International Livestock Research Institute

# Training on veterinary and medical entomology and molecular methods for diagnostics

Guwahati, India

24–27 September 2018

ILRI
INTERNATIONAL LIVESTOCK RESEARCH INSTITUTE

CGIAR

SLU

ASSAM AGRICULTURAL UNIVERSITY

UPPSALA UNIVERSITET

ILRI thanks all donors and organizations which globally support its work through their contributions to the CGIAR Trust Fund.



Compiled by Johanna Lindahl and Chi Nguyen

**Citation**

ILRI (International Livestock Research Institute). 2018. *Training on veterinary and medical entomology and molecular methods for diagnostics*. Nairobi, Kenya: ILRI.

*Patron: Professor Peter C Doherty AC, FAA, FRS*
*Animal scientist, Nobel Prize Laureate for Physiology or Medicine–1996*

Box 30709, Nairobi 00100 Kenya
Phone +254 20 422 3000
Fax +254 20 422 3001
Email ilri-kenya@cgiar.org

ilri.org
*better lives through livestock*

ILRI is a CGIAR research centre

Box 5689, Addis Ababa, Ethiopia
Phone +251 11 617 2000
Fax +251 11 667 6923
Email ilri-ethiopia@cgiar.org

*ILRI has offices in East Africa • South Asia • Southeast and East Asia • Southern Africa • West Africa*

# Contents

# Abbreviations and acronyms

| | |
|---|---|
| AAU | Assam Agricultural University |
| ICMR | Indian Council of Medical Research |
| ILRI | International Livestock Research Institute |
| PCR | polymerase chain reaction |
| RMRC | Regional Medical Research Centre |
| RNA | ribonucleic acid |

# Acknowledgements

This workshop was financed by Uppsala University through the *Metropolitan mosquitoes* project and the CGIAR Research Program of Agriculture for Nutrition and Health, with support from Assam Agricultural University.

# Introduction

Vector-borne diseases cause a high disease burden globally, and Assam is no exception. With climate changes, urbanisation and population growth, we are likely to see increasing numbers of outbreaks from mosquito-borne diseases, including Japanese encephalitis, Dengue, Zika and West Nile virus. Vector-borne diseases are among the most complex of all infectious diseases to predict, prevent or control. Not only is it difficult to predict the habits of the vectors, but most vector-borne agents can infect animals as well. Mosquitoes and ticks are notoriously difficult to reach and often develop resistance to insecticides. Almost all vector-borne pathogens are zoonoses which can live in animals as well as in humans. Only a few vector-borne diseases can be prevented by use of vaccines. Every year there are more than 1 billion cases and over 1 million deaths from vector-borne diseases globally. Increasing deforestation and irrigation, as well as urbanization along with poor sanitation and water system, poor waste disposal and water storage are contributing to rise in vector-borne diseases. The World Health Day 2014 campaign, 'small bite: big threat' was aimed at raising awareness about the threats posed by insect vectors and the bacteria, viruses, and parasites they carry, and to motivate families and communities to protect themselves through simple measures.

Enormous impacts of vector-borne diseases on human and economic health have been well studied, but there are unique challenges associated with assessing and controlling zoonotic vector-borne diseases for which an animal host is a major component and even more so when multiple host species can play epidemiologically significant roles. The scope, relevance, and evaluation of vector-borne pathogens are highly dependent on organizational priorities. As different listings highlighted, known vector-borne diseases are of great importance and concern to local, regional, national and international organizations for their existing or potential burden to human and animal health. Vector-borne viruses account for 29% of the 593 known mammalian viruses across the globe. These pathogens have three times the host range compared to non-vector-borne viruses meaning that multiple animal species may act as hosts or reservoirs for any vector-borne disease. Additionally, some vector-borne viruses can be transmitted by multiple vector species. When a vector-borne disease affects both people and animals, humans are typically an incidental host and do not serve an important role in transmitting the disease to additional vectors. However, this does not exclude humans from being affected both directly and indirectly by vector-borne diseases for which they are not the primary host. Vector-borne diseases can have serious effects on human and animal health as well as significant economic implications and are therefore considered poverty promoting.

