OFFICE OF THE DEPARTMENT OF MICROBIOLOGY
ASSAM MEDICAL COLLEGE, DIBRUGARH

NO.2015/AMC/MICRO/DBT/002 Dated, Dibrugarh the 24th January2015

To,
The Prof. A.K. Mukherjee
Head & Coordinator
DBT Nodal Centre
Tezpur University, Tezpur

Subject: Submission of updated Annual Progress Report

Dear Sir,
With reference to the subject cited above I am sending herewith the annual progress report for the year 2014 of DBT Molecular Biology/ Virology Laboratory, Department of Microbiology, Assam Medical College & Hospital, Dibrugarh.

Thanking you,

With regards

Professor & Head
Department of Microbiology
Assam Medical College
Dibrugarh

(Dr. Lahari Saikia)
PI, DBT Project &
Professor & Head
Dept. of Microbiology
Assam Medical College & Hospital
Dibrugarh

ANNUAL REPORT FOR THE YEAR 2014

Name of Laboratory : DBT Molecular Biology/ Virology Laboratory

Name of PI : Dr. Lahari Saikia

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1. Completion/taking forward of sanctioned objectives

Our ongoing project is under DBT infrastructure and Development scheme. The Major sanctioned objectives are:

a. A study of viral aetiology of acute encephalitis Syndrome
b. Estimation of HIV and HBV viral load
c. Diagnosis of pulmonary and extra pulmonary tuberculosis.

Our first objective to detect the main viral cause of acute encephalitis Syndrome (AES) with a view to aid clinical diagnosis and treatment. Till date we have processed total 517 CSF samples in Real time PCR for detection of four viruses i.e. JEV,WNV, Enterovirus and HSV 1& 2.out of which only one sample showed positive result for HSV. Currently we have been processing the ELISA negative JE samples for the year 2014.

Regarding the second objective our lab has been providing patient service for HIV and HBV viral load. Till December total 77 no's of HBV viral load cases and 23 no's of HIV viral load cases have been successfully completed.

With respect to the third objective our lab has also started molecular diagnostic service on extra pulmonary tuberculosis. Recently we have started the multi drug resistance (MDR) monitoring service by HAIN twin incubator using line probe assay.

DBT tissue culture lab has been made functional. Initially we will start the isolation and identification of Influenza virus. We will also start the maintenance, isolation, detection of JE virus by using mosquito cell line.

Space dedicated to DBT Healthcare lab

A. **DBT Molecular Biology/ Virology Lab: (29x18) =522 sq. feet**
B. **DBT Tissue Culture Lab: (29x 22) =638 sq. feet**

1. Enhancing patient services:

Ongoing Patient services from Virology Lab:

Patient services on Hepatitis B (HBV) viral load estimation and HIV viral load estimation are being provided by Virology Lab routinely using the DBT sponsored infrastructure facility.

The kits and reagents are being provided by Assam Medical College & Hospital Management.

Name of Kits:

Name of test	Name of Kit	Company	Approval
HBV Viral Load	Cobas TaqMan HBV test kit	Roche Diagnostics Pvt. Ltd.	**IVD** approved
HIV Viral load	Cobas TaqMan HIV-1 test kit, v2.0	Roche Diagnostics Pvt. Ltd.	**IVD** approved

- All tests are performed on Roche, Cobas TaqMan 48 platform (Real Time PCR)
- Approved price charged for viral load assay:
  1. For HBV viral load: **Rs. 3500.00** per test
  2. For HIV viral load : **At free of cost**
- The revenue generated from the said services is deposited under Assam Medical College & Hospital Management fund.
- The said viral load diagnostics services will be continued by Department of Microbiology under support of the Assam Medical College & Hospital Management.

Sl.No.	Name of the major Equipment	Time Period	Diagnostic facility/Parameter	Number of Cases	Sample Tested	Remarks/revenue if any
1	**Cobas TaqMan**	**June 2013 to**	**HBV viral load**	**77**	**77**	**Rs.**

	48 Real Time PCR	**December 2014**				**2,59,000.00 (excluding the free service cases)**
		August 2013 to March 2014	**HIV viral load**	**23**	**23**	**All service have been provided at free of cost**
2	**HAIN Twin incubator (provided by Hospital Management Society)**	**From September 2014 to December 2014**	**Multi Drug Resistance (MDR) assay in MTB positive cases**	**28**	**28**	**Service is being provided at free of cost for initial period**
3	**8 Capillary DNA Sequencer**	**July 2014 to December 2014**	**Sequencing Service**	**266**	**266**	**At Present service is being provided as non-profit Scheme. In future it will be continued as Revenue generate model**

2. Quality Control Measures:

Regarding quality control measures for viral load assay we have been using IVD approved kits from ROCHE Diagnostics. The kit itself provide three internal quality controls i.e. NC (negative Control. LPC (low positive control) and HPC (High positive control). These three controls are run with each and every batch of test samples for internal quality control test. For EQAS & ILS communication has already been made with NIMHANS for viral load Testing.

