CAPACITY BUILDING AND NETWORKING OF LABORATORY PROFESSIONALS FOR ZOONOTIC DISEASE CONTROL: INTENSIVE TRAINING IN IDENTIFICATION OF PATHOGENIC BACTERIA

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Bacteria are common causes of zoonotic infections. In Southeast Asia, *Streptococcus suis* infection is a leading cause of death, blindness and hearing loss among infected cases. *S. suis* can be transmitted to humans through close contact with sick or carrier pigs, and by consuming raw or uncooked pork. In Thailand there have been sporadic outbreaks of *S. suis* infection in humans, the first report being in 1987, followed by a series of outbreaks in the north and northeast regions during 1997-2012. Many zoonotic gram-negative bacteria are maintained in human and animal reservoirs and contaminate food supply via excreta causing outbreaks of food-borne illness, e.g. Salmonella.

Although laboratory skills in the detection of bacterial zoonosis have been widely imparted in government sectors under the supervision of the Ministry of Public Health, analysis by THOHUN NCO in Year 1 of gaps and needs revealed laboratory skills are still required as perceived by health professionals of Thailand One Health Workforce. Faculty staff of medical, public health and veterinary schools serve an important role in preparing future workforces for rapid identification of pathogens and reservoirs in mitigating foodborne outbreaks. Therefore, maintaining and improving the One Health Core Competency (OHCC) and laboratory skills of these faculty staff members are crucial for their professional career.

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Objective 3: Education and In-Service Training of Health Officers, Practitioners and Professionals (Current Workforce)

The intensive training "Networking and capacity building of biomedical personnel in Southeast Asia on clinical laboratory identification of bacteria causing zoonosis and foodborne illness" focused on enhancing OHCC, previous knowledge and laboratory skills for early detection and bacterial identification using standard and advanced techniques. This 2 week hands-on workshop aimed to strengthen faculty staff capacity for effective detection of bacterial zoonosis as well as to create a professional network among professionals from Malaysia, Myanmar, Thailand and Vietnam to ensure sustainable future workforce development. The expected outcomes were that all participants have (1) knowledge regarding One Health strategy, (2) skills and know how in the identification of zoonotic bacteria, food-borne bacteria and others clinically-relevant bacteria, (3) acquired soft skills, and (4) established laboratory networking.

In the first week, on day one, a lecture was delivered in the morning on the One Health strategy and how One Health links to one's health. In the afternoon, participants were informed on zoonotic and specimen handling. On days two to five, participants spent 1.15-1.30 hours on soft skill training in leadership, positive thinking, collaboration and team building, systems thinking and mind mapping, communication and informatics, analytical thinking, communication, and work planning and team working using a number of activities, e.g. NASA moon model, Marshmallow challenge, Blind fold, video presentation and lectures on the concept underlying each skill and how to make summaries of each activity learnt. Then, current knowledge was taught regarding clinically important and zoonotic gram-positive cocci, viz. *Enterococcus* and gram-positive bacilli (*Bacillus, Corynebacterium, Gardnerella, Listeria, Erysipelothrix,* and *Clostridium*), *Streptococcus* and *Staphylococcus,* The lecture also reviewed the properties of zoonotic *S. suis.* This was followed by lectures on biochemical identification of gram-positive bacterial species and a review of antimicrobial resistance in gram-positive bacteria and molecular methods in bacteria detection and identification.

In the laboratory training section, participants were required to identify two unknown gram-positive bacterial samples using techniques starting from streak plating, selecting colonies for staining, selecting colonies for inoculating biochemical test media, and selecting colonies for drug susceptibility testing. One sample was ***S. suis*** and the other gram-positive bacteria closely resembling ***S. suis*** or other clinically important bacteria, e.g. *Streptococcus pneumoniae, Streptococus pyogenes, Streptococcus agalactiae, Streptococcus bovis, Enterococcus faecalis, Enterococcus faecium, Staphylococcus aureus, Staphylococcus haemolyticus,* and *Listeria monocytogenes.* Participants were

required to up date their progress daily and to supply a detailed history regarding the source and origin of their samples based on knowledge from the lectures. Daily questions posed to participants on their laboratory results for Sample 1, which was cultured from blood culture specimen of patient presenting fever, nausea and headache, and had a history of drinking alcohol, were such as (i) on day 1: what is/are the organism(s) in your mind after reading the history of the sample? (ii) on day 2: from the colony morphology and Gram staining, can you guess this organism might be? And (iii) on day 3-4: after identification, what is/are biochemical test(s) you wish to perform?

