1.02)
	Thick blood smear (double)	294	4(1.36)
	Thick and thin blood smears	294	4(1.36)

#### 3.4.3 Hepatitis A infection

Hepatitis A virus remains a major public health problem wherever hygiene and sanitation are inadequate. There is some concerns that areas with large populations of immigrants from the neighboring countries of Myanmar, Cambodia and Laos may still be hyper-endemic for HAV, although little is known about the HAV situation among these immigrants or in their countries of origin (Rianthavorn et al., 2011). The result showed the age related seroprevalence of HAV among the people residing in the border area was significantly higher than that of the general Thai population but lower than Myanmar immigrant workers (Rianthavorn et al., 2011).



**Figure 3.4.** Age-related seroprevalence of hepatitis A virus among Thai population residing near Myanmar border ( $\nu$ ), general Thai population ( $\lambda$ ), and Myanmar immigrant workers ( $\triangle$ )

#### 3.4.4 Filariasis

In Thailand, lymphatic filariasis has been well controlled and endemic areas are limited to only 5 provinces: Tak, Kanchanaburi, Mae Hong Son, Surat Thani, and Narathiwat, with the low prevalence rate of 3.7 per 100,000 population. The migration of Myanmar labor into Thailand for better jobs and incomes has had a significant impact on public health of Thailand. These Myanmar immigrants have not only come with their labor but also with many infectious organisms. Out of many infections they carry lymphatic filariasis needs considerable attention. The survey of microfilaria rate in the Myanmar migrant working in Maesot district, Tak province, Thailand was done in by Triteeraprapab (1999). The result showed that the microfilaria rate in 654 Myanmar immigrants working in Maesot district was 4.4 per cent. The highest microfilaria rate was found in male aged 21-30 years (6.8%) and the majority of Myanmar migrants (55.5%) have been staying in Thailand 1-6 years; most of them (82.0%) have never been back to Myanmar.

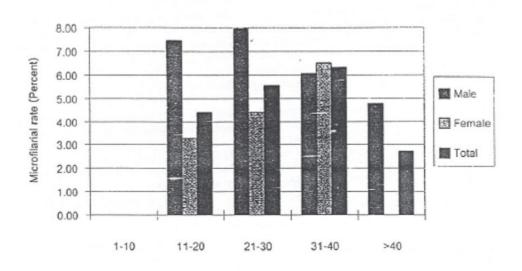


Figure 3.5. The microfilaria rates of Myanmar migrants classified by age group.

# CHAPTER IV MATERIALS AND METHODS

## 4.1 Design, period and place of study

This study was a prospective study carried out at the Immigrant Clinic, Samut Sakhon Hospital, Thailand during March until June 2013.

## 4.2 Study population

Study population was adult immigrant workers in fishery and marine food preparation factories of Samut Sakhon Province. Participants who fulfilled the eligible criteria were interviewed and proceeded to physical examination and laboratory investigation.

#### 4.3 Inclusion criteria

- 1) Adult male and female participants were in the age 18 60 years.
- 2) Participants were Non-Thai people.
- 3) Participants can understand and communicate in Thai language.
- 4) Participants were willing to comply with the study protocol.
- 5) Individual written informed consent was obtained.

#### 4.4 Exclusion criteria: None

#### 4.5 Methods

The collection data was divided in two phases:

- 1) Phase 1: Principal investigator asked permission from Dr. Molee Wanichsuwan, Director of Samut Sakhon Hospital, to collect the participant's data and samples.
- a. History taking and physical examination were done by local authorized medical doctors, including:
- i. History taking: identity (code), age, sex, date of screening, original and underlying diseases.
- ii. Physical examination: weight, height, blood pressure, heart rate, respiratory rate, head, ears, eyes, throat, heart, thorax, abdomen, skin, extremities
  - b. Laboratory investigations:
    - i. Routine laboratory [by hospital laboratory staff]
    - 1) Blood smear for malaria and filarial worms

(thick smear)

2) VDRL test for syphilis (venereal disease

research laboratory)

- 3) Chest X-ray
- 4) Urine pregnancy test for female
- ii. Non-routine laboratory [by hospital laboratory

staff]

Rectal swab for stool examination in normal saline

solution

- 2) Phase 2: Principle investigator used the samples (rectal swab samples and thick blood film) to be tested at Laboratories of Department of Helminthology, Faculty of Tropical Medicine and The Bangkok School of Tropical Medicine, Mahidol University
- a. Microscopic examination for intestinal parasites (helminth and protozoa)

b. Microscopic examination for malarial and filarial parasites.

#### A. Intestinal parasitic examination

A total of 198 rectal swab samples, which put in to small bottle with agar gel after getting rectal swab examination informed consent, were studied. Samples were come from adult immigrant workers who worked as fishery or marine food preparation. They came to immigrant clinic Samut Sakhon Hospital for health check by hospital staff. The staff in Samut Sakhon hospital collected and sent the samples for the first 20 immigrant every single day to the laboratories of Department of Helminthology Faculty of Tropical Medicine to be examined. Rectal swab sample that having delay for examination was stored in cool temperature room in order to minimize the risk of fungal contamination.

Each rectal swab sample specimen was separated from the agar by using straw, then additional 0.05 milliliter normal saline (with micropipette) mixed in small tube (0.5) before proceeding to centrifuge methods at 2000 rpm for 5 minutes. After centrifuge, the supernatant was separated with micropipette and the sedimentation was taken by another new micropipette to slide (object glass). Every sample was examined with 2 routine methods, with normal saline and iodine solution.

#### **Procedure**

One drop of normal saline solution was put in the middle of the left half of the slide and one drop of *Lugol's* iodine in the middle of the right slide. The drop of normal saline on the left half of the slide should be mix first, continue with right iodine part. Then, cover slips were added to each left for normal saline and right for iodine part with slowly from the side to avoid bubbles. Entire cover slip was ready to be examined. For the saline preparation, the 10x objective and the 40x objective were for helminthic infection used, and for the Lugol's iodine preparation were used the 40x objective.

#### B. Malaria and filarial examination

Blood specimens were collected from adult immigrant workers that came to the immigrant clinic, Samut Sakhon Hospital by hospital staff. All specimens were examined by *Giemsa* stain.