While climate change is commonly cited as a major contributor to increasing vector-borne disease prevalence and distribution, it is important to recognize that numerous human and ecological factors play a major role in disease emergence and spread. Patterns of vector-borne diseases can be attributed to a wide range of variables that vary by disease, location, and circumstance. Additionally, identifying the drivers that are associated with vector-borne disease emergence and spread presents an opportunity for prevention, education, and control. Changes in land use, war and famine, breakdown of public health measures, global trade and travel, and human behaviour are all associated with vector-borne disease emergence. By identifying situations where we anticipate vector-borne disease emergence, we can more effectively target prevention and intervention

strategies. A One Health approach that considers the links between humans, animals, and the environment can provide a more robust view around causes and possible solutions to vector-borne diseases. The ability to discern patterns of vector-borne diseases in animals hinges on consistent surveillance, prioritization and integrative strategies. While vaccines present an opportunity at the individual animal or herd level, the associated cost-benefit relationships pose additional challenges. A fundamental quality of vector-borne diseases, their dependence on the ecology of vectors and hosts, points to the need for the earnest engagement of the ecological sciences. Skilled veterinary and medical entomologists are critical for future work, and the number in this field are dwindling. There is an urgent need for ongoing support and training in veterinary and medical entomology to meet emerging demands.

Mosquito-borne diseases, including Japanese encephalitis and dengue, are major public health concerns in the north-eastern state of Assam, deterring equitable socioeconomic and industrial development. Among these, Japanese encephalitis has been the most predominant infection and is spread across the state. Japanese encephalitis, formerly endemic in upper Assam, is currently spreading fast across the state, with confirmed cases and a high case-fatality rate affecting all ages. Dengue has recently invaded the state, with a large concentration of cases in Guwahati city that are spreading to suburban areas. Since West Nile virus was first serologically detected in Assam during 2006, it has become recognised as another important etiological agent of acute encephalitis syndrome in addition to endemic Japanese encephalitis virus. While circulation of West Nile virus is evident, the role of vectors and avian hosts involved in the transmission remains unclear.

This workshop aims at building more capacity within Assam to diagnose and monitor mosquito-borne infections in the future, as well as to foster both national and international collaborations for disease prevention. Vector-borne diseases need expertise within veterinary and human health, ecology and entomology for fruitful mitigation, and therefore, a One Health approach is of utmost need.

# Training process

The workshop took place on 24–27 September 2018 at the College of Veterinary Science, Assam Agricultural University (AAU), Guwahati, India. It was co-hosted by AAU, the International Livestock Research Institute (ILRI), Uppsala University and the Swedish University of Agricultural Sciences. Altogether 18 participants (8 male and 10 female) were trained (see list at the end of this report).

**Day 1**

The first day of the workshop started with the inaugural session by AAU Vice Chancellor K.M. Bujarbaruah. In his inaugural speech, he referred to the alarming situation created by vector-borne diseases in Assam and other North Eastern states. He pointed out that around 17% of all infectious diseases are vector-borne and loss of 7 lakh lives every year has been due to these diseases. He expressed concern at the current Japanese encephalitis scenario in the State. He stressed the need for controlling vectors, capacity building and competitiveness through improved infrastructure, by enhancing the facilities for monitoring and surveillance of such diseases and above all, by bringing about sensitisation among the people. Johanna Lindahl, a senior scientist at ILRI, laid emphasis on collaboration both across and within the border with multidisciplinary approaches. She opined that one should think out of the box and find out what strategic research and innovative ideas are needed to address the diseases that are posing a threat to the health of humans and animals as well as the economy of the nation.

The morning session was followed by two presentations on vector-borne-diseases in India and Asia and Epidemiology of Japanese encephalitis virus and West Nile virus in Assam by Johanna Lindahl (ILRI and Uppsala University) and S.A. Khan (Indian Council of Medical Research [ICMR]), respectively.

The afternoon session was followed with four presentations elaborating entomological research work in Assam and the presence of mosquitoes as vectors of arboviral diseases in this North East state. The afternoon session concluded with an exercise on mosquito sampling. Participants learned how to monitor, sample and trap mosquito eggs, larvae and adults.

**Day 2**

The second day continued with mosquito sampling exercise which was facilitated by Johanna Lindahl (ILRI and Uppsala University) and Jenny Hesson (Uppsala University). This was followed by a lecture on mosquito systematic and identification. In the afternoon, trainers guided participants on how to identify adult mosquitoes (and larvae) through practical exercise. And, they continued with mosquito sampling exercise from the morning session.

**Day 3**

The third day the students continued practicing mosquito identification, and thereafter there was an exercise on ribonucleic acid (RNA extraction as well as quantitative, real-time, polymerase chain reaction (PCR).