After all the identification results were known together with their histories, a group discussion was held on such topics as pitfall of each test that would give rise to a false positive or negative result. For instance, cooling the sterile loop by stabbing in blood agar before picking up a colony for subsequent test might produce a false positive result in a catalase test because the blood in blood agar might be the cause of the bubbles indicative of a positive catalase reaction.

In the second week, in the mornings of day 1 to day 3, participants were divided into three groups and each group spent 1.15-1.30 hours discussing the issue of "Capacity building and laboratory professional networking for zoonotic disease control". The following three case studies were presented for discussion.

Case 1. One village in Mae Chaem district (150 km from Chiang Mai) held a special event after the rice harvest, a tradition of this village. They have a special dish known as "Lue" (northern Thai dish using fresh pig blood). After the party, seven men, 35-65 years of age, came down with fever and meningitis from day 1 to day 7. The men said they bought two pigs from a market in Mae Chaem and two more from Chom Thong district (close to Mae Chaem). The patients were admitted to Chom Thong Hospital and referred to Nakhon Ping Hospital (a Chiang Mai central hospital) where CSF and blood samples were cultured and the isolates sent to the Division of Clinical Microbiology, Faculty of Associated Medical Sciences, Chiang Mai University for identification by standard methods and confirmation by PCR. All isolates were identified as *Streptococcus suis* type II. Unfortunately, two patients died, four suffered hearing loss and one lost both hearing and sight.

Discussion topic. Control measures against spread of *S. suis*.

Daily questions were posed for discussion employing the soft skills learnt during the first week followed by presentation.

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Questions on day 1. (a) Who is/are the stakeholder(s) involved in the control of an epidemic of *S. sui*? (b) How are they involved?

Questions on day 2. (a) What are the responsibilities of each stakeholder involved in the control an epidemic of *S. suis*? (b) What steps should be taken to set up network(s) or working group(s) to control an epidemic of *S. suis*? (c) Who will act as leader of each network or working group?

Questions on day 3. In a network or working group involved in laboratory investigations: (a) Who will act as coordinator or leader? (b) How will collaboration be set up? (c) What is/are activity(ies) that the network or working group should do to serve members?

Case 2. A patient presented with a wound in the left hand and high fever. Pus and blood from the wound were sent for culture before being prescribed the antibiotic ceftriazone. Three days later, laboratory result indicated methicillin-resistant *Staphylococcus aureus* (MRSA). The patient worked at a pig farm and received the wound during the course of his work.

Discussion topic. Control of a MRSA epidemic.

Daily questions were posed for discussion employing the soft skills learnt during the first week followed by presentation.

Questions on day 1. (a) Who is/are stakeholder(s) involved in the control of a MRSA epidemic? (b) How are they involved?

Questions on day 2. (a) What are the responsibilities of each stakeholder involved in the control a MRSA epidemic? (b) What steps should be taken to set up network(s) or working group(s) to control a MRSA epidemic? (c) Who will act as leader of each network or working group?

Questions on day 3. In a network or working group involved in laboratory investigations: (a) Who will act as coordinator or leader? (b) How will collaboration be set up? (c) What is/are activity(ies) that the network or working group should do to serve members?

Case 3. A patient presented with fever, abdominal cramp and diarrhea. The symptoms began 24 hours after consuming a soft-boiled egg from his farm. Stool and blood were sent for culture before being prescribed an antibiotic. After three days, laboratory report indicated the presence of *Salmonella* Enteritidis. The patient had about 1,000 hens, which produce 700-900 eggs per day.

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Discussion topic. Control of an epidemic of *Salmonella* Enteritidis.

Daily questions were posed for discussion employing the soft skills learnt during the first week followed by presentation.

Questions on day 1. (a) Who is/are stakeholder(s) involved in the control of an epidemic of *Salmonella* Enteritidis? (b) How are they involved?

Questions on day 2. (a) What are the responsibilities of each stakeholder involved in the control an epidemic of *Salmonella* Enteritidis? (b) What steps should be taken to set up network(s) or working group(s) to control an epidemic of *Salmonella* Enteritidis? (c) Who will act as leader of each network or working group?