#### Thick film

The drop of blood in a circle, the size was spread by corner of a clean slide of a dime (diameter 1-2 cm). The thick films were waited until completely dry before staining.

For Giemsa 3% solution preparation, 97 mL of buffered water (ph 7.2) was poured into a 100 mL graduated cylinder. Using Pasteur pipette, 3 mL of Giemsa stain was added to the graduated cylinder in order to make 3% concentration. The top of the graduated cylinder was covered and inverted several times until completely mixed.

The smears were allowed to dry quickly by application a fan or blower at room temperature. Thick blood film was placed in a staining rack and stain singly, or in batches in staining jar to avoid cross-contamination. Films were stained for 30-45 minutes out of the sunlight. Clean tap water was poured gently into the trough to float off the iridescent scum on the surface of the stain. Then slides were rinsed briefly and gently under running tap water.

The stained slides was placed in a drying rack to drain and dry, film side in downwards position, it would make sure the film did not touch the slide rack. During preparation and storage time, slides should be protected from exposure to dust and insects.

### 4.6 Data analyses

Data were collected according the approved CRF. Afterwards, data entry was carried out with coding and verified. Validity and consistency of data gathering were checked. Data were analyzed by using SPSS program.

Descriptive statistics (mean, mode, quartile, minimum maximum data, rang, etc) were used to describe the percentage distribution or proportion of demographic data, clinical symptoms and signs and laboratory tests of the participants.

Categorical variable associations were determined association using either Chi-square test or Fisher exact test as appropriate. Normally continuous distributed data were compared by the Student's t-test. Statistical significance was considered when p-value <0.05.

### 4.7 Sample size calculation

To get sufficient power of study we conduct sample size estimation.

n =  $Z_{1-\alpha/2}^2 P(1-P)/d^2$ 

n = sample size

 $Z^{2}_{1-\alpha/2}$  = confidence interval (CI 95% = 1.96 [two-tail test])

P = estimated proportion (Previous Prevalence data)

d = desired precision (0.05)

The latest study from The Department of Parasitology and Community Health, Faculty of Medical Technology Mahidol University August 2012 show the prevalence of Intestinal parasites among immigrant workers in Samut Sakhon Province = 13.6% (Ngrenngarmlert, 2012)

$$n = ([1.96x1.96] \times [0.136] \times [1-0.0136]) / [0.05x0.05]$$
  

$$n = 180$$

198 of 200 participants (immigrant workers who work in marine food preparation – fishery group who came for work checkup at immigrant clinic, Samut Sakhon hospital) were enrolled in this study. The reasons of 2 cases who excluded were (1) data missing and lower age (1).

# CHAPTER V RESULTS

## 5.1 Immigrant workers demographic data

A prospective study of 198 Myanmar immigrant workers who checked up at the immigrant clinic, Samut Sakhon Hospital was enrolled. 2 immigrants were excluded due to age exclusion and data missing. (Figure 5.1)

Gender distributions (M: F) of participants were 41:59. The number of female participants was higher than male group. (Figure 5.2)

Immigrant workers participants were divided into 2 age groups. In this study 172 participants (86.9%) were less than or equal to 35 years old, and 26 participants (13.1%) were more than 35 years old. The distribution age less than or equal to 35 is the highest number of participant group in this study. (Figure 5.3a)

Immigrant workers participants were divided into 2 duration of working groups. In this study 24 participants (12.1%) were worked for less than 1 year and 174 participants (87.9%) were worked for more than 1 year. The working duration for more than 1 year has the higher number of participant group in this study. (Figure 5.4)

### 5.2 Physical examination data

Median and range result for body weight, height, basal metabolism index, systolic and diastolic pressure, temperature and pulse rate from 198 immigrant workers in this study were 53 (40-85), 159 (125-180), 20.9 (15-33.9), 120 (86-120), 80 (50-98), 37.2 (36-38) and 79 (60-101) respectively. (Table 5.1)

In all BMI (Body Mass Index) classification group based on WHO 2004 criteria guidelines, there was only one outlier (data outside the 1.5 time lower box scale) in female underweight group (1 participant with BMI 15kg/m<sup>2</sup>). (Figure 5.5)

Only one (0.05%) immigrant had physical examination abnormality there was hypothyroid. Table (5.2)

### 5.3 Laboratory examination data

We conducted chest X-ray investigation for possibility pulmonary abnormality related with tuberculosis, smear for both malaria and filarial parasites, and VDRL test for syphilis. We found only 1 abnormality in chest X ray immigrant, but unfortunately we could not identified as tuberculosis due to we did not yet proceed to smear procedure as a golden standard. For malaria and filarial laboratory investigation, we detected negative parasites (thick blood smear) from 198 blood samples. We found the only positive result from our tropical diseases study among immigrant who checked up to Samut Sakhon Hospital routine survey was intestinal parasites (include helminthes and protozoa). We found 5 species of parasites: *Entamoeba hystolytica*, *Entamoeba coli*, *Endolimax nana*, *Giardia lamblia* and 1 species helminth *Enterobius vermicularis* from 9 participants from total 198 immigrant workers (4.5%). (Table 5.3)

Intestinal protozoa infection (3%) was dominant infection in this fishery immigrant workers compared to intestinal helminthes (1.5%). Among the intestinal protozoa infection, *Entamoeba coli* and *Giardia lamblia* were the most common infection in equal percentage (1%) than *Entamoeba hystolytica/dispar* and *Endolimax nana* (0.5%), whereas *Enterobius vermicularis* infection (1.5%) was the single species found and the highest infection of overall species (1.5%).

The overall prevalence of intestinal parasites infections in this study was 4.5%. The prevalence of intestinal parasites infections was more common in male (2.5%) rather than female (2%). The highest prevalence was 3.5% in the group more than 35 years old. In our study group, the type of infection was single infection only (4.5%). (Table 5.4a)

Prevalence of intestinal parasitic disease higher in underweight group and statistically significant difference with normal BMI group with p-value 0.04 less than cut-off point 0.05. All type infection in immigrant worker participant was single infection parasite. (Table 5.4b)

Intestinal parasites infections have higher percentage in both underweight group (22.2%) than in normal group (4.8%) in male gender, and 11.1% underweight female group to 2.6% normal female group. Male group infection (22.2% and 4.9%) tended higher than female group (11.1% and 2.6%). (Figure 5.6)

From population pyramid chart showed the intestinal parasitic infection immigrant age distribution predominates within range 20-30 (infection rate 66.6%). Figure (5.7)

### 5.4 Intestinal parasites and Others Tropical diseases

There were no associations between intestinal parasites occurrences with others tropical diseases. The result of abnormality findings in Chest X-ray represent tuberculosis was P-value more than cut off point 0.05 and there were no measurement of association among the result of blood thick smear represent malaria - filarial and VDRL test with intestinal parasites infection in this group of study due to constant variable result.

