

Laboratory Quality Assurance Procedures and Results in IBBA

Executive Summary

Integrated Behavioral and Biological Assessment survey of key population groups in 29 districts was conducted in 29 districts of five districts and highways in 2005-2006. The prevalence of HIV and STI were measured by performing standardized test through establishing three tiered laboratory network and quality systems selected laboratories. The proficiency of the laboratories is monitored using a structured quality assessment schemes and supportive supervision.

Key Quality Systems and Procedures implemented are

1. *Building infrastructure capacities.*
2. *Coordination of network lab*
3. *Capacity Building of Staff*
4. *Specimen tracking system*
5. *Quality Systems: Quality control (internal and external), proficiency programs*
6. *Supportive supervision*
7. *Data management*

Introduction

In India, sentinel surveillance is used annually to estimate the prevalence of Human Immunodeficiency Virus (HIV) infection in the country and to monitor trends in the epidemic. In 2005, it was estimated that more than 5.3 million people* in India are infected with HIV. The majority of people living with HIV in India are in six states: Tamil Nadu, Andhra Pradesh, and Karnataka in the south, Maharashtra in the west, and Manipur and Nagaland in the northeast. The national HIV prevalence was 0.9% with higher prevalence rates in selected areas and among key populations. People living with HIV represent all sectors of Indian society, but was found to be high among sex workers, their clients and partners, men who engage in risky sex with men, and injecting drug users.

Avahan India AIDS Initiative of the Bill and Melinda Gates Foundation, a national HIV prevention program was established in 2003. The Avahan initiative supplements existing HIV/AIDS efforts in India by providing funding and technical support to a wide range of organizations and works closely with the National AIDS Control Organization and the State AIDS Societies. The goal of the Avahan Focused Intervention program for key populations (Sex workers, MSM, IDU and Clients/truck drivers) is to improve access to effective prevention services in the southern and western states of Tamil Nadu, Andhra Pradesh, Karnataka, and Maharashtra, where HIV is primarily transmitted by sex work, and in the northeastern states of Manipur and Nagaland, where injecting drug use drives infection rates.

To assess major outcomes and measure the impact of Avahan interventions implemented by the Avahan India AIDS Initiative in 71 districts in five states and national highways, first phase of an integrated behavioral and biological assessment (IBBA) was initiated and implemented in 2005. The project was implemented by the FHI in close collaboration with National AIDS Control Organization (NACO) and State AIDS Control Societies (SACS).

Objective

The overall objective of the IBBA is to collect necessary information for assessing the outcomes and the impact of HIV interventions in Avahan project districts. The conduct of IBBA would strengthen the capacities (infrastructure and human resources) of national and state level institutes including the Indian Council of Medical Research (ICMR), the National AIDS Research Institute (NARI), The National Institute of Epidemiology (NIE), The National Institute of Nutrition (NIN), the Regional Medical Research Council (RMRC), and the National Institute of Medical Statistics (NIMS).

Laboratory Network:

The goal of the laboratory network is to strengthen the performance of laboratories to support diagnosis, disease surveillance; control and prevention programs. Clinical Laboratory Networks are primarily set up globally by the private sector to provide support to vast number of physicians working in hospitals and clinics. A complete range of clinical laboratory services are offered by the network to its clients with a special focus on customer (patient) and primary stakeholder (physicians/insurance companies). Quality has become the mantra of these clinical laboratories.

Similarly, Global Laboratory Networks were established for public health program such as control of Polio, measles, rubella Yellow fever and Japanese B Encephalitis by WHO. A Sentinel Laboratory network for surveillance of syphilis was also implemented by the EURO Surveillance

following syphilis outbreaks in Belgium. Laboratory network for STI//TBHIV have also been established in Africa.

The Concept of Laboratory Network in IBBA Study

The concept of laboratory network in the context of the IBBA is to establish a web of laboratories interconnected through development of guidelines on laboratory standards, methods, proficiency, training, logistics, resource utilization and communication systems. The proficiency of the laboratories is monitored using a structured quality assessment schemes and supportive supervision. The IBBA systematically identified laboratories that met criteria for performance of tests required for the biological component of IBBA in five high prevalent states in India.

Levels of Laboratories

Three levels of laboratories were established for the conduct of biological component of IBBA in 2005-2006

Level 1: Field specimen collection sites

Biological samples were collected at field site make shift laboratories from study participants after obtaining consent.

Level 2: District laboratories

Twenty nine districts in 5 states and highways were chosen for implementation of IBBA survey. The study sites were selected after extensive consultations with the NACO, SACS and District authorities. The survey methodologies for behavioral and biological assessment were developed, documented and implemented during the survey. District laboratories were assessed using a simple criteria and were selected to participate in the survey.