Questions on day 3. In a network or working group involved in laboratory investigations: (a) Who will act as coordinator or leader? (b) How will collaboration be set up? (c) What is/are activity(ies) that the network or working group should do to serve members?

At the completion of each daily discussion topic, each group presented their conclusions. On day 4, each group summarized their conclusions regarding the feasibility of establishing a laboratory network for the control of an epidemic of their respective zoonotic bacterial pathogen.

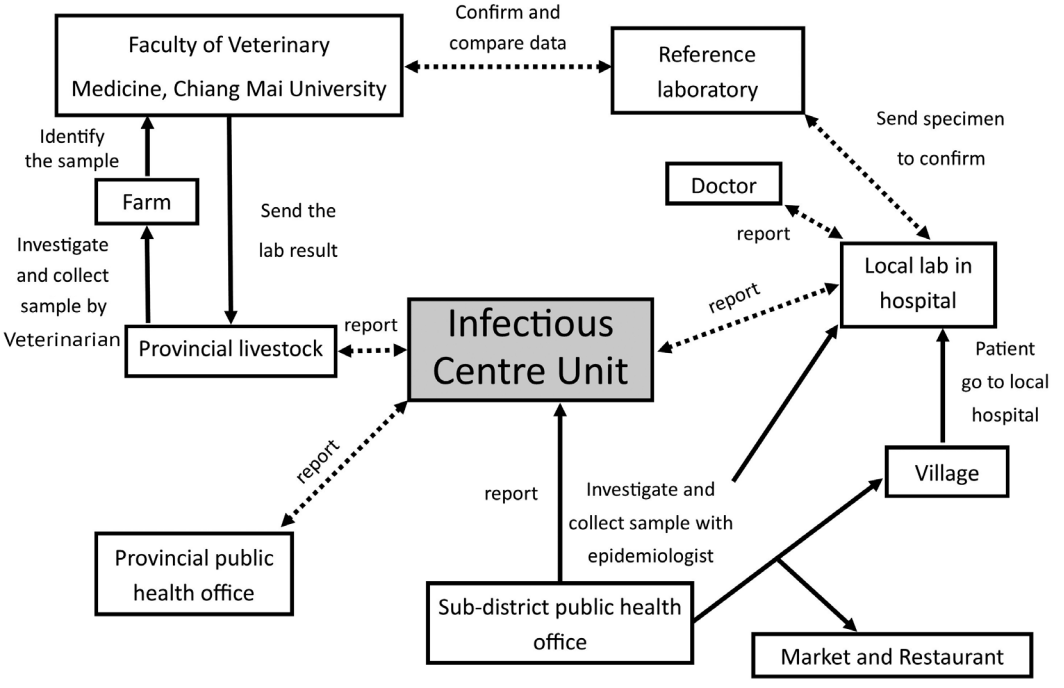
During this period of the training, lectures focused on the clinical importance of zoonotic gram-negative bacteria, such as *Bartonella* especially in family Enterobacteriaceae, e.g. *Eschericial coli, Salmonella, Shigella,* and other coliform group, *Brucella,* and *Francisella*. Lectures also included clinical importance and identification of *Aeromonas, Mycobacterium tuberculosis, Plesiomonas,* and *Vibrio*. There were lectures on drug resistance of gram-negative bacilli, causes and control.

Laboratory training sessions on identification of pathogenic gram-negative bacteria were conducted using the same format as in the first week. One sample was a salmonella specimen (*Salmonella* Choleraesuis, *Salmonella* Enteritidis, *Salmonella* Paratyphi A, or *Salmonella* Typhi) and the other one of the following gram-negative bacilli: *Aeromonas hydrophila, Enterobacter cloacae, Escherichia coli, Klebsiella pneumoniae, Pasteurella multocida, Shigella flexneri, Shigella sonnei, Vibrio cholerae,* and *Vibrio parahaemolyticus.*

A typical power point presentation point presentation from each group is shown in Fig 1.

Group 1. Control of *Streptococcus suis* epidemic.

Involvement of Stakeholder or network.