Our lab is also has been communicating with central laboratory **NIMHANS** (Bangalore) for managing **EQAS & ILC** for the respective diagnostic test. As the other laboratories of our department have already got NABL accreditation so we have a plan to include the Virology laboratory in the scope of NABL in future.

3. Training:

Name and workshop/training programme ATTENDED From August 2013-	**Duration & Venue**	**Organized /conducted by**	**Name & no. of participant(s) from project**	**Source of fund availed**

August 2014				
Training on animal tissue culture	For Seven (7) days, at RMRC, Dibrugarh	N/A	one	No expenditure
Onsite training on Sequencing	On 11.08.2014 to 13.08.2014 at DBT Molecular Biology/Virology Lab	DBT Molecular Biology Lab	All staff	DBT training fund.
Short term training on Animal Cell Culture	For Seven 15 days, at RMRC, Dibrugarh	N/A	one	No expenditure

Workshop/training programme CONDUCTED From August 2013-August 2014	**Duration & Venue**	**Organized /conducted by**	**No. of participant(s) attended**	**Objective of workshop /training**	**Source of fund availed and total amount spent.**
CME on "**Hepatitis B viral Infection"**	12.03.2014, Venue: Conference Hall, Department of Microbiology, AMCH	Department of Microbiology in association with DBT Molecular Biology/Virology Lab	25	Awarness about the Molecular Diagnostic parameters for Hepatitis B viral Infection and Diagnostic facilities available in our DBT Molecular Biology lab	From Departmental Fund, total amount spent is **Rs. 35,200.00**
3 Days Workshop on **Sequencing and post Sequencing analysis**	Held on 10th, 11th and 12 th September 2014,DBT Molecular Biology Lab,AMCH	DBT Molecular Biology/ Virology Lab	23	To provide training on PCR, Cycle Sequencing, Purification, Capillary Electrophoresis, Sequence analysis, Data submission , phylogenetic analysis .	From DBT project fund under Training Head, total amount spent is Rs.**5,06,143.00**

Sl No	Name (please mention designation if project personnel)	Thesis Title (please also mention PhD/MD/MS)	Thesis Status	Equipment Used
1	Dr. Saurav Patgiri	A study of viral aetiology of acute encephalitis syndrome with special reference to Japanese Encephalitis- A hospital based study	Submitted	Biohazard safety Cabinet, Table Top Refrigerated Microcentrifuge , Micropipette Set , Cobas TaqMan 48 Real Time PCR
2	Dr. Anusmita Das	Spectrum of bacterial pathogens causing community acquired pneumonia in children below 5 years.	Submitted in 2014	Biohazard safety Cabinet, PCR, Gel Electrophoresis & Power Pack, UV Transilluminator, Gel Documentation System, DNA Sequencer
3	Dr. Bhaswati Sharma	"Occurrence of Respiratory Syncytial viruses, Influenza A and B virus, Parainfluenza1,2,3 virus and Human Metapneumovirus in acute respiratory tract infections in paediatric patients-A Hospital based Study"	Submitted in 2014	Biohazard safety Cabinet, PCR, Gel Electrophoresis & Power Pack, UV Transilluminator, Gel Documentation System, DNA Sequencer

4. Science based research activity:

Research work for detection of possible viral aetiological cause in AES cases by molecular method i.e. Real time PCR is in progress. We are using Taqman based real time PCR kits for detection of four viruses i.e. JEV, West Nile Virus, Enterovirus, Herpes Simplex Virus 1 &2.

In the study we have considered only the ELISA negative CSF samples.

Real Time PCR report for AES cases from December 2013 to December 2014:

Time Period	JEV			WNV			Enterovirus			HSV 1 & 2		
December 2013 to December 2014	No. of cases	JE Positive	JE negative	No. of cases	WNV positive	WNV negative	No. of cases	Positive	Negative	No. of cases	Positive	Negative
	49	0	49	42	0	42	37	0	37	53	0	53

Till now total **266 no's** of samples have been processed in DNA sequencer. Sequencing service is being provided by Molecular Biology/Virology Lab.

Work going on under CDC project "Strengthening laboratory support for surveillance to JE in India, with NIMHANS:

A surveillance to AES is undergoing for detection of other than JE pathogen in Assam for bacterial agents such as **Neisseria Meningitidis, S. Pneumoniae, H. Influenzae** and viral agents such as **Herpes simplex virus-1, Enterovirus and Chikungya** Molecular diagnosis for both bacterial and viral Meningitis will be done based on TaqMan principle in Real Time PCR platform .

Both serological and Molecular diagnosis is undergoing in this project.