The role and responsibilities of District Laboratory are

1. Ensuring adequate laboratory supplies to field staff
2. Performing RPR test as per the SOPS
3. Quality documentation of data
4. Management of package of samples for transport to the State laboratories

Coordination with the field staff

Level 3: State Laboratories: ICMR Institutions

3A. National Institute for Nutrition (Andhra Pradesh): The National Institute of Nutrition is premier research institute established to conduct multi disciplinary research (laboratory, clinical and community) activities in food and nutrition. NIN has four major laboratory research programs such as biophysics, biochemistry, molecular biology and analytical chemistry. Molecular biology focuses on gene – nutrient interaction and virology department has conducted clinical research on maternal infections and pregnancy outcomes (Toxoplasmosis and Rubella and Immunology). The institute has an instrumentation section which provides support research laboratories.

3B The National Institute of Epidemiology (Tamil Nadu). This institute was established in 1999 in Chennai, TN and carry out research in interventional studies, disease modeling, health systems, diseases control and evaluation of health schemes. This institute has a basic laboratory to support research and field activities in leprosy and HIV serology.

3C. The Regional Medical Research Center (Manipur & Nagaland). The Regional Medical research center in Dibrugarh, Assam was established by ICMR to promote and address regional health priority issues and health systems in northeast region of India. The center has been conducting research in non communicable and communicable diseases (Five divisions) including HIV. The center has its own laboratory which has essential laboratory equipment and an animal house to support its arbovirology work. The laboratory performs diagnostic tests in microbiology, biochemistry, virology and molecular biology.

Level 4: IBBA Reference Laboratory (NARI)

The National AIDS Research Institute (NARI), a leadership ICMR organization was established in 1992 with the mission to provide leadership in biomedical research on HIV/AIDS in India and also to compliment and strengthen the National AIDS Control Programmes. The institute has been perusing international research programs in prevention, treatment, care & support and behavioral strategies. NARI has been conducting clinical trials in compliance with Good Clinical Practices. The Institute has been promoting GCLP and has been instrumental in developing national schemes on quality assurance for HIV diagnosis.

The National AIDS Research Institute was given the responsibility of coordinating and implementing the first phase IBBA study in five high prevalent states and highways in India in 2005. The novel component of this survey is an attempt to collect biological indicators of STI (syphilis, antibodies to herpes type 2, NG/CT).

Coordination of laboratory activities and policies (ICMR, FHI, UNC and BMGF)

The coordination of IBBA activities includes development of all laboratory related documents, ensure smooth delivery of supplies to ICMR institutes and field sites, timely procurement of supplies from different vendors, communicating with lab managers of different ICMR institutes, liaison with consultants at UNC and Delhi and liaison with Bill and Melinda Gates Foundation focal person for Monitoring and evaluation grant.

- 1. Procurement of supplies:** FHI was responsible for procurement of all supplies, reagents and kits and instruments for setting up district laboratories and specialized equipments for APTIMA test. All procurements were done through two suppliers; one for APTIMA Combo2 test for NG/CT including the DTS 400 system, the other one was for all supplies, consumables, reagents, kits and instruments.
- 2. Development of laboratory related documents:** FHI was involved in coordination of development of all laboratory related document. This includes SOPs for all tests, procedure for specimen collection in field, specimen storage and transportation, development of form for chain of custody management, results recording sheet and laboratory data entry forms.
- 3. Coordination of supplies:** After procurement was signed the supplies were initially stored at supplier's warehouse. Based on the sample size and number of district hospital and the need of state ICMR institutes a supply chain management matrix was developed. Supplies for each district and ICMR institute were packed in separate boxes with clear marking of the name of the district. Accordingly all supplies for one state including field, district hospital and state ICMR institute was sent to the state ICMR institute. Transportation of supplies to district hospitals was done by state ICMR institutes as necessary. FHI was involved in issuing, collecting all necessary documents for transporting the supplies and ensuring enough supplies in each field site and at district hospital.
- 4. Coordination of laboratory activities with ICMR institutes:** FHI was involved in routine coordination IBBA laboratory activities with all state ICMR laboratories. This includes receiving updates of specimen collection at different field sites, testing at district hospital, transportation of specimen to state ICMR institutes, testing at ICMR institutes, recording of results, laboratory data entry and long term storage of specimen at NARI.
- 5. Liaison with consultants:** FHI was involved in coordination of recruitment of consultants needed for different activities of IBBA. This includes consultants from UNC and local consultants for training and manual and SOP development.
- 6. Quality control:** FHI was involved in coordination of internal and external quality assurance program of IBBA. This includes ensuring retesting of 10% specimen at ICMR institutes for Syphilis test done at district hospital and retesting of 10% specimen at NARI for all tests done at different ICMR institutes.

