Meeting Report

Laboratory Training and Risk Management Workshop

6-10 July 2015

Mahidol University, Amnatcharoen Campus

under project

Reducing Biosecurity Threats from Infectious Diseases of Pandemic Potential in Southeast Asia

Contents

Summary

The project entitled “Reducing Biosecurity Threats from Infectious Diseases of Pandemic Potential in Southeast Asia” is jointly cooperated by 3 networks (1) Asia Partnership in Emerging Infectious Diseases Research (APEIR), (2) Mekong Basin Disease Surveillance Network (MBDS) and (3) ASEAN+3 Field Epidemiology Training Network (FETN). As the leader in research, APEIR realized the impact of emerging infectious diseases caused by dangerous pathogens. After grant reallocation, APEIR researchers resumed the project process and conducted “Laboratory Training and Risk Management Workshop”. In addition to meet the project’s goal which is to build laboratory capacity to handle dangerous pathogens, this workshop was set up to share knowledge on potential pathogens.

The project was proposed to implement in 4 sites including Thailand –Laos–Vietnam (Mukdahan–Savannakhet–Quang Tri), Lao PDR–Thailand (Champasack–Ubon Ratchathani), China–Laos (Mengla–Luang Namtha) and Thailand–Cambodia (Sakaeo–Banteay Meanchey), however, only the largest site was selected at inception meeting (11-12 September 2014 at Savannakhet, Lao PDR). In the meeting, all research partners also agreed that laboratory training is necessary to practice the protocol indicated in SOP or revise based on all researchers’ agreement. Around the same time, potential infectious diseases other than targeted diseases e.g. EBOLA virus and MERS-CoV drew the interested worldwide. It is the good opportunity to prepare healthcare professionals for these EID and therefore situation update and management protocol were included in the workshop.

Group photo of workshop participants

The workshop is organized at the Mahidol University, Amnatcharone campus, near East_West Corridor with participants from 4 countries, Cambodia, Laos, Thailand and Vietnam, including the members of both APEIR and MBDS networks. During the event, knowledge sharing of updated potential risks was discussed. Dr. Moe Oo Ko, MBDS secretariat, updated MBDS network’s work while Dr. Chakrarat Pittayawonganon, Director of Thai FETP (Field epidemiology training program, shared FETN’s works and the interesting process in MERS-CoV first-case investigation in Thailand. Thai APEIR researchers also update the information of potential pathogens including influenza, virus, MERS-CoV, other corona viruses, *Salmonella*, *Burkholderia pseudomallei*, *Mycobacterium tuberculosis.* As being in genomic era, microbiome and its application was also updated by Dr. Pravech Ajawatanawong-the invited speaker from Mahidol University.

In addition to practice laboratory protocol, participants learned how to manage and prepare for potential diseases. This represent the readiness of the project to move forward our research project and also provide sustainability knowledge on disease control.

Laboratory Training

Laboratory training is essential for dealing with potential microorganisms. The researchers/laboratory technicians were invited from study sites including Lao PDR (Vientiane and Savannakhet) and Vietnam (Hanoi and Quangtri). Even though Cambodia will not be an implemented site of the project, the researchers were also invited as trainees. Laboratory technicians from Mukdahan and Amnatcharoen, nearby areas of the study sites in Thailand, also got a chance to improve their capacity in handling targeted organisms as well as general organisms.

Subproject influenza

Sample collection from live and dead poultry was demonstrated by Assist. Prof. Dr. Witthawat Wiriyarat, who is currently responsible for screening influenza in wildlife in Thailand. Trainees practiced sample processing procedure in biosafety cabinet (BSC). They all also practiced sample inoculation into eggs, which was the critical step for viral isolation. Inoculated eggs were observed daily. Positive result was prepared beforehand with non-pathogenic virus for the sake of demonstration. Viral harvesting and hemagglutination were performed by trainees. RNA was extracted and subjected to RT-PCR.

Subproject *B. pseudomallei*

Soil sample collection was done following the published method at nearby area of the University. The soil sampling site was divided into a 2x 2 grid of 1 m x 1 m square. Global Positioning System (GPS) receiver and ArcGIS software were used to record and map the location of sampling. Samples at the depth of 30 cm were collected, weighed and processed. Similar to influenza virus, isolation and identification of *B. pseudomallei* have to perform using BSC. Prepared *B. thailandensis*, a closely related non-pathogenic species culture was used for subsequent training. Trainees observed colonies and biochemical test for *B. pseudomallei.* Multilocus Sequence Typing (MLST) was planned together with research partners with the aim to reveal epidemiology of the bacteria among countries.