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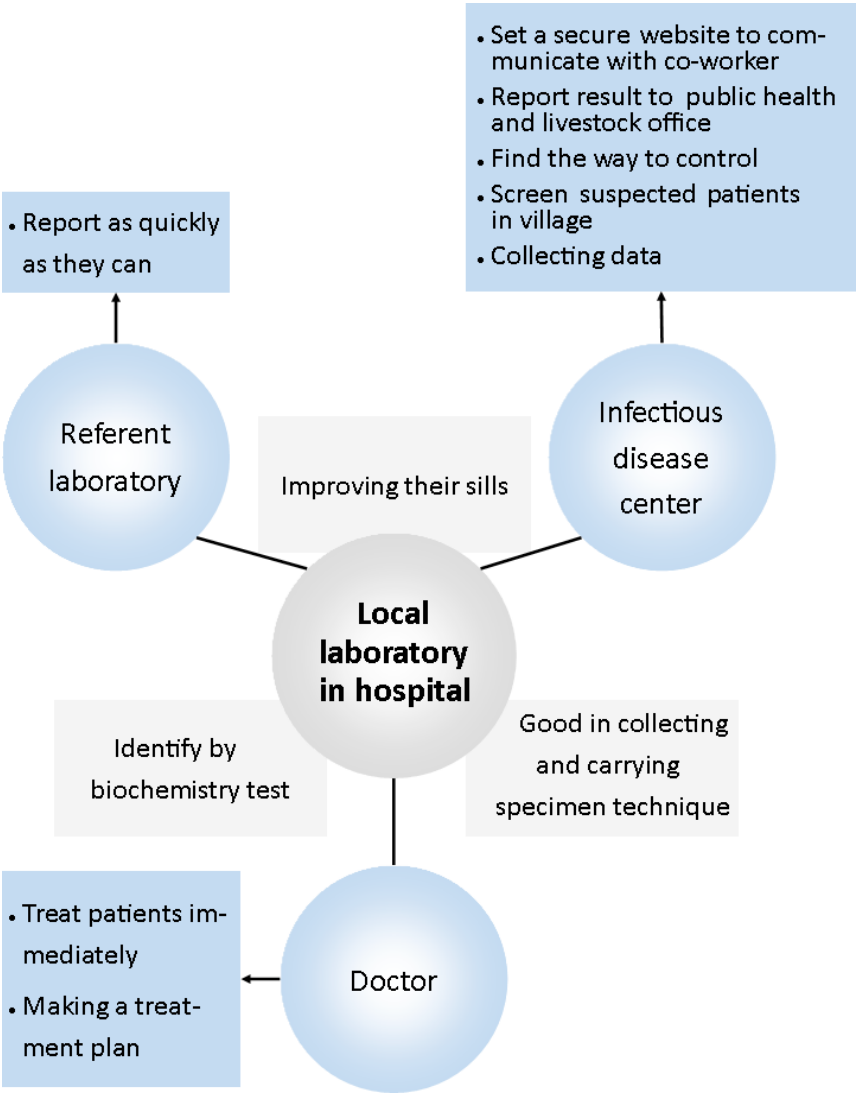
Laboratory networking and activity of each stakeholder