7. **Monitoring and supervision:** One of the key responsibilities of FHI was to monitor the IBBA laboratory activities. This includes periodic visit to field, district hospital laboratories and state ICMR laboratories. During the visit technical assistance was provided as necessary.
8. **Trouble shooting:** FHI was involved in day to day trouble shooting of instruments, supplies and technical assistance. When ever complains received either from ICMR institutes or from NARI, FHI contracted vendors for necessary repair of the instrument or identify local sources for repair.

Liaison with Bill and Melinda Gates Foundation focal person for Monitoring and evaluation grant: FHI maintained close contact with BMGF key person for M & E grant and keep then updated about progress of the laboratory activities. Any change in the test protocol was informed to BMGF and approval was sought before implementation of the change

Laboratory Quality Assurance

Traditionally, National Laboratory Accreditation Programs concentrate quality control and quality assurance in laboratories providing diagnostic testing services for patients. However, Quality Assurance program for disease surveillance, control, evaluation programs are now being recognized as an important component of public health laboratory programs. More and more public health and research laboratories are recognizing the need for quality output “the right test result on the right specimen from the right patient that is accurate, timely and properly interpreted” . This concept can only be achieved through establishment of quality assurance which is defined as “the total process whereby the quality of laboratory reports can be guaranteed”

Four Tenants of Quality Assurance

- A. Quality Assurance focuses on systems and processes
- B. Quality Assurance uses data to analyze laboratory service delivery processes
- C. Quality Assurance is oriented towards meeting the expectations of stakeholders
- D. Quality Assurance encourages a team approach to problem solving and quality improvement

Good Laboratory Practice

Good Laboratory Practices in the context of IBBA survey is when the network laboratories consistently perform laboratory investigations using up to date standard operating procedures, and their reports are consistently reliable and accurate.

Laboratory Standards

- Standard 1- Laboratory Infrastructure (Testing Facility and Operations)
- Standard 2- Human Resources (Organization and Personnel)
- Standard 3- Preventive Maintenance (Facilities and Equipment)
- Standard 4- Specimen Management and Tracking
- Standard 5- Test Validation and Performance Specifications
- Standard 6.-Quality Control
- Standard 7- Laboratory Safety
- Standard 8 – Data Management (Records and Report

Laboratory Quality Assurance

Field Site Laboratory

Field sites in 29 districts and the national highways were selected for conducting interviews of participants and collection of biological samples.

Standards for Specimen Collection sites

1. Infrastructure and Supplies:

- Adequate stock of infrastructure and supplies available at the field site (Annex 1)

2. Human Resources: The field team consist of

- Medical Officer (Leader and supervisor of the biological samples)
- Laboratory technician
- Supervisor, Community liaison and interviewers (3)

Responsibilities of Field Laboratory Technician

- Ensuring adequate stock and maintenance of all consumables and non-consumables required for the biological component of the survey
- Correct labeling of all specimens
- Collection of biological specimens (blood, urine and dry blood spot (DBS)) after appropriate instructions to the participants
- Correct documentation of all specimens (lab forms, etc.)
- Management of gel packs and transportation of boxes
- Proper packaging of all specimens
- Assist the medical officer in collection of specimen from the ulcer
 - Ensuring proper waste disposal
 - Supporting the field team

3. Standard Operating Procedures (SOPS)

A comprehensive field manual on procedures for collection of blood, urine, swabs and dried blood samples and transport of specimens to the district laboratory was prepared.

4. Capacity building and Competencies of the field team

Each district team was trained for xx days on field operations that included selection of site for interviews, consent, and clinical examination, collection of samples, handling and transport to district laboratories.

Table 1: Training Programs for Field Team

Number of Training Conducted:

Andhra Pradesh: 8

Tamilnadu:

Maharashtra

Manipur and Nagaland

Highways

Total Number of field teams attended

Total Number of participants attended

5. Quality Control of Field Operations

1. Infrastructure and Supplies: Available for at least for three days
2. Consent for survey including specimen collection
3. Specimens to be collected are; blood (10 ml), Urine (30ml in 50ml containers), DBS (Five spots per one Sheet x 2), swab if GUD present
4. Aliquot 2ml of urine into urine transport tube
5. Stock register and Indent form
6. Lab submission form
7. Infection control practices- Universal precautions, HBV vaccination

6. Forms for transport of specimens

1. A lab submission form for transport of specimens and waste disposal bags/sharp containers developed
2. Number of specimens in each thermacol box and waste disposal bags to be sent to the district laboratory is checked by the field supervisors/ and medical officer

The following quality parameters were checked during transport of specimens.