**Day 4**

For the last day of the workshop, the participants were guided with laboratory exercise on gel electrophoresis, and there were discussions on how to interpret results from the real-time PCR as well as from the gels. In the afternoon, trainers introduced participants with basic knowledge about how to survey and control mosquito vectors. The workshop closed with observations and remarks from facilitators. R.A. Hazarika of AAU hoped for a successful mission together with multidisciplinary collaborative approaches for executing the gain acquiring through the workshop. A. Chakraborty of AAU expressed satisfaction with the issues of vector-borne diseases discussed at the workshop. He said that through research and training in collaboration with universities and other organizations from abroad, veterinary and medical institutes in the State including experts from AAU can help evolve a solution to reduce the burden of vector-borne diseases in the State.

# Trainers

The workshop was facilitated by nine people. Below are their contact details.

Dr. Razibuddin Ahmed Hazarika, Professor and Head, Department of Veterinary Public Health, College of Veterinary Science, AAU rah1962@rediffmail.com

Dr. Johanna Lindahl, veterinary epidemiologist, ILRI j.lindahl@cgiar.org

Dr. Jenny Hesson, Scientist, Zoonosis Science Centre, Department of Medical Biochemistry and Microbiology, Uppsala University, Sweden jenny.hesson@imbim.uu.se

Dr. Jiaxin Ling, Scientist, Zoonosis Science Centre, Department of Medical Biochemistry and Microbiology, Uppsala University, Sweden jiaxin.ling@imbim.uu.se

Dr. Sailen Kumar Das, Professor and Head, Department of Veterinary Microbiology, College of Veterinary Science, AAU drskdas53@gmail.com

Dr. Dilip Kumar Deka, Professor and Head, Department of Veterinary Parasitology, College of Veterinary Science, AAU dilipkumar.deka@rediffmail.com

Dr. Nagendra Nath Barman, Professor, Department of Veterinary Microbiology, College of Veterinary Science, AAU nnbarman@gmail.com

Dr. Saidul Islam, Professor, Department of Veterinary Parasitology, College of Veterinary Science, AAU isaidul@yahoo.com

Dr. Siraj Ahmed Khan, Scientist, Deputy Director, ICMR - Regional Medical Research Centre (RMRC), Dibrugarh sirajkhanicmr@gmail.com