Infrastructure of DBT Molecular Biology/Virology laboratory has been utilizing for DNA/RNA extraction purpose and the quantitative PCR will be performed at LC-480 in an another laboratory MDRL(ICMR unit)

Total 118 no.s of CSF sample will be processed for Molecular diagnosis. Currently RNA & DNA extraction of CSF samlple is going in Molecular Biology/Virology Lab.

Serological test i.e. ELISA is being performed for detection of West Nile and Scrub Typhus.

Paper Published:

- **Saurav Jyoti Patgiri, A K Borthakur, B Borkakoty, Lahari Saikia, R Dutta, S K Phukan, "An appraisal of clinicopathological parameters in Japanese encephalitis and changing epidemiological trends in upper Assam, India"; *Indian Journal of Pathology and Microbiology*-57(3), July-September 2014.**

Paper Submitted by PGT (under review):

- **Anusmita Das, Lahari Saikia, S J patgiri, P Dowarah, R Nath, "Bacterial pathogens associated with community acquired pneumonia in children below 5 years of age";*Indian Pediatrics*. Date of submission-26/11/2014.**

- **Bhaswati Sarma, Lahari Saikia, S J patgiri, P Dowarah, R Nath, "Occurrence of eight common viruses causing acute respiratory tract infections in paediatric patients, a hospital based study"; *Indian Journal of Medical Microbiology*. Date of submission- 27/11/2014.**

5. Intra institutional & extra institutional use of laboratory facility :

Sl.no	Name of Dept/Institution/Person	Equipment used	Time period	Purpose
1.	Virology Division, RMRC, Dibrugarh, Assam	DNA Sequencer	From July 2014 onwards	Sequencing of PCR products
2	DBT Project, Dept. of Microbiology, GMCH, Guwahati, Assam	DNA Sequencer	From August 2014 onwards	Sequencing of PCR products
3	DBT project, Molecular Mycology Lab, Dept of Microbiology, AMCH	DNA Sequencer	From August 2014 onwards	Sequencing of PCR products
4	Dr. Anusmita Das	DNA Sequencer	From August 2014	Sequencing of PCR products
5	Dr. Bhaswati Sharma	DNA Sequencer	From August 2014 onwards	Sequencing of PCR products
6	Department of Paediatrics, AMCH, Dibrugarh	-80^0 C Deep Freezer	From June 2014 to September 2014	For Storage of Clinical Samples for a DBT project
7	DBT –Tuberculosis Project, Dept. of Microbiology,AMCH	Conventional PCR	From September 2014 onwards	Multiplex PCR for MDR TB detection.

6. Sustenance& Revenue Generation Strategy:

Visions for sustenance of DBT Molecular Biology & Virology Lab:

DBT Molecular Biology, Virology Laboratory was proposed with an idea to establish a well-equipped molecular biology laboratory for molecular detection of major viral diseases such as Japanese encephalitis, HIV, HBV etc. Apart from this it was also aimed to establish scope of research and to carry out academic activities for post graduate medical students such as M.D. thesis work.

The Molecular Biology Lab was established in 2009 under funding of DBT, New Delhi, Govt. of India. DBT sponsored the infrastructure project entitled "Up gradation of Infrastructure in Medical Colleges in NER, India" for 5 years. Following are the visions for sustenance of Molecular Biology Laboratory beyond December 2014.

Proposed plan for enhancing diagnostic and research activity:

1. Existing HBV & HIV viral load diagnostic services will be continued.
2. Molecular detection of Pulmonary and extra pulmonary tuberculosis and MDR tuberculosis has already been started and will be continued in future as revenue generating model.
3. Communication has been started with NACO to recognize the Virology Laboratory to support for HIV viral load detection. Already all the necessary documents have been submitted to NACO and preliminary inspection by NACO team has been completed.
4. The Virology laboratory has been recognized as an Apex Laboratory by CDC, under a project named as "Strengthening laboratory support for surveillance to JE in India".
5. Tissue culture Laboratory has already been made functional and media and reagent preparation is going on. Two manpower have completed short term training on tissue culture in RMRC Dibrugarh. Our preliminary objective is isolation and identification of Influenza A & B virus using MDCK cell line.
6. The existing facilities of Molecular Biology, Virology Lab will be utilized to carry out MD thesis work and Extra Mural Research project works. One PGT (Dr. Priyanka Mukherjee) is about to start her thesis work entitled "Prevalence and Genotyping distribution of HPV in antenatal Women" in DBT Molecular Biology/Virology Lab.

7. Optimal utilization of DNA Sequencer is being continued by providing sequencing service to RMRC Dibrugarh, GMC and other DBT sponsored projects of Assam Medical College. In future also this will be continued as revenue generating model
8. To develop more trained manpower laboratory will organize various training workshop in future.

Signature of PI:

Full name: Dr. Lahari Saikia

Designation: Professor & Head, Department of Microbiology, AMCH

Date: 24/01/2015