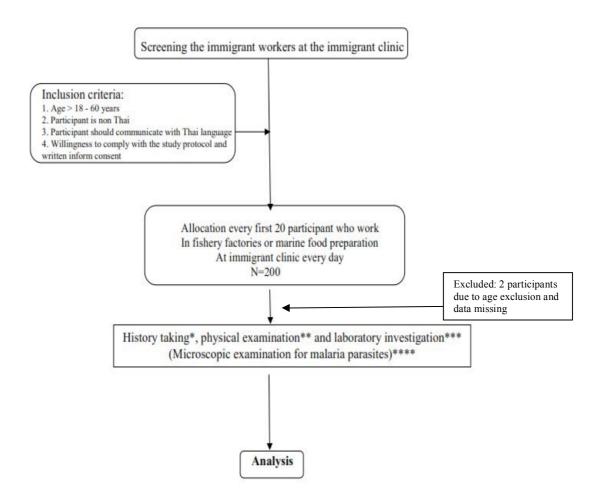


Figure 5.1: Enrolment of patients

- \*History taking: identity (code), age, gender, working duration, underlying disease
- \*\* Physical examination: weight, height, blood pressure, head, ears, throat, heart, skin, extremities and conclusion examination
- \*\*\* Routine immigrant clinic Laboratory program: VDRL test for syphilis
- \*\*\*\* Additional laboratory examination: blood smear for malaria and filarial (thick smear), and rectal swab for stool examination.

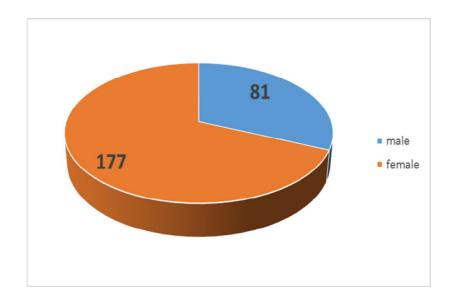


Figure 5.2: Participant's gender group

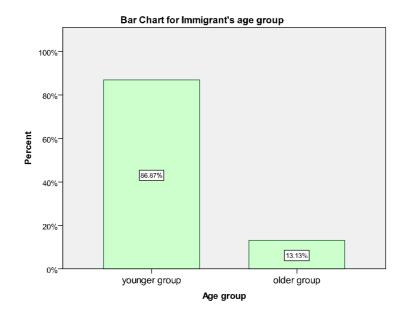


Figure 5.3: Participant's age group (Bar Chart)

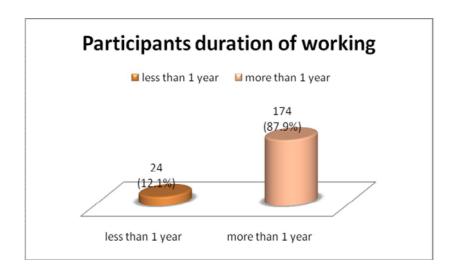


Figure 5.4: Participant's duration of working

 Table 5.1: Vital sign result

Characteristic	Median	Range
Body weight (kg)	53	40-85
Body height (cm)	159	125-180
Basal Metabolism Index (Kg/m²)	20.9	15-33.9
Systolic blood pressure (mmHg)	120	86-130
Diastolic blood pressure (mmHg)	80	50-98
Body temperature (°C)	37.2	36-38
Pulse rate (min)	79	60-101

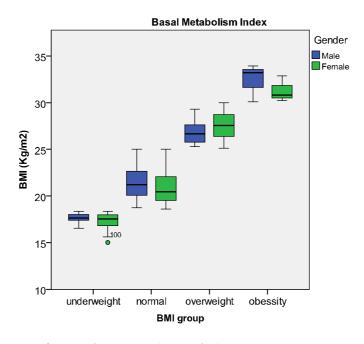


Figure 5.5: BMI for gender group (Box Plot)

**Table 5.2:** Physical exam result (n=198)

Physical exam	Number (abnormal)	Percentage
HEENT	1 (hypothyroid)	0.5 %
Cardiovascular	0	0 %
Lung	0	0 %
Abdomen	0	0 %
Skin	0	0 %
Extremities	0	0 %

 Table 5.3: Laboratory examination result

Laboratory result	Positive result N=198	Description
Stool examination	9 (4.5%)	
	2 (1%)	Cyst form
Protozoa		
Entamoeba coli	1 (0.5%)	Trophozoite
Entamoeba hystolytica		
Endolimax nana	1 (0.5%)	Cyst form
❖ Giardia lamblia		
Helminth	2 (1%)	Cyst form
Enterobius vermicularis		
	3 (1.5%)	Egg
Chest X-ray	1 (0.5%)	
Thick Blood smear (malaria and filarial)	0	
VDRL (reactive)	0	

Table 5.4a: Prevalence of intestinal parasites found in immigrant workers

Characteristic	Immigrant (n)	Positive (n)	Prevalence (%)	p-value
Age				
≤ 35	172	7	3.5%	0.336*
>35	26	2	1%	0.550
Gender				
Male	81	5	2.5%	0.491*
Female	117	4	2%	0.171
Working duration				
Less than 1 year	24	1	0.5%	0.999*
More than 1 year	174	8	4%	

<sup>(\*)</sup>No statistically significance different between age, gender and working duration group to intestinal parasites infection among immigrant workers in Samut Sakhon Hospital (Significance p-value Fisher's exact test >0.05)

Table 5.4b: Prevalence of intestinal parasites found in immigrant workers

Characteristic	Immigrant (n)	Positive (n)	Prevalence (%)	p-value
BMI				
Underweight	27	4	14.8%	0.04
Normal	139	5	3.6%	0.01
Parasitic infection				
Mono infection	198	9	4.5%	
Mix infection	198	0	0 %	