Subproject *salmonella*

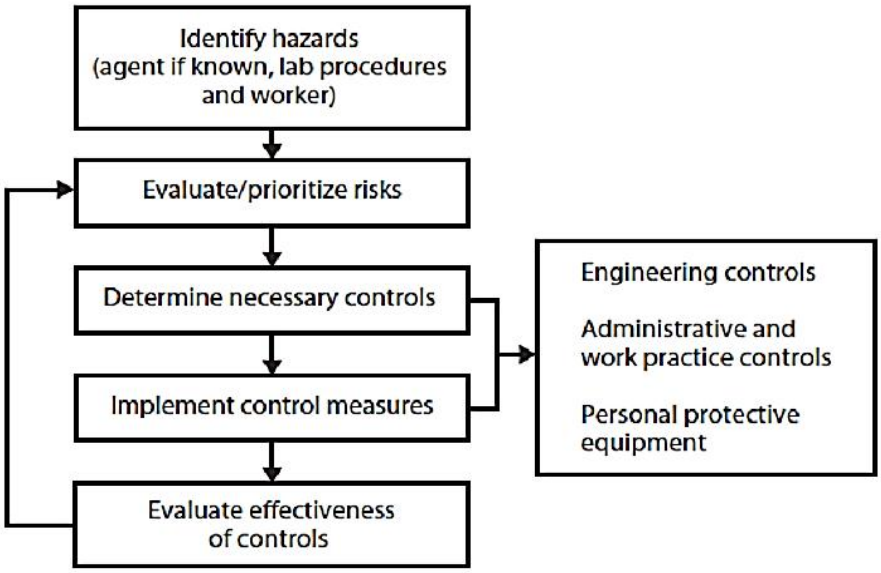
Samples were collected from poultry similar to subproject influenza. Bacterial culture and identification was performed using standard method. Molecular method was used to confirm bacterial species. Antibiotic susceptibility testing was determined as drug-resistant *Salmonella* causes

problems worldwide. The protocol of MLST typing is also discussed as *Salmonella* and its resistance are commonly food-associated in the regions and have potential to spread across borders following the contaminated food.

Researchers from Lao PDR, Vietnam and Thailand discussed in details on each subproject. All three diseases remained focused and the main contact person in each step were clarified. The SOP of the procedure for the detection of highly pathogenic avian influenza viruses, H5N1 and H7N9, is added. Positive control for molecular detection will be shared among countries to ensure that no bias results from any country. Vietnamese and Thai teams possess the ability to perform all laboratory procedures. Lao team is able to process some parts; however, they are glad to support the project in every aspect.

Training on Risk management for laboratory handling of dangerous pathogens

Biohazard materials are biological organisms or materials produced that potentially hazardous to humans, animals or the environment. To restrict biohazard, biosafety risk management, which is the occupational safety action is needed to implement and maintain. Risk management is the application of knowledge, techniques and equipment to prevent personal, laboratory and environmental exposure to potentially infectious agents or biohazards. Key steps of risk assessment are 1) determination of hazards and evaluate risks, 2) using all relevant available data and 3) determining controls needed to minimise the risks (Flowchart). On the other hand, biohazard is also needed to evaluate i.e. what the risk handled is, what the potential consequences are, how the people exposed are and what the necessary controls are.



General process of risk management

Biosafety risk management require the combinations of three main components that are containment facilities, personal protection device and good standard operation protocols. Different containments, appropriate with various levels of risks are categorized into 4 Biosafety Levels (BSLs) including BLS1 – for agents not known to cause any diseases, BSL2 – for agents associated with human disease, BSL3 - indigenous/exotic agents associated with serious human disease and with potential for aerosol transmission and BSL4 -dangerous/exotic agents of life threatening nature and without adequate therapeutic measures. The need for working in each containment facility depends on the natures of the pathogens, the amount of biohazard materials, the required procedures and the effectiveness of available personal protection equipment.