Table 2: Quality Parameter Checked during transport of specimen
1. Laboratory forms completely filled
2. Supervisor checked specimens in the box
3. Temperature checked
4. Duplicate forms sent with the box
5. Field team Supervisor, medical officer signed the forms

Level 2: District Laboratories

District laboratories were selected in 29 survey districts to support biological investigation in IBBA survey. The main function of the district laboratory is to perform Rapid Plasma Reagin (RPR) Test for syphilis. Other activities of the laboratory are;

- A) Supply of laboratory logistics to the field team
- B) Receiving of all types of specimens from the field
- C) Freezing the gel packs for the cool box for the field
- D) Sorting of specimens
- E) Ensuring right temperatures
- F) Processing of specimens
- G) Packaging and Transportation of specimens (aliquots) to the state laboratory.
- H) Safe Disposal of waste generated from field and district labs
- I) Monitoring and supervision of field team activities

Pre Analytical Phase

a. Assessment of Laboratory Practices

District laboratories were assessed by the State laboratory manager with support from the National Reference Laboratory and FHI. The criteria used were

1. Availability of laboratory space
2. Sentinel site for NACO sentinel surveillance study.
3. Minimum equipment for RPR test
4. Infection control practices
5. Laboratory reporting practices
6. Quality Control procedures
6. Competencies of laboratory technicians to perform RPR test.

Based on the above criteria, district laboratories sites were selected to participate in the program.

Table 3: District Laboratory Site in IBBA Implementing States

District Laboratory Site	AP	Maharashtra	Tamilnadu	North East	Highways
Medical Colleges	6	3			
District Hospital	-	2			
Private Laboratories	2	-			
Others	-	1			

b. Laboratory Infrastructure: The minimum space and equipment required for performing activities in the district laboratory is

Space

1 room (lab) + 1 store

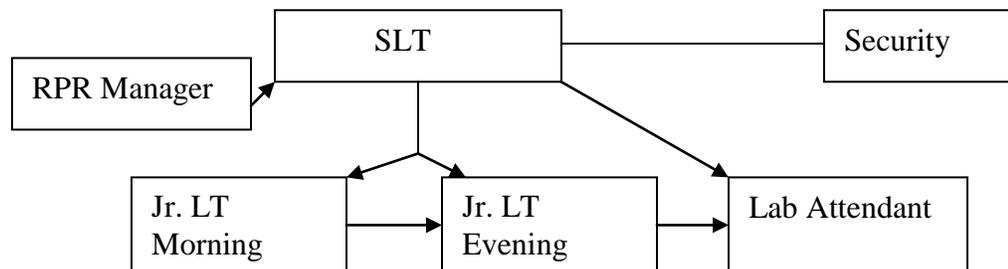
Equipment

1. +4.C Cabinet
2. Freezer of -20 deg C
3. RPR rotator
4. Bio-safety cabinet
5. Autoclave for waste disposal
6. Serum separation centrifuge
7. Micropipette
8. Small oven in sites collecting DBS (for sites where blood is not collected)
- 9.

The IBBA Project provided all equipment and supplies required for the laboratory component.

c. Human Resources (Organization and Personnel).

The generic organogram of the district laboratory for IBBA is shown below



The Senior Laboratory Technician has the overall responsibility of the district laboratory component of IBBA survey.

1. Overseeing aliquoting of serum specimen and proper storage of all samples
2. Performing RPR
3. Preparing RPR reports for dispatch to field
4. Maintaining records
5. Maintaining stores – indenting from state & issuing to field
6. In charge of administrative issues including accounts
7. Training of field LT & Jr. LTs
8. Supervising Junior LTs

d. Capacity Building of Staff (Annex Training Agenda)

Training sessions were conducted for the laboratory staff on RPR test, quality control, reporting and sorting& handling of specimens at the district laboratory. The training session also included biosafety practices, equipment maintenance and transport of specimens to the State laboratories.

Table 5: Capacity Building Programs for the Staff in District Laboratories

State	No Training Sessions Conducted	No Participants attended
AP	8	32
Maharashtra		
Tamilnadu		
North East		
National Highways		

District laboratory technicians received IBBA training on field operations, testing and data management for six days and one day of mock run.

e. Preventive maintenance (Biosafety cabinet, -20C Freezer, 4C Refrigerator, RPR rotator, micropipettes etc)

A preventive maintenance protocol for equipment in the district laboratory and responsible staff was developed and implemented.

- A. **-20C +/- 1Freezer and 4C+/-1 Refrigerator-** Daily temperature log at the beginning of shift
- B. **Bio-safety Cabinet:** Daily cleaning. Air flow, Certification of the equipment on instillation by the company
- C. **Autoclave:** Indicator strip used during operation
- D. **RPR Rotator:** manual reading of rotations 100+/-5 rpm per minute

f. Bio-safety Practices

1. Safe Disposal of Biohazard waste from the field

The bio-hazardous wastes generated during the survey were appropriately taken care at the district laboratory. As the field sites are not well laid out to handle waste and resources are limited, district laboratory was given this responsibility of proper disposal of potential infectious waste. The procedure ensures maximum safety for the staff handling the wastes.