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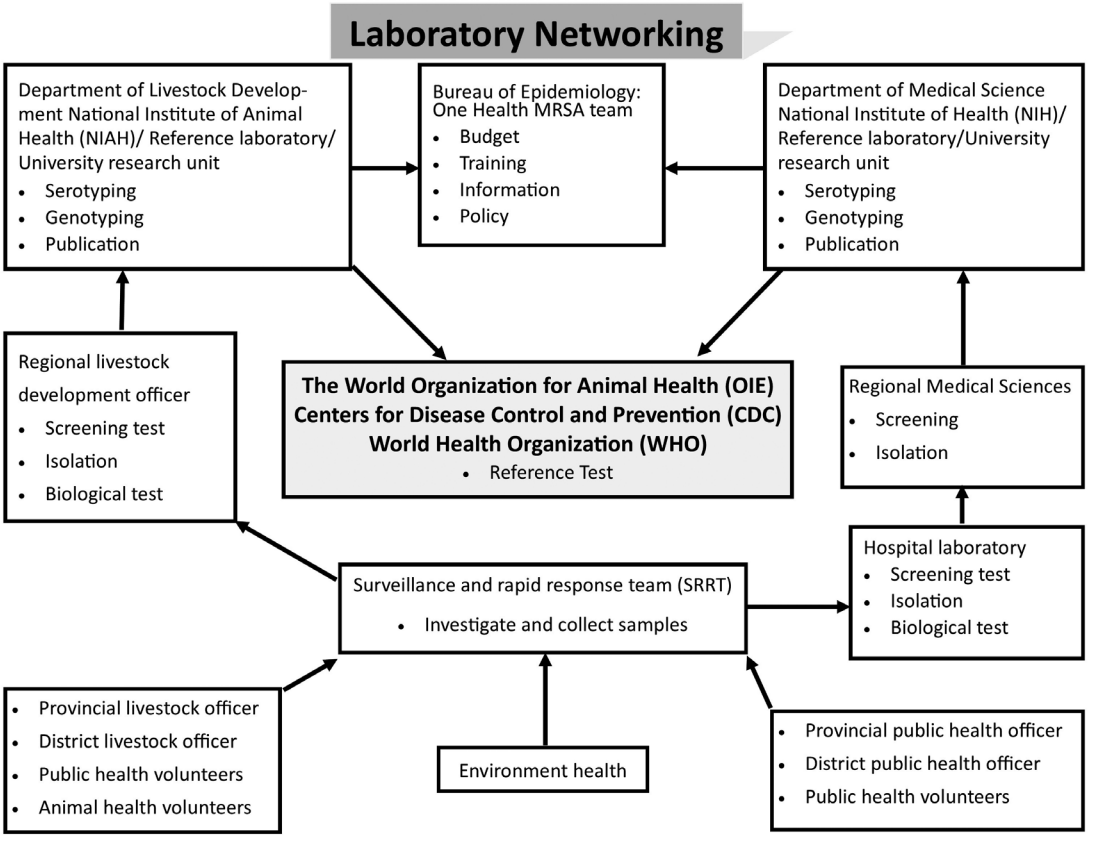
Group 2. Control of an epidemic of methicillin-resistant *Staphylococcus aureus*. Laboratory networking.



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Group 3. Control of an epidemic of *Salmonella enteritidis*.

Laboratory networking.

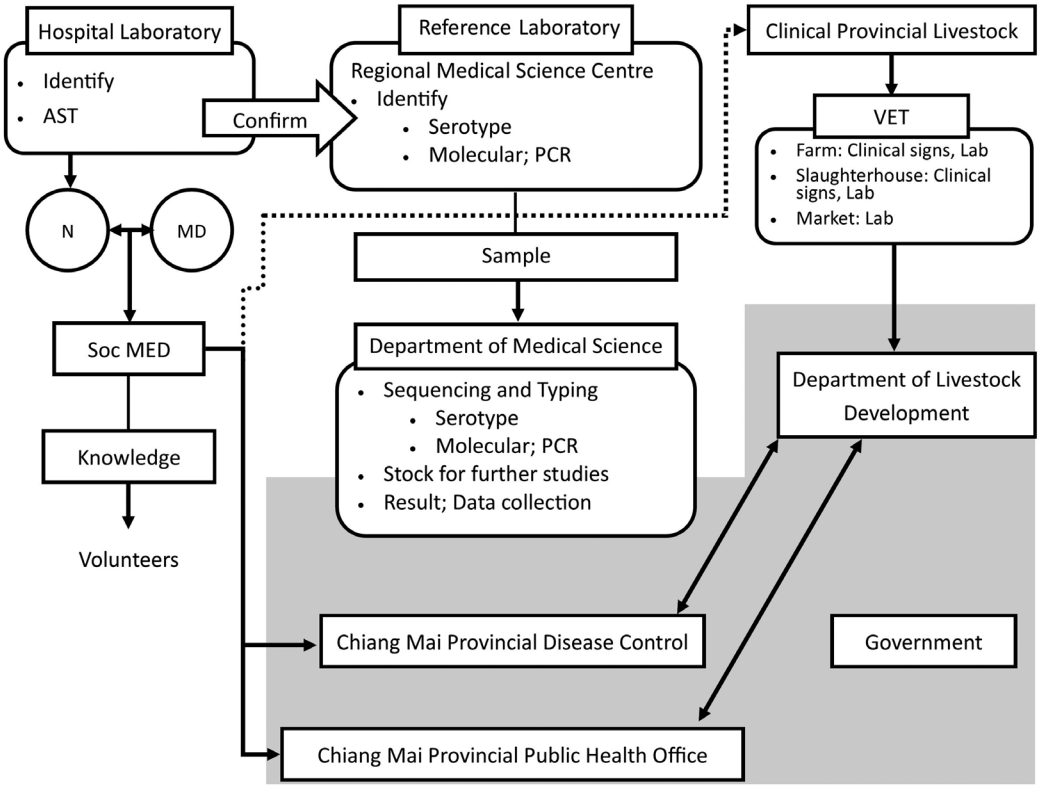

Fig 1 – Typical power point presentations on control strategies of zoonotic bacterial epidemics.