Personal Protective Equipment (PPE) is required for potential infectious disease exposure. Various microorganisms are different in virulence and spreading ability. PPE are various in types and needed to use properly in combination with biocontainment facility. If needed, all parts of body can be protected by different PPE including gloves -protect hands; gowns/aprons – protect skin and/or clothing; masks and respirators– protect mouth/nose; respirators – protect respiratory tract from airborne infectious agents; goggles – protect eyes, face shields – protect face, mouth, nose, and eyes; cap - used to contain hair; shoes – protect feet. To be completely protected, key points of using are needed to concern i.e. don before contact with the microorganisms, don generally before entering the laboratory, use carefully and avoid spread contamination, remove and discard carefully at the doorway or immediately outside the laboratory and wash hands immediately after use.

Updates on emerging infectious disease (EIDs)

As the first case of MERS-CoV infection has just been confirmed in Thailand, update information on this virus and the experience to handle its victims and on other high potential viruses is essential for healthcare and public health workers.

Respiratory tract infection caused by impact viruses draw global attention as their spreading are quite rapid. Animals are revealed to play role as a viral reservoir i.e. wild aquatic birds and bats are thought to be the reservoir for many virus families in nature including influenza and Nipah viruses while camel is thought to serve as reservoir for MERS-CoV. Thus, the observation of these animal migration should be performed to (at least) alert public health system in prevention and control.

Tuberculosis (TB), although has emerged for a long time, its resistance to most therapeutic drug is the great concern. New TB cases worldwide are ca. 9,000,000 cases (WHO, 2014), however, 136,000 cases of MDR-TB were detected and only 97,000 got treatment. As *M. tuberculosis* has ability in immune system evasion, some infected individuals may not apparently show off -so called Latent TB. These individuals may progress to active TB. The more concern is that more MDR cases may results from the growing economy and migration of the region.

Update on molecular approaches for biohazard risk management

In order to specify the chain of transmission of dangerous pathogens, they can be categorised in a subspecies levels. Microorganisms can be classified by “phenotyping” or “genotyping”. A common phenotyping method is based on drug resistance patterns. However, some phenotypes, particularly the drug resistance, may be a result of horizontal gene transfer, which can occur crossing species. Thus, they may not be epidemiologically useful. The use of genotyping in the field of epidemiology and public health becomes more popular e.g. defining an outbreak in endemic areas and consequently define the settings that the transmissions occur, identify hospital-acquired infections, defining/identify occupational hazard or sources of infections of health personnel, identify the origins emerging infectious diseases and prognosis prediction via virulence identification.

Omics approach is a high-throughput, data-driven, holistic and top-down methods to integrated study microorganisms. The approach requires bioinformatics to analyse and manipulate the data. The information allow people to understand the set of genes expressing in a particular condition that facilitate more understanding of microorganism related to human. In addition, omics data utilization is advantage for various fields such as narrowing spectrum of antibiotics based on microbiome data and developing true probiotics based on gut microbiota information.

Networks sharing

Dr. Chakkrarat Pittayawonganon, Director of FETP-Thailand, shared FETN's roles in enhancement of region capacity for high potential pathogen such as EBOLA virus infection. He also shared experience in dealing with MERS-CoV. The information about the case was told form the time of suspect, during investigation and after confirmation. The limitation revealed to be the laboratory test. There were confusing results at first but finally the case was confirmed and the protocol to quarantine were applied to control disease spreading. With well cooperation in surveillance, no more case found in Thailand. This MERS-CoV case provides lesson learn for disease investigation and control. Also, the situation reminded us to prepare for any risk that may occur in our region.

Dr. Moe Oo Ko, MBDS secretariat, update the network's work on disease surveillance. There are 5 member countries, including Myanmar, China, Cambodia, Vietnam and Thailand, setting up as partners in reporting disease information. The report schedule of exchange depends on the diseases of interest- reporting within 24 hours of diagnosis: H1N1/H5N1, AFP (acute flaccid paralysis), SARS, cholera/ severe diarrhea, encephalitis, tetanus, meningitis, diphtheria and other public health emergency of international concern (PHEIC); reporting weekly: leptospirosis, chikungunya, dengue fever, typhoid fever and measles; reporting monthly: malaria and pneumonia; report quarterly: HIV/AIDS and tuberculosis.

Mobile Application Training

The workshop served a stage for mobile application training for MBDS network. The application was developed by Dr. Ekkarat Boonchieng, Chiang Mai University Thailand to share and report the news related to Biothreat Surveillance. With mobile phone and tablet, this tool will facilitate evidence-based surveillance. Reporters are allowed to share the photo indicating infectious disease occurrence via the application that will alert members to prevent or aware of the disease. However, the detail of report still needed to discuss among members.