- Sharp disposal containers and the bags (autoclavable and disposable) should be autoclaved at the district laboratory

- Material discarded after autoclave into municipal bin.

- Sharps were transferred into card board box and discarded.

2. Bio-safety practices during Laboratory Testing, Handling and Transport of specimens.

- Adherence to Universal precautions by all staff.

- Disinfection of laboratory waste

- Hepatitis B vaccination of laboratory staff

g. Standard Operation Procedure manual for District laboratory

A district laboratory SOP manual was developed (Annex 3) for use by the laboratory staff.

h. Reagents Supply and Management of Stocks:

The district laboratory has a secure room (away from heat and humidity) for storage of supplies and reagents. This room has dedicated 4⁰C Refrigerated cabinet for cold storage of survey specimens. A protocol was developed for storage of stocks of supplies and indenting of supplies. A stock status register was developed for recording supplies on receipt, balance on hand and distribution. Minimum Stock maintenance and reorder level was developed (Annex 4)

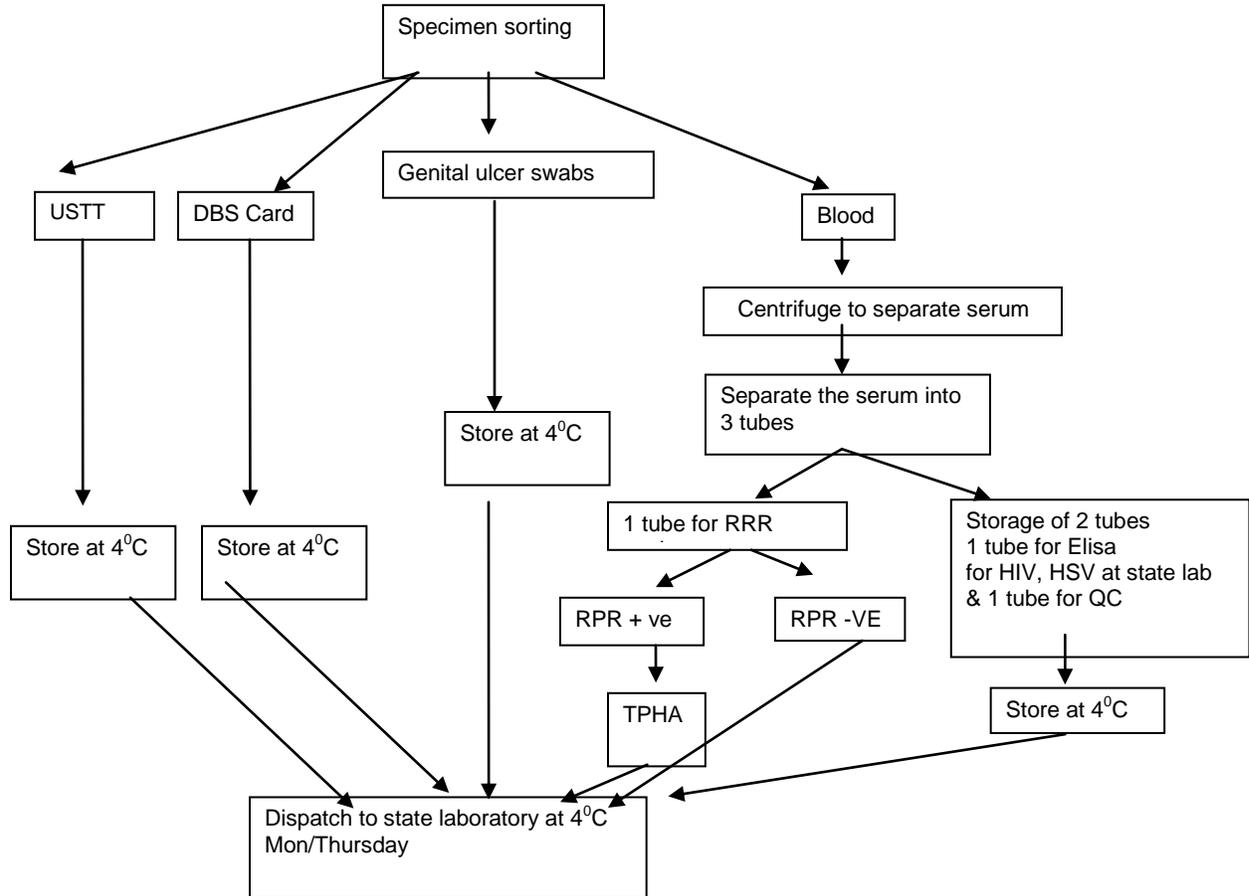
Stock Out in the District Laboratories -----
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Analytical Phase

a. Sample Receipt and Sorting:

- i. District laboratories receive blood in vacutainers/DBS, 2ml urine in transport tubes, ulcer swabs and waste bags.
- ii. Each lot of specimens is accompanied with two copies of lab form.
- iii. Specimens are sorted and stored as shown below

Fig 1: Flow Chart showing Specimen sorting at the District Laboratory



b. Audit of Samples received from the field: During sorting of samples received from the field, an audit (Audit Form) was conducted to determine number and type of samples, sample integrity, correctness of labeling and data in the forms.