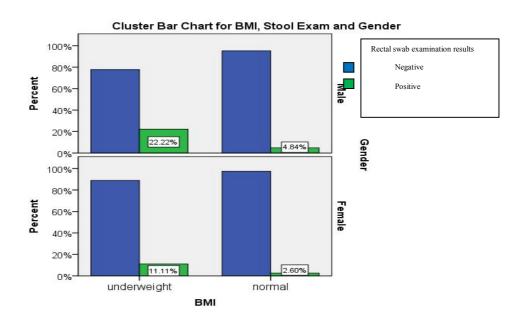


Figure 5.6: Intestinal parasitic infection between gender and BMI group

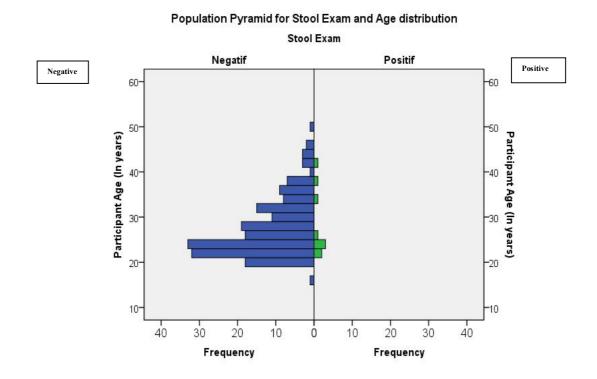


Figure 5.7: Age distribution among intestinal parasitic infection immigrant

## CHAPTER VI DISCUSSION

In this study we aimed to obtain the prevalence of intestinal parasitic infections including other helminthic-protozoan infections and possible tropical diseases (tuberculosis, malaria, filariasis, and syphilis) among adult immigrant workers who visited at immigrant clinic in Samut sakhon hospital. The results showed that the prevalence of intestinal parasites was common in male, younger age, longer duration of work and underweight immigrants.

Our study revealed that prevalence of intestinal parasitic diseases among Myanmar immigrant adult worker who visit Samut Sakhon immigrant clinic in order to get working permit was 4.5%. This percentage was decreasing from the previous studies that were conducted by Ngrenngarmlert et al in 2012. They reported 8.2% (7 from 85 examined cases).

Even though the percentage was decreasing, but the actual number was raised. The number of samples being examined was also double up in our study, 85 cases in 2012 comparison with 198 cases in 2013. In this study, we used modified technique by means different with the routine one. In the routine examination, small amount of stool was used directly with normal saline solution or iodine. But, due to specimen availability from the Immigrant Clinic Samut Sakhon Hospital routine screening survey (utilization small bottle with agar gel for culture purpose) we used modified technique for separation fecal debris in cotton from rectal swab examination with agar gel using straw. After separation using straw, we proceed to centrifuge method in order to get fecal sedimentation before giving the normal saline solution or iodine in object glass.

There were 2 possibilities why our study revealed low prevalence. First, immigrant who came to clinic already knew that they would have a problem if the finding of the disease was positive. That was why a chance for them to provide self-medication in this case anthelminthic drug was undoubtable. The second one was

possibilities for low sensitivity from our diagnostic tools. Our method had limitation from both utilization rectal swab specimen instead direct stool materials in routine procedures and our methods by simple smear examination itself.

Our finding was equal with the low prevalence of parasitic infection in immigrant workers in Thailand with prevalence 4-12% reported by Yoon et al. (2002), Maneeboonyang et al. (2004), Changsap et al. (2005), Nuchprayoon et al. (2006), Ngrenngarmlert et al. (2007) and Nuchprayoon et al. (2009) This phenomenon correlated with the usual condition, which happens among immigrants such as poor hygienic condition, overcrowding, limitation to health facilities, low health education and low socio-economic status.

Nowadays, mass migration to developed countries is not only cause large impact to global population but also resulted in new public health problems. These conditions reflected by the rapid appearance of tropical diseases in developed countries (so do in Thailand) such as malaria case in the United States reported by Thwing et al. (2007) and tuberculosis in Sweden and many more diseases by Lifson et al. (2002), LEZ et al. (2003), and Montresor et al. (2004). CDC report 1,528 cases of malaria only during 2005 and EURO surveillance published 552 cases in 2008 reported by Devaux et al. (2010). Routine screening test is absolutely needed for destination countries to prevent and stop disease transmission from foreign workers.

Our data is also supported with several studies on common relationship between the diseases problems and intestinal parasitic infection immigrant workers in many countries. The prevalence varies from 8-40% reported by Wang et al (2004), Lopez et al (2003), Lifson et al (2002) and Arfaa et al (1981).

In 1997, The government of Myanmar claimed that they had achieved crucial improvement for the national sanitation week (NSW) system and social mobilization for sanitation and hygiene from 45% in 1997 to 67% in 2001 reported by Bajracharya (2003). However, intestinal parasitic infection cases still persist in high level even after 4 years improvement in that county (18.5% in 2005) reported by Ohnmar (2005)

The prevalence of intestinal parasitic infection varies from one to other region. Montresor et al. (2004) emphasize that the distributions of intestinal parasites was influenced with geographical areas. There was significance difference distribution

among hilly, plains, delta and coastal area. Montresor mentioned the hilly (69% infection) area was the highest burden of parasite after being confirmed with the previous study in Myanmar. In our study, Samut Sakhon area is located in the coastal area; theoretically it was categorized to lower transmission than the hill area but the rapid migration phenomenon in Samut Sakhon makes the exception due to heterogeneity of immigrant origins area.

Distribution of intestinal parasites depends on climate. Soriano et al in 2001 investigated the relationship between intestinal parasites with environment (climatic factors) in 69 children in Patagonia, Argentina brought high prevalence 50.7% intestinal parasites and conclusion strong correlation between these 2 factors (p-value 0.0001) reported by Soriano et al. (2001)

Another study in two periurban population in La Plata Argentina by Inés et al. (1996) taken conclusion the importance of personal or community sanitation and hygiene in controlling intestinal parasitic infection as well study from Qatar, Kenya, Taiwan, United States Lao and Myanmar. (Salas et al., 1990; Adams et al., 1994; Geltman et al., 2003; Garg et al., 2005; Cheng et al., 2006; Abu-Madi et al., 2010; Hsieh et al., 2010)

In our study, the younger age group had higher risk of infection for both protozoa and helminthes. This might be explained by younger age group have more activity contact related with occupation, in this field as fishery or marine food preparation or higher chance to expose with infective agent. In contrast with the older age group that less likely active and raised level of immune status, as note by others.