Discussions on current and further collaboration between MBDS, FETN and APEIR

Since three networks gathered, fruitful discussion on future collaboration was done. Several MBDS members attended the risk management workshops. Many of them were from bordering towns and eager to explore further collaboration in disease surveillance and research on cross-border transmission of infectious diseases. APEIR will facilitate the research originated from MBDS member needs and interests. On the other hand, FETN works on new emerging outbreaks which are unlikely to be in current APEIR projects. However, APEIR will try to organize research that will facilitate FETN works. According to the discussion, some proposed projects are as following:

- Rabies: the project has been currently funded but would be ended soon
- Toilet practices of cross border road travelers: impact on health (such as parasitic dissemination)
- Cross-border tuberculosis transmission
- Food safety issues, such as illegal transport of meat products across the borders (Thai chicken to Laos and residual agricultural chemicals)
- Rapid cross border disease screening methodology
- Cross border medical care
- Other diseases such as dengue fever, leptospirosis, rickettsial diseases, etc

Annex

Participant

Attendees

No.	Participant	Affiliation	Country
1	Dr.Hansa Taesiri, M.D. hansa.tae@mahidol.ac.th	Vice President, Mahidol University, Amnatcharoen	Thailand
2	Dr. Paskorn Chaiyaseth, M.D.	Provincial Chief Medical Officer, Amnatcharoen	Thailand
	Speakers/ Trainers /Mahidol Staffs		
3	Prof. Prasit Palittapongarnpim prasit.pal@mahidol.ac.th	Head of Department of Microbiology, Mahidol University	Thailand
4	Dr. Chakrarat Pittayawonganon c.pittayawonganon@gmail.com	Director of International FETP-Thailand, Bureau of Epidemiology, Department of Disease Control	Thailand
5	Dr. Moe Oo Ko moekooo2003@yahoo.com	MBDS Secretariat	Thailand
6	Dr. Pravech Ajawatanawong pravech.aja@mahidol.ac.th	Lecturer, Mahidol University	Thailand
7	Assist. Prof. Arunee Thitithanyanont arunee.thi@mahidol.ac.th	Department of Microbiology, Mahidol University	Thailand
8	Assist. Prof. Witthawat Wiriyarat wiriyarat@yahoo.com	Deputy Dean for Research, Faculty of Veterinary Sciences, Mahidol University	Thailand
9	Assist. Prof. Soraya Chaturongkakul soraya.cha@mahidol.ac.th	Department of Microbiology, Mahidol University	Thailand
10	Dr. Kamolchanok Rukseree kamolchanok.ruk@mahidol.ac.th	Lecturer, Mahidol University, Amnatcharoen	Thailand
11	Dr. Sirirak Supa-Amornkul pung_654@hotmail.com	Lecturer, International school of dentistry, Mahidol University	Thailand
12	Ms. Suwimon Wiboon-ut swiboonut@gmail.com	Research Assistant, Department of Microbiology, Mahidol University	Thailand
13	Ms. Phajongjit Karraphan aom_kat77@hotmail.com	Research Assistant, Mahidol University, Amnatcharoen	Thailand
14	Dr. Arunee Thong-on arunee.tho@mahidol.ac.th	Lecturer, Mahidol University, Amnatcharoen	Thailand
15	Dr. Wongwarut Boonyanugomol wongwarutb@gmail.com	Lecturer, Mahidol University, Amnatcharoen	Thailand
16	Dr. Thidarat Netikul t_netikul@hotmail.com	Lecturer, Siam University	Thailand
	Laboratory Trainees		
17	Ms. Samran Lekngam	Technician, Amnatcharoen Provincial Health Department	Thailand
18	Ms. Prapawee Bandasak	Technician, Amnatcharoen Provincial Health Department	Thailand
19	Miss Orawan Ottiwet	Technician, Mukdahan Provincial Health Department	Thailand