Table 6. Audit Form

Quality Indicator
1. Samples received within 24hrs
2. Number of samples recorded in the form and in the box correct
3. Temperature of the box on receipt is maintained
4. Labeling of specimens correct and matches with the list
5. Volume and quality of blood and urine is as per the guidelines
7. Laboratory forms are legible and two forms received
8. Waste disposal bags received
9. Sharp disposal container received
10. Supervisor/Medical officer signature

c. SOP for RPR was prepared which includes

1. Specimen processing
2. Equipment and supplies required
3. RPR Kit (Span Diagnostics India)
4. Summary of test procedure
5. Interpretation
6. Reporting: One Report was sent back to the referral clinic, and another report was sent to State Lab for TPHA

d. Internal Quality Control – Positive and negative control sera supplied in the kit was run along with the test samples. Validity of the test run was determined by the results of control serum samples

e. External Quality Assessment- Retesting at the State Laboratory: In order to validate RPR test results at the district laboratories, all positive RPR samples and 10% of randomly selected negative samples submitted to the state laboratory were retested by the same RPR kit. The concordance and discordant rates between district and state laboratories will be measured.

Table 7 RPR External Quality Control Results

	No Sample tested	District Lab Results		State Lab Results	
		NR	R	NR	R
District Labs					
Andhra Pradesh					
Maharashtra					
Tamilnadu					
Highways					

NR: Non Reactive R: Reactive

f. External Quality Assessment Proficiency Scheme for RPR implemented by the State Laboratory

At the beginning of the survey, a panel of proficiency test for RPR was sent. Results were 100% correct for all district laboratories except for one district laboratory.

Table 8: RPR EQAS Results

	No Panels tested	District Lab Results		State Lab Results	
		NR	R	NR	R
District Labs					
Andhra Pradesh					
Maharashtra					
Tamilnadu					
Highways					

Supportive Supervision: Laboratory Manager at the State Laboratory and/or Laboratory Coordinator, IBBA Reference laboratory conducted capacity building during fortnightly supportive supervisory visits to district laboratories. The activities performed during these visits were;

- A Monitoring of GLP in the district laboratory
- B. Inspection of preventive maintenance logs and RPR register
- C. Inspection of Stock register
- D. Quality control forms of RPR test
- E. Capacity building of staff

Post Analytical Phase

a. Lab reports:

Specimen submission form: A signed copy of specimen submission form was returned within 24-48hrs as an evidence of specimen receipt and feedback on sample integrity. If errors are noted, corrective action will be recommended

Participant RPR Report was sent with in 5-7 days to the referral clinics in the district

Participants RPR report with quantitative result was sent to the State laboratory for TPHA

RPR register was maintained at the district laboratory during the survey period and was sent to the State laboratory.

b. Turn around time as stated below was achieved during survey period

District Laboratory to the Referral clinic —5-7 days

District Laboratory to the State laboratory- Once a week

Specimen Transport to the State Laboratory: All samples received from the field were processed in the district laboratory and were sent to the state laboratory as per the guidelines. Quality parameters/check list as shown below was used to determine conformity to the guidelines.

Table 9. Check List Used while specimen transport to the State Laboratory

1. Four vials (Non Reactive vial, RPR Reactive vial, HSV/HIV/BED vial and QC vial) sent at 4C
2. Preprinted labels correctly pasted
3. Volume – up to ¾ of the cry tube
4. Vials sorted in four cry tube boxes
5. Two copies of Cryo-submission forms completed
5. USTT tube sent at 4C in a Zip lock bag
6. DBS in Zip lock bags
7. District laboratory submission form correctly filled

Level 3: State Laboratories

Four ICMR institutions were selected to implement IBBA survey (Behavioral and biological components) in four states and the national highways. These research agencies have experience in conducting and providing support to biological components of surveys. These institutions have variable levels of laboratory infrastructure and skills/competencies based on primary role of the institutions. These laboratories are expected to perform moderately complex to highly complex tests on samples collected from IBBA survey participants

Pre analytical Phase-

i. Assessment of the State ICMR Laboratories and Recommendations on improvement of Laboratory Infrastructure and Equipment

An assessment of the state ICMR laboratories selected to conduct IBBA survey was done by NARI and FHI/UNC. (Annex 5: Assessment Tool for State Laboratories)

A. National Institute of Nutrition (AP) The microbiology lab would be the designated lab space for this project. The staff has basic experience with PCR and ELISA tests. The necessary equipment is available through various departments, but would need to be centralized in the microbiology labs for the test period. There is ample laboratory space between three rooms to accommodate the large volume of samples to be tested, though some reorganization is needed. The most important renovation for these two rooms is to have the windows sealed, and air conditioning installed to better control the lab environment.