Male gender was more chance to have parasitic infection. This findings support a previous study from Nacher et al (2003) that male tend to get parasitic infection which might be due to their immunity, activity and gender related habits. Working duration also plays important role in diseases susceptibility. Immigrants will susceptible to infection or re-infection if contact with infectious agent lasting longer.

In this study, among positive cases protozoa predominated (66.6%) to helminthes infection (33.3%). It was interesting to analyze that the intestinal protozoa predominate in this immigrant worker population while the occurrence of intestinal helminthes was lower. Our findings make impression the wide distribution of intestinal protozoa, particularly *Giardia lamblia* (22.2%), *Entamoeba coli* (22.2%),

Entamoeba hystolytica/dispar (11.1%) and Endolimax nana (11.1%) in this immigrant population. Our findings similar with 3 studies, two from Thailand by Ngrenngalermlert in 2007, that intestinal protozoon (10.7%) predominate to intestinal helminthes (0.5%) in 1920 children on Phuttamonthon district by Ngrenngarmlert et al. (2007)

The second one from Argentina by Soriano et al (2001) that gave explanation climatic factors, in conjunction with soil characteristic (sandy, low organic matter content, nutrient retention and capacity of water related with humidity), air humidity as well temperate determine a not very favorable environments for the development and survival of parasitic specific cyst (protozoa infected form), eggs or larva (helminthes infected form). This condition would justify the parasitic species distribution found in stool sample, where only protozoan species were detected.

The third one by Nuchprayoon (2009) that stated among Myanmar immigrant, the fecal-oral transmitted parasites (i.e. *Giardia lamblia, Blastocystis hominis*) were common. From 177 infected cases of 284, 89.3% was infected by *Giardia lamblia*. In contrast, the parasites commonly identified in Thais was helminthes group such as *Ascaris lumbricoides* (16-49%), hookworms (22-38%), *Strongloides stercoralis* (4-33%) and *Ophistorchis viverini* (12-24%) while *Giardia lamblia, Blastocystis hominis* was found with low prevalence.

We found the fecal-oral transmitted parasites (*Entamoeba hystolytica/dispar*, *Giardia lamblia*, *Endolimax nana*, *Entamoeba coli and Enterobius vermicularis*) were common found in immigrant workers. This was might be due to Myanmar immigrants prefer consumes fresh vegetable while came to new environments, blend with Thai society proposed by Nuchprayoon et al. (2009), and raw food that have a big change contaminated with infective form cyst (protozoa) or infective egg (parasites). In addition, they lived in poor crowded area that still limited hygiene facilities and medical service.

These parasites can effect to health condition including malnutrition, malabsorption syndrome and growth retardation for long time consequences analyzed by Oberhelman et al. (1998). He stated the prevalence of intestinal parasites among children in rural Nicaragua was 40%, 14.6% among them got malnutrition with lowering DENVER score as consequences growth and intelligence retardation.

Overall, poor hygienic factor, unwell-food or water plays huge role factors as disease transmission. Therefore, well-cooked food consumption promotion, clean water utilization and sanitation improvement including ownership own family latrine should be enhanced.

Jumma et al (2005) stated that the simplest way and the most important things to minimize diseases transmission of these intestinal parasitic infections were likely to be hand-washing promotion program. Medication after screening for those infected was crucial part to stop the transmission. Adams et al (1994) revealed the importance of medication in 55 Kenyan primary school children with intestinal parasites infection. There was significant improvement in growth, activity, appetite and clearance parasitic rate in medication group compare with placebo (p-value <0.0002).

The routine screening program (followed with medication for those infected) in Immigrant clinic Samut Sakhon Hospital for Myanmar workers should include stool examination for intestinal parasites. Health education every visit to clinic and poor socio-economic improvement program should be more emphasized in order to suppress the disease transmission and control intestinal parasitic infection from foreign immigrant workers.

There was a study from Lopez et al (2003) as part of European Union program that conducted screening survey for tropical diseases among immigrant from Africa and Latin America. 988 immigrant followed the testing protocol include medical history, complete physical examination and laboratory examination similar with protocol in our study.

The result indicated higher prevalence of tropical diseases case among immigrant workers in Spain (2003) compare with Samut Sakhon, Thailand (2013). This condition might be there were more endemic diseases by means of quantity and variety in immigrant origins from Africa and Latin America compared to Myanmar. Hotez et al reported the estimate global burden of tropical diseases for intestinal parasites, malaria, tuberculosis and STD in Sub-Sahara Africa region only was 18.3 million, 40.9 million, 9.3 million and 64 million respectively reported by Hotez (2009). In the other studies, Hotez reported the estimate global burden of tropical

diseases for intestinal parasites, malaria, tuberculosis and STD in Latin America region was 56 million, 46.5 million, 34.7 million and 84.5 million respectively.

In our study, as a pilot project, we only used standard routine procedure due to limitation of financial resources to examine the immigrant such as simple smear for intestinal parasites, thick blood smear for malaria and filarial, VDRL test for syphilis and radiology chest X ray for pulmonary abnormality tuberculosis as compare with additional advance laboratory examination such as DNA detection by PCR, CT scan, USG, ELIZA, Western blot and many sophisticated tools that have higher specificity and sensitivity diagnostic tools than our standard procedure.

## CHAPTER VII CONCLUSION

The economic growth of Thailand has attracted a large number of people from the neighboring countries for seeking better life that bring consequences to new threat for a role in diseases transmission. We found the prevalence of intestinal parasitic disease (in our tropical diseases study) as 4.5%.

Regular screening and proper medication for those infected combine with mass education preventive promotion program hopefully can prevent even stop disease transmission from immigrant to local community.