This training workshop emphasized acquisition of both soft and hard skills. Groups composed of participants with different careers and expertise formed a multidisciplinary units that allowed viewpoints from a variety of perspectives and arriving at multifaceted solutions. In the laboratory sessions, participants were diligent in perfecting skills they lacked and in many cases they were helped by fellow participants. Participants' comments on the laboratory training included: (a) "It's very nice learning how to identify unknown samples", (b) "I gained many techniques (in) identification of the unknown bacteria (and) all the facilitators gave good information and help(ed) me a lot during the

lab training", (c) "I need more organism(s) to improve my lab skill, (d) "Got knowledge and technique to do clinical laboratory (tests)", (e) "All staffs were sincere to help us and supported us (via) question, problem, result", (f) "Laboratory training is quite good and interesting", (g) "Excellent", and (h) "Very good laboratory to learn about microorganism and identification".

The consensus of the participants on the control of zoonotic bacterial epidemics a network be established with the Faculty of Associated Medical Sciences acting as the center of collaborating institutions (Fig 2). The network will be divided in two main laboratory groups: one involved in human and the other on animal infections. Each member will help each other and respond immediately to an outbreak through communications via a line group, such as "Zoo Lab Net" and Facebook Laboratory networking for zoonotic disease control (LNZDC) group. In addition, THOHUN should hold similar workshops annually in other regions of the country and ultimately to establish collaborative networks of professionals capable of providing expert training in bacteria identification and in infectious diseases surveillance for future One Health workforces.

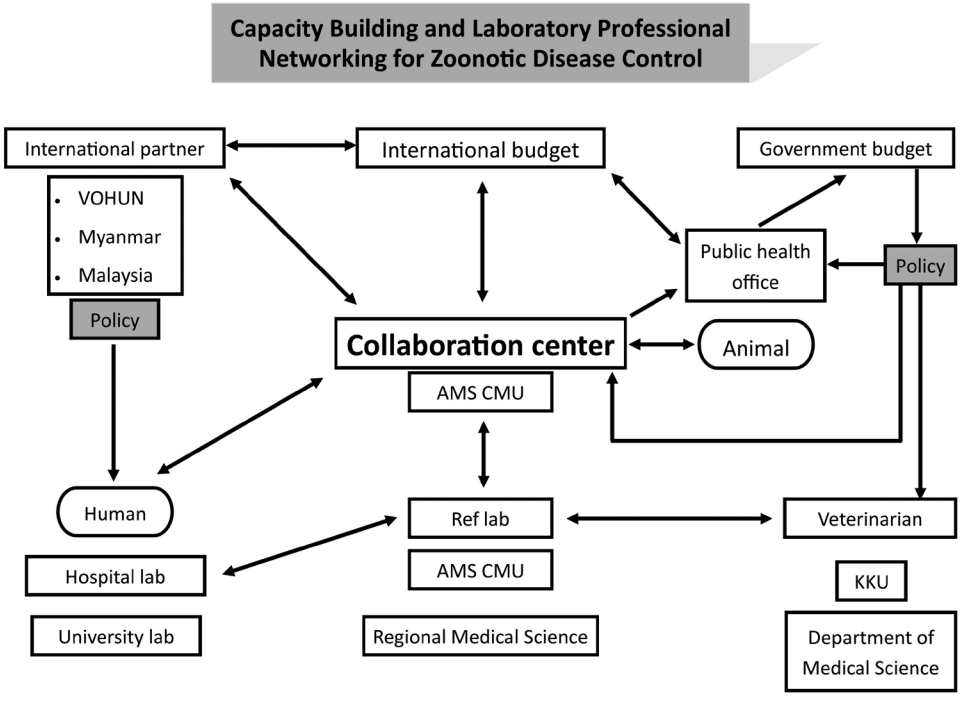

Fig 2 – Thailand network for control of zoonotic bacterial epidemics.

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