Infrastructure available is:

- Equipment available includes the AB 9700 thermalcycler, plate washer and reader, laminar flow hood, two -20°C freezers, centrifuge, and generator.
- Excellent potential for lab space to accommodate high volume of samples

Recommendations:

Room 1- This room would be used for specimen reception. It has two -20°C freezers, and plenty of space for unloading the thermocol boxes. Table could be cleared to such provide space. Shelves can be added to hold supplies. A -70°C needs to be purchased for specimen storage.

Room 2- This room has a laminar flow hood, and would be ideal as an 'Area 1' room.

- Equipment to left of hood to be removed, and bench space cleared and extended.
- This bench space immediately left of the laminar hood would hold a dead space hood for reagent preparation. The bench space to the left of that would be for organization of specimens.
- Place current microbiological supplies in a centralized location.
- Current bench space behind the laminar hood would hold the centrifuge and heat block for specimen preparation.

Room 3- This room would be ideal as an 'Area 2' used for serology and ELISAs.

- Bench space to left of door upon entry should be reorganized and extended along the left wall and across the back wall to allow for four work spaces for each assay.
- Wall space to right of door extends to the sink in the corner. This space can be used to hold the plate washer and reader.
- Wall with windows has space to hold refrigerator and two desks could be placed perpendicular to provide space for paperwork and data management.
- A computer should be placed at the desks, and communications infrastructure (phone, fax, and internet) should be established. Shelves can be added to provide storage space for supplies

B National Institute of Epidemiology, Chennai TN: NIE had previously done HIV ELISA, but have not for some time. The capacities of current staff were also assessed. Major equipments needed were identified (thermalcycler, plate washer and reader, incubator, -20°C freezers, microfuge, and pipettes. laminar flow hoods). There is adequate lab space which needs to be reorganized.

Recommendations:

- Reorganization of space
- Electrical outlets need to be checked, and the air conditioning needs service or installation. Windows need to be sealed in lab space.
- A partition should be installed to separate the entire floor space in two, so that the back half of the floor would be the lab so the environment can be better controlled (due to open windows and grates).
- One room currently has two smaller rooms partitioned, with one containing a laminar flow hood, and the second is empty. From the second room, the small laminar hood should be moved into the second room. This will become the reagent prep room, and the room with the larger hood will become the specimen prep room. In this larger room will house the micro centrifuge and a heat block (needs to be purchased)
- The large open second room that originally housed the smaller hood and a second laminar hood should be completely cleared, including removal of the remaining large hood.
- There are desks in storage that could be used as bench space. These should be organized around the walls particularly in the second room to allow at least four work spaces for the four tests that need to be performed.
- The second room has a water supply, should be checked, and site needs to install a water distillation unit for a laboratory grade water supply.
- The second room will house the incubator, plate washer and readers, and thermalcycler.
- Shelving needs to be installed in the lab rooms for in-use supplies.
- The area outside these two rooms can be used as the specimen reception and data management area.
- Phone, fax, and internet communications need to be established or serviced.
- There is a room that can be used for storage opposite the lab rooms. This could hold the material supplies for the state and district labs.
- A vertical -70°C freezer needs to be purchased for specimen storage.
- Servicing of the lab equipments