### REFERENCES

- Abu-Madi, M. A., J. M. Behnke and S. H. Doiphode (2010). "Changing trends in intestinal parasitic infections among long-term-residents and settled immigrants in Qatar." <u>Parasites & vectors</u> **3**(1): 98.
- Adams, E. J., L. S. Stephenson, M. C. Latham and S. N. Kinoti (1994). "Physical activity and growth of Kenyan school children with hookworm, Trichuris trichiura and Ascaris lumbricoides infections are improved after treatment with albendazole." The Journal of nutrition **124**(8): 1199.
- Arfaa, F. (1981). "Intestinal parasites among Indochinese refugees and Mexican immigrants resettled in Contra Costa County, California." The Journal of family practice 12(2): 223.
- Bajracharya, D. (2003). "Myanmar experiences in sanitation and hygiene promotion: lessons learned and future directions." <u>International Journal of Environmental Health Research</u> **13**(S1): S141-S152.
- Binford, C. H. and D. H. Connor (1976). "Pathology of tropical and extraordinary diseases. An atlas. Vols 1 and 2." <u>Pathology of tropical and extraordinary diseases</u>. An atlas. Vols 1 and 2.
- CDC (2012). Domestic Intestinal Parasite Guidelines. <u>Immigrant and Refugee Health</u>. ATLANTA, Centers for Disease Control and Prevention.
- Changsap, B., P. Boontan and N. Arnat (2005). "A survey on the prevalence of intestinal parasitic infection in Khlong Toei district." <u>Songklanagarind Medical Journal</u> **23**(4): 219-227.
- Cheng, H.-S., Z.-F. Haung, W.-H. Lan, T.-C. Kuo and J.-W. Shin (2006).

  "Epidemiology of Blastocystis Hominis and Other Intestinal Parasites in a Vietnamese Female Immigrant Population in Southern Taiwan." The Kaohsiung Journal of Medical Sciences 22(4): 166-170.
- Cook, G. C. and A. Zumla (2008). <u>Manson's Tropical Diseases E-Book</u>, Saunders Limited.

- Eccleston, K., L. Collins and S. P. Higgins (2008). "Primary syphilis." <u>International</u> journal of STD & AIDS 19(3): 145-151.
- Egglestone, S. and A. Turner (2000). "Serological diagnosis of syphilis. PHLS Syphilis Serology Working Group." Communicable disease and public health/PHLS **3**(3): 158.
- Fischbach, F. and M. B. Dunning (2008). <u>A manual of laboratory and diagnostic tests</u>, Lippincott Williams & Wilkins.
- Garg, P. K., S. Perry, M. Dorn, L. Hardcastle and J. Parsonnet (2005). "Risk of intestinal helminth and protozoan infection in a refugee population." <u>The American journal of tropical medicine and hygiene</u> **73**(2): 386-391.
- Geltman, P. L., J. Cochran and C. Hedgecock (2003). "Intestinal parasites among African refugees resettled in Massachusetts and the impact of an overseas pre-departure treatment program." The American journal of tropical medicine and hygiene **69**(6): 657-662.
- Hall, A., G. Hewitt, V. Tuffrey and N. De Silva (2008). "A review and meta-analysis of the impact of intestinal worms on child growth and nutrition." <u>Maternal</u> & child nutrition 4: 118-236.
- Hooper, P., M. Bradley, G. Biswas and E. Ottesen (2009). "The Global Programme to Eliminate Lymphatic Filariasis: health impact during its first 8 years (20002007)." <u>Ann Trop Med Parasitol</u> **103**(Supplement 1): 17-21.
- Hotez, P. J., D. H. Molyneux, A. Fenwick, J. Kumaresan, S. E. Sachs, J. D. Sachs and L. Savioli (2007). "Control of neglected tropical diseases." New England <u>Journal of Medicine</u> **357**(10): 1018-1027.
- Hsieh, M.-H., W.-Y. Lin, C.-Y. Dai, J.-F. Huang, C.-K. Huang, H.-H. Chien, C.-L. Wang, W.-L. Chung, J.-R. Wu and E.-R. Chen (2010). "Intestinal parasitic infection detected by stool examination in foreign laborers in Kaohsiung." The Kaohsiung journal of medical sciences 26(3): 136-143.
- Inés, G. M., F. J. Basualdo, L. Kozubsky, M. Costas, R. E. Cueto and H. Lahitte (1996). "Intestinal parasites in two periurban populations in La Plata, Argentina]." <u>Boletín chileno de parasitología</u> **51**(1-2): 37.

- Ishida, M., G. Rubinsky-Elefant, A. Ferreira, S. Hoshino-Shimizu and A. Vaz (2003). "Helminth antigens Taenia solium, Taenia crassiceps, Toxocara canis, Schistosoma mansoni and Echinococcus granulosus and cross-reactivities in human infections and immunized animals." Acta Trop 89(1): 73-84.
- Jumaa, P. A. (2005). "Hand hygiene: simple and complex." Int J Infect Dis 9(1): 3-14.
- Kent, M. E. and F. Romanelli (2008). "Reexamining syphilis: an update on epidemiology, clinical manifestations, and management." The Annals of pharmacotherapy **42**(2): 226-236.
- Kritsiriwuthinan, K. and W. Ngrenngarmlert (2011). "Asymptomatic malaria infections among foreign migrant workers in Thailand." <u>Asian Pac J Trop Med 4(7): 560-563</u>.
- Kulkumthorn, M. and M. Surang Nuchprayoon (2006). "The prevalence of intestinal parasitic infections among school children in the central region of Thailand." J Med Assoc Thai 89(11): 1928-1933.
- Lam, S. K. (1998). "Emerging infectious diseases--Southeast Asia." <u>Emerging</u> infectious diseases **4**(2): 145.
- Lifson, A. R., D. Thai, A. O'Fallon, W. A. Mills and K. Hang (2002). "Prevalence of tuberculosis, hepatitis B virus, and intestinal parasitic infections among refugees to Minnesota." Public health reports 117(1): 69.
- Lopez, R., H. Huerga and M. Turrientes (2003). "Infectious diseases in immigrants from the perspective of a tropical medicine referral unit." <u>Am. J. Trop.</u>

  <u>Med. Hyg</u> **69**(1): 115-121.
- Maneeboonyang, W., C. Charusabha, S. Prommongkol, S. Puangsa-art, N. Thanyavanich and K. Buchachart (2004). "Status of intestinal helminth infections among Thai troops working along Thai-Myanmar border, Suanphung, Ratchaburi, Thailand." <u>J Trop Med Parasitol</u> 27: 59-64.
- Martin, P. L. (2004). <u>Thailand: improving the management of foreign workers</u>, ILO Geneva.