No.	Participant	Affiliation	Country
20	Ms.Daovieng Vanthady daovieng.daao9@gmail.com	Technical staff Surveillance Division, Department of Communicable Disease Control	Lao PDR
21	Ms.Manivanh Vongsouvath manivanhvsv@hotmail.com	Deputy Head of Microbiology Unit, Mahosot Hospital, Vientiane	Lao PDR
22	Mr. Chanhthalom Southammavong shmavong.cs@gmail.com	Technical staff, National Animal Health Laboratory, Department of Livestock and fishery, Vientiane	Lao PDR
23	Dr. Vatsana Sopraseuth vat.sana@hotmail.com	Head of Laboratory Section, Savannakhet Hospital	Lao PDR
24	Mrs. Somneuk Keokhounying	Technician, Savannakhet Hospital	Lao PDR
25	Dr. Tran Huy Thinh huythinhda@yahoo.com, tranhuythinh@hmu.edu.vn	Deputy Head, Institute for Preventive Medicine and Public Health	Vietnam
26	Mr. Do Nam Khanh khanhdonam@gmail.com	Lecturer,Institute for Preventive Medicine and Public Health, Hanoi Medical University, Hanoi	Vietnam
27	Mr. Do Duy Thang duythangdo@gmail.com	Researcher, Central Laboratory, Institute for Preventive Medicine and Public Health, Hanoi Medical University, Hanoi	Vietnam
28	Dr. Teng Srey tengsrey72@gmail.com	Deputy-Director of Communicable Disease Control Department	Cambodia
29	Mr. Chin Savuth Savuth_Chin@niph.org.kh	Head of Virology unit, National Public Health Laboratory/National Institute of Public Health	Cambodia
MBDS			
30	Dr. Ekkarat Boonchieng ekkarat@boonchieng.net	Lecturer, Chiang Mai University	Thailand
31	Mrs. Punchawee Sukbut mbdsmuk@yahoo.co.th	Associate Province Coordinator, Senior Technical Health Office, Mukdahan Health Office	Thailand
32	Mr. Somkiat Thonglek	Chief of Epidemiology Sector, Sakaeo Provincial Health Office	Thailand
33	Mrs. Waleerat Aphaibunditkul	Public Health Technical Officer, Assistant Chief of Disease Control Group, Ubon Ratchathani Provincial Health Office	Thailand
34	Mr. Thapon Tiawsirichaisakul	Technical Officer, Nongkhai Provincial Health Office	Thailand
35	Mr. Narin Suriyon	Chiang Rai Provincial Public Health Office,	Thailand
36	Ms. Jittra Thajeen jittra.thajeen@gmail.com	Project Assistant, MBDS Secretariat Office	Thailand
37	Ms. Thin Mar Soe tmarsoe@gmail.com	ICT Assistant, MBDS Secretariat Office	Thailand
38	Mr. Kolakanh Phichitchay pkolakanh@yahoo.com	Deputy of Epidemiology, MBDS Coordinator, Savannakhet Health Department	Lao PDR

No.	Participant	Affiliation	Country
39	Dr. Chanthalay Sayavong sengthong_2006@yahoo.com	Head of Epidemiology Division, Vientiane Capital City	Lao PDR
40	Mrs. Ketchanh Sysavat ketchanh@gmail.com	Provincial MBDS Coordinator, Epidemiology Unit, Bokaeo province	Lao PDR
41	Mr. Le Huu Le lehuule2007@yahoo.com	Deputy Director, Quang Tri Provincial Department of Health	Vietnam
42	Dr. Keo Pechsovann dr_kpsovann@yahoo.com	PHD Site Coordinator, Banteay Meanchey province	Cambodia
Dictyostelium Team from Department of Microbiology, Mahidol University			
43	Ms. Kulawadee Wonganun	Master Student, Department of Microbiology, Mahidol University	Thailand
44	Mr. Pitayapol Rattanaloed	Master Student, Department of Microbiology, Mahidol University	Thailand
45	Mr. Phongsathon Yodsuwan	Intern Student, Department of Microbiology, Mahidol University	Thailand
46	Mr. Thanakun Kanyapornkun	Intern Student, Department of Microbiology, Mahidol University	Thailand
47	Mr. Chalermkiet Pumeim	Intern Student, Department of Microbiology, Mahidol University	Thailand
48	Ms. Panawun Palittapongarnpim panawunp@gmail.com	Research Assistant, Department of Microbiology, Mahidol University	Thailand
49	Mr. Isaac Pelagio Chomitz	Visiting student, Wisconsin University	Thailand
50	Ms. Marisol Wolf	Visiting student, Wisconsin University	Thailand