# Program

| Time | Activity | Person in charge |
|---|---|---|
| **Day 1–** Venue: Conference Hall College of Veterinary Science (morning), Department of Veterinary Parasitology (afternoon) | | |
| 0930 – 1030 | Inaugural Session | |
| 1030 – 1100 | Tea break | |
| 1100 – 1120 | Introduction to mosquito-borne diseases | Dr. R.A. Hazarika |
| 1120 – 1145 | Vector-borne diseases in India and Asia | Dr. Johanna Lindahl |
| 1145 – 1215 | Epidemiology of Japanese encephalitis virus and West Nile virus in Assam | Dr. S.A. Khan |
| 1215 – 1245 | Group discussion: Mitigation options | Dr. Johanna Lindahl |
| 1245 – 1300 | Group feedback and finalization | |
| 1300 – 1400 | Lunch break | |
| 1400 – 1430 | A brief outline on entomological research work in Assam | Dr. D.K. Deka/ Dr. S. Islam |
| 1430 – 1500 | Mosquito/vector ecology. | Dr. Jenny Hesson |
| 1500 – 1530 | Mosquitoes as vectors of arboviral diseases present in Assam | Dr. S.A. Khan |
| 1530 – 1730 | Mosquito sampling exercise: The agricultural, peri-urban and rural system: breeding sites, monitoring/sampling/trapping of eggs, larvae and adults | Dr. Johanna Lindahl Dr. Jenny Hesson Dr. Jiaxin Ling |
| **Day 2 –** Venue: Department of Veterinary Parasitology/ Nearby Pig and poultry farms | | |
| 0800 – 1030 | Mosquito sampling exercise: Emptying traps. Larval/egg sampling and monitoring | Dr. Johanna Lindahl<br>Dr. Jenny Hesson |
| 1030 – 1100 | **Tea break** | |
| 1100 – 1300 | Lecture: Mosquito systematic and identification | Dr. Johanna Lindahl<br>Dr. Jenny Hesson |
| 1300 – 1400 | **Lunch break** | |
| 1400 – 1730 | Mosquito identification exercise: Morphological identification of adult mosquitoes (and larvae)<br>Mosquito sampling exercise**:** The urban system: breeding sites, monitoring/ sampling/ trapping of eggs, larvae and adults | Dr. Johanna Lindahl<br>Dr. Jenny Hesson<br>Dr. Dr. Jiaxin Ling |
| **Day 3 –** Venue: Department of Veterinary Parasitology (before 9 a.m.), Department of Veterinary Microbiology (10:00a.m. - 4.30 p.m.) | | |
| 0800-0930 | Collection of traps, Continuing mosquito identification exercise<br>Identification of larvae | Dr. Johanna Lindahl<br>Dr. Jenny Hesson |
| 0930 – 1000 | **Tea break** | |
| 1000 –1300 | A brief outline on mosquito-borne virus molecular research works | Dr. S.K. Das/Dr. N. N. Barman |
| | Lecture: Introduction to virology | Dr Jiaxin Ling |
| | Lecture: Molecular work with mosquitoes - RNA extraction and viral RNA detection<br>Laboratory exercise: RNA extraction | Dr. Johanna Lindahl<br>Dr. Jiaxin Ling |
| 1300–1400 | **Lunch break** | |
| 1400–1730<br>(including tea break) | Laboratory exercise: qPCR<br>During the qPCR run time: Discussion on problems faced with PCR | Dr. Johanna Lindahl<br>Dr. Jiaxin Ling |
| **Day 4 – Extra optional day** | | |
| If required | Specific project discussion time | Dr. Johanna Lindahl<br>Dr. Jenny Hesson<br>Dr. Jiaxin Ling |
| 0930 – 1030 | Interpretation of qPCR results | Dr. Johanna Lindahl<br>Dr. Jenny Hesson<br>Dr. Jiaxin Ling |
| 1030 – 1100 | **Tea break** | |
| 1100 – 1300 | Laboratory exercise: Gel electrophoresis | Dr. Johanna Lindahl<br>Dr. Jenny Hesson |
| 1300 – 1400 | **Lunch break** | |
| 1400 – 1530 | Lecture: Surveillance and control of mosquito vectors: The basics | Dr. Johanna Lindahl<br>Dr. Jenny Hesson<br>Dr. Jiaxin Ling |
| 1530 – 1545 | **Tea break** | |
| 1545 – 1700 | Final discussion and closing of the workshop | Dr. R.A. Hazarika<br>Dr. Johanna Lindahl<br>Dr. S.K. Das<br>Dr. D.K. Deka<br>Dr. N.N. Barman<br>Dr. S. Islam<br>Dr. Jenny Hesson<br>Dr. Jiaxin Ling |

# List of trainees

| Name | Title/ Position | Organization | Sex |
|---|---|---|---|
| Dr. Archana Talukdar | Assistant Professor | Assam Agricultural University | F |
| Dr. Sarat Sonowal | Assistant Professor | Assam Agricultural University | M |
| Dr. Aditya Barua | | Assam Agricultural University | M |
| Dr. Phunu Talukdar | | Assam Agricultural University | F |
| Ms. Bhanita Talukdar | | Assam Agricultural University | F |
| Dr. Kuntola Roy | Assistant Professor | Assam Agricultural University | F |
| Dr. D. P. Bora | Assistant Professor | Assam Agricultural University | M |
| Dr. Sutopa Das | Professor | Assam Agricultural University | F |
| Dr. Ditul Barman | Assistant Professor | Assam Agricultural University | M |
| Dr. Deepa Lahkar | Assistant Professor | Assam Agricultural University | F |
| Mr. Biplob Sarmah | DHR Young Scientist Fellow | ICMR-RMRC, Dibrugarh | M |
| Ms. Anisha Shah | Research Assistant | ICMR-RMRC, Dibrugarh | F |
| Mr. Manash Singha | Research Assistant | ICMR-RMRC, Dibrugarh | M |
| Dr. Anupam Brahma | Junior Scientist | Assam Agricultural University | M |
| Dr. Seema Rani Pegu | Scientist | Indian Council on Agricultural Research | F |
| Dr. Manju Goswami | | Animal Health and Veterinary Department, Government of Assam | F |
| Dr. Barnali Saikia | | Animal Health and Veterinary Department, Government of Assam | F |
| Dr. Sidhatha Mohakud | | Assam Agricultural University | M |