- Miller, J., H. Boyd, S. Ostrowski, S. Cookson, M. Parise, P. Gonzaga, D. Addiss, M. Wilson, P. Nguyen-Dinh and S. Wahlquist (2000). "Malaria, intestinal parasites, and schistosomiasis among Barawan Somali refugees resettling to the United States: a strategy to reduce morbidity and decrease the risk of imported infections." <a href="https://example.com/Am J Trop Med Hyg">Am J Trop Med Hyg</a> **62**(1): 115-121.
- Montresor, A., T. T. Zin, E. Padmasiri, H. Allen and L. Savioli (2004). "Soil-transmitted helminthiasis in Myanmar and approximate costs for countrywide control." <u>Tropical Medicine & International Health</u> **9**(9): 1012-1015.
- Nacher, M., P. Singhasivanon, S. Treeprasertsuk, U. Silamchamroon, W. Phumratanaprapin, A. Fontanet and S. Looareesuwan (2003). "Gender differences in the prevalences of human infection with intestinal helminths on the Thai–Burmese border." <u>Annals of Tropical Medicine and Parasitology</u> **97**(4): 433-435.
- Ngrenngarmlert, W. (2012). "Prevalence of Intestinal parasitic infection among Myanmar Workers in Bangkok and Samutsakorn." <u>Asia Pac J Public Health</u> **3**(july 2012): 53-58.
- Ngrenngarmlert, W., C. Lamom, S. Pasuralertsakul, R. Yaicharoen, N. Wongjindanon, S. Sripochang, T. Suwajeejarun, B.-O. Sermsart and R. Kiatfuengfoo (2007). "Intestinal parasitic infections among school children in Thailand." Trop Biomed 24: 83-88.
- Nuchprayoon, S., V. Sanprasert, S. Kaewzaithim and W. Saksirisampant (2009).

  "Screening for Intestinal Parasitic Infections Among Myanmar Migrant
  Workers in Thai Food Industry: A High-Risk Transmission." Journal of
  Immigrant and Minority Health 11(2): 115-121.
- Ohnmar, T.-T.-S. and S.-S. Ko-Ko-Zaw (2005). "Soe-Win. Household income, health and education in a rural area of Myanmar." <u>Southeast Asian J Trop Med Public Health</u> **36**(2): 529-533.
- Rianthavorn, P., et al. (2011). "Seroprevalence of Hepatitis A among Thai Population Residing Near Myanmar Border." <u>Journal of health, population, and nutrition</u> **29**(2): 174.

- Salas, S. D., R. Heifetz and E. Barrett-Connor (1990). "Intestinal parasites in Central American immigrants in the United States." <u>Archives of internal medicine</u> **150**(7): 1514.
- She, R. C., E. W. Taggart and C. A. Petti (2010). "Comparison of 10 indirect fluorescent antibodies to detect and type influenza A specimens." <u>Archives of pathology & laboratory medicine</u> **134**(8): 1177-1180.
- Singh, A., E. Houpt and W. A. Petri (2009). "Rapid diagnosis of intestinal parasitic protozoa, with a focus on Entamoeba histolytica." <u>Interdisciplinary perspectives on infectious diseases</u> **2009**.
- Soriano, S., L. Barbieri, N. Pierangeli, A. Giayetto, A. Manacorda, E. Castronovo, B. Pezzani, M. Minvielle and J. Basualdo (2001). "Intestinal parasites and the environment: frequency of intestinal parasites in children of Neuquén, Patagonia, Argentina." <a href="REVISTA LATINOAMERICANA DE MICROBIOLOGIA-MEXICO-43">REVISTA LATINOAMERICANA DE MICROBIOLOGIA-MEXICO-43</a>(2): 96-101.
- Stauffer, W. M., A. M. Newberry, C. P. Cartwright, J. E. Rosenblatt, K. L. Hanson, L. Sloan, D. T. Tsukayama, C. Taylor and B. A. Juni (2006). "Evaluation of malaria screening in newly arrived refugees to the United States by microscopy and rapid antigen capture enzyme assay." The Pediatric infectious disease journal 25(10): 948-950.
- Taylor-Robinson, D. (1988). "Immunological Diagnosis of Sexually Transmitted Diseases." Journal of Clinical Pathology **41**(5): 599.
- Thwing, J., J. Skarbinski, R. D. Newman, A. M. Barber, S. Mali, J. M. Roberts, L. Slutsker and P. M. Arguin (2007). "Malaria surveillance—United States, 2005." <a href="MMWR Surveill Summ">MMWR Surveill Summ</a> 56(6): 23-40.
- Triteeraprapab, S. and J. Songtrus (1999). "High prevalence of bancroftian filariasis in Myanmar-migrant workers: a study in Mae Sot district, Tak province, Thailand." <u>JOURNAL-MEDICAL ASSOCIATION OF THAILAND</u> **82**(7): 734-739.
- Versalovic, J. (2011). "Manual of clinical microbiology, American Society for Microbiology." Washington, DC.
- Wang, L.-C. (2004). "Changing patterns in intestinal parasitic infections among Southeast Asian laborers in Taiwan." <u>Parasitology research</u> **92**(1): 18-21.

- WHO (2011). "Global Tuberculosis Control." WHO report 2011.
- WHO. (2011). "Thailand GNI per capita, Atlas method (current US\$)." Retrieved 18 october 2012, from http://data.worldbank.org/country/, .
- Wong, T. and B. S. A. Yeoh (2003). <u>Fertility and the family: An overview of pronatalist population policies in Singapore</u>, Asian MetaCentre for Population and Sustainable Development Analysis Singapore.
- Wyler, D. J., G. Mandell, R. Douglas Jr and J. Bennett (1990). "Plasmodium species (malaria)." <u>Principles and practice of infectious diseases.</u>: 2056-2066.
- YOON, S. N., C. PORKSAKORN and I. N. YOON (2002). "Prevalence of parasitic infections among Thai patients at the King Chulalongkorn Memorial Hospital, Bangkok, Thailand." <u>J Med Assoc Thai</u> **85**: S415-423.

### **APPENDICES**

## APPENDIX A DEFINITIONS

**Age group** is defined as adults (age ≥18 years until 60 years) **Immigrant** is defined as people who move into another country or region to which they are not native in order to settle there with any kind of purposes **Prevalence** is defined as the proportion of a population found to have a condition (typically a disease or a risk factor such as smoking or obesity or hygienic factor) **Tropical Diseases** is defined as diseases that are mainly of parasitic origin and are common in tropical or subtropical areas (between the latitudinal lines of the tropic of Cancer (23.5N) and the tropic of Capricorn (23.5S))

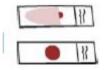
# APPENDIX B THICK BLOOD SMEARS FOR MALARIA INFECTION AND FILARIASIS

### **Blood film examination**

1) Thick film: the drop of blood is spread by using the corner of a clean slide in a circle the size of a dime (diameter 1-2 cm).



2) We wait until the thick films are completely dry before staining.

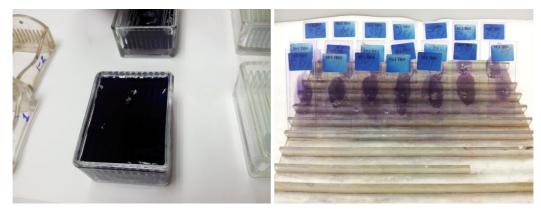


3) For Giemsa 3% solution preparation, 97 mL of buffered water (ph 7.2) was poured into a 100 mL graduated cylinder. Using Pasteur pipette, 3 mL of Giemsa stain was added to the graduated cylinder in order to make 3% concentration. The top of the graduated cylinder was covered and inverted several times until completely mixed.





4) The smears were allowed to dry quickly by application a fan or blower at room temperature. Thick blood film was placed in a staining rack and stain singly, or in batches in staining jar to avoid cross-contamination. Films were stained for 30-45 minutes out of the sunlight. Clean tap water was poured gently into the trough to float off the iridescent scum on the surface of the stain. Then slides were rinsed briefly and gently under running tap water.



5) The stained slides was placed in a drying rack to drain and dry, film side in downwards position, it would make sure the film did not touch the slide rack. During preparation and storage time, slides should be protected from exposure to dust and insects.



### **APPENDIX C**

### RECTAL SWAB EXAMINATION FOR PARASITE INFECTION

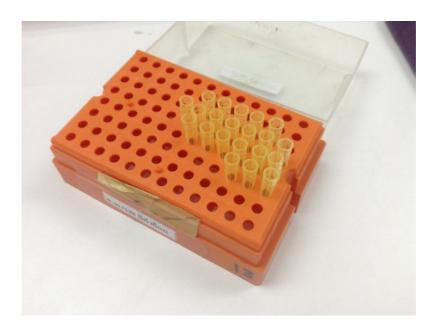
1. Rectal swab samples were sent to Faculty of Tropical Medicine for examination. If delay for examination, the rectal swab samples were stored in cool temperature room in order to minimize the risk of fungal contamination.



2. Each rectal swab sample specimen was separated from the agar by using straw, then additional 0.05 milliliter normal saline (with micropipette) mixed in small tube (0.5) before proceeding to centrifuge methods at 2000 rpm for 5 minutes.



3. After centrifuge, the supernatant was separated with micropipette and the sedimentation was taken by another new micropipette to slide (object glass).



- 4. Every sample was examined with 2 routine methods, with normal saline and iodine solution.
- 5. One drop of normal saline solution was put in the middle of the left half of the slide and one drop of *Lugol's* iodine in the middle of the right slide.

- 6. The drop of normal saline on the left half of the slide should be mix first, continue with right iodine part.
- 7. Then, cover slips were added to each left for normal saline and right for iodine part with slowly from the side to avoid bubbles.
- 8. Entire cover slip was ready to be examined. For the saline preparation, the 10x objective and the 40x objective were for helminthic infection used, and for the Lugol's iodine preparation were used the 40x objective.

# APPENDIX D CASE RECORD FORM

Data collection sheet – 1				
Study code: SKH-				
Date of visit: (dd/mmm/yy	/yy)/_	/		
Demographic data:				
Date of birth: (dd/mmm/yy	yyy)//_		No data	
Age: Years	_Months			
Sex :				
(1) Male (2) Female				
Ethnicity:				
$(1) Myanmar \qquad (2)K$	Carean (3) Lao(4	4) Mon	(5) Cambod	ian
(6) Others, specify				
Duratian of staying in Sam	ut sakhonSamut sal	kon:		
(1) Less than 1 month (2) 1	-6 month (3) 6mor	nth – 1 year		
(4) 1 year – 5 year (5)	more than 5 year, s	pecifyy	rs	
Underlying disease:   N	No Yes, spe	ecify		
Weight:kgs Heig	ght:cms E	BMI:	_	
Temperature: °C l	Blood Pressure:	mmHg	Pulse rate:	/min
Physical Examination	(1) Normal	(2) Abnorma	ıl	(9)No data
HEENT				
Cardiovascular				
Lung				
Abdomen				
SKIN				
EXT				

Laboratory Result		(9)No data
Blood smear	Thick smear: positive / negative	
Chest X-ray	Normal Abnormal, Specify:	
VDRL	Reactive / non-reactive	
Filaria	Thick smear:	
Stool Examination	Intestinal helmint: Intestinal protozoa:	

### **BIOGRAPHY**

NAME Muhammad Luthfi Almanfaluthi

**NATIONALITY** Indonesia

**DATE OF BIRTH** 19 May 1984

PLACE OF BIRTH Malang, Indonesia

**EDUCATION** Brawijaya University, Indonesia 2002 – 2008

Doctor of Medicine

Mahidol University, Thailand 2012

Diploma (DTM&H)

**HOME ADDRESS** Permata Hijau A 17 RT 3 RW 9

Tlogomas, Malang, East Java, Indonesia

Tel: (+66) 572223

Email: lutfi\_dr\_1984@yahoo.co.id