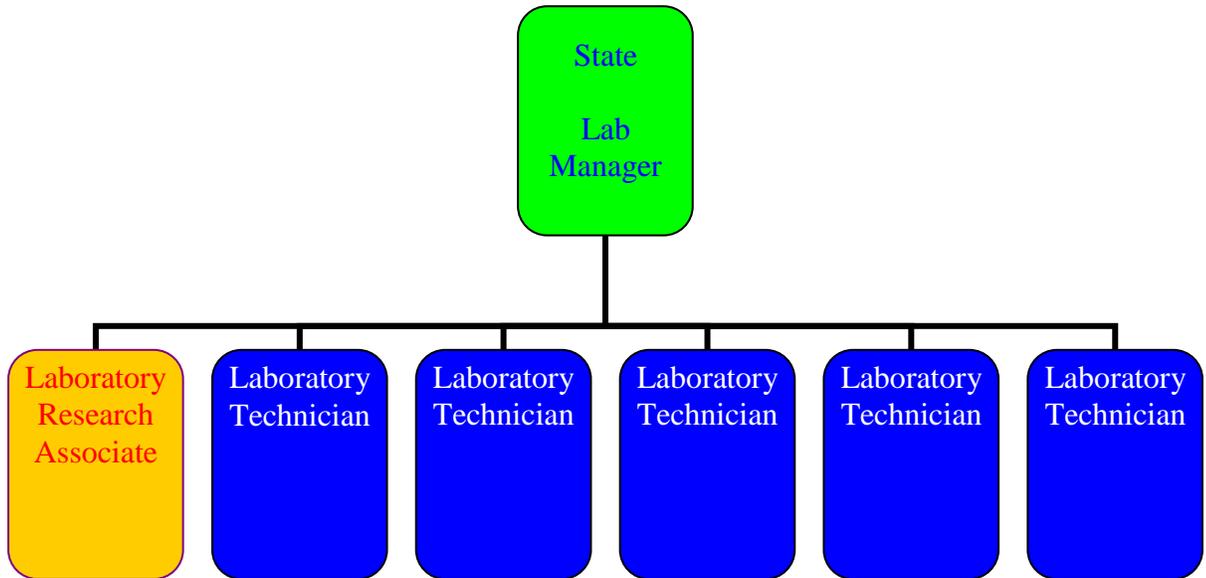
C. Regional Medical Research Council (Manipur and Nagaland). This facility is well equipped for basic research, and is evidenced as such with each department having current state of the art equipment to perform quality research. The site was assessed evaluating the infrastructure and personnel to meet the requirements for this project. The FHI project falls under the area of clinical research, and the labs were assessed critically for quality control as would be required internationally when patient testing is involved.

Recommendations:

- The laboratory will be reorganized to prepare an ‘Area 1 and 2’ based rooms. This includes moving the biosafety cabinet into the larger room. The second biosafety cabinet will be installed in this room, plus the necessary equipment (i.e. centrifuge, heat block, refrigerator/freezer, etc.)
- The Area 1 room will be designed to have bench space upon entry into the room as a specimen reception area which will allow better specimen management.
- Good Laboratory practice needs to be enforced in movement of supplies between the two rooms to avoid contamination.
- Care should be taken to monitor the air flow within the rooms by the air conditioners. Avoid air blowing directly over bench space where samples will be handled.
- All unnecessary equipment in the current rooms should be removed to another room. Moving the autoclave to a central waste processing area is recommended.
- A calibration plate should be purchased for the plate reader, and monthly QC should be performed to monitor shifts in performance between scheduled maintenance calls.
- Records need to be kept in the laboratory of all maintenance performed on the equipment.
- Revise the lab results documentation forms. Forms should include the name of the test, brand of kit, kit lot number, expiration date of kit, date the test was performed, name of tech who performed the test and a place for supervisor the sign indicating review of results.
- A copy of all test results should be kept in the lab.
- Parallel testing should be performed when a new kit lot number is put into use. This is done by testing three known patient samples with both the old kit lot and the new lot. Document and store all files with the QC records.
- Enrollment in an external quality control program for proficiency testing
- Employee files should be maintained that document training, experience, certificates of training, and job description.
- Organizational chart should be devised for the study team.

Outcome of the Assessment: Based on the assessment report and recommendations, all state laboratories were reorganized/refurbished, necessary infrastructure procured, and equipment serviced and calibrated prior to testing.

b. Laboratory Management Structure (Organogram) Roles and Responsibilities.



Responsibilities of State Laboratories:

The state laboratory has an overall responsibility for laboratory activities within the state/ highway. Project The laboratory is headed by a laboratory manager and staff who perform laboratory activities. It will also be the responsibility of the state level laboratories to provide technical assistance and oversight to the district laboratories. The state laboratories will have the following responsibilities to the district laboratories and field teams:

- Must assure adequate supplies of specimen collection materials.
- Monitor usage of supplies and current stores of supplies on site.
- Monitor the use of personal protective equipment for safety to the laboratory technicians, and quality of the test result.
- Monitor proper collection and shipment of samples coming from the field through the district laboratory to the state laboratory, and finally to NARI (Pune) for final quality testing and storage.
- Maintain all quality control records on site for the duration of the project. This includes:
 1. relevant temperature charts
 2. maintenance logs
 3. records of all assay reagent lot numbers used in the laboratory
 4. records of assay control results
 5. assay result log sheets and computer files
 6. back-up of computer result files daily
- Ensure proper storage of samples

c. Capacity Building and Training Program for the State Laboratory Staff.
(Annex: Capacity Building Program schedule)

1. Train the Trainer Program:

- Good Clinical Laboratory Practices (GCLP)
- Laboratory Bio Safety
- Testing Procedure: RPR, TPHA, HIV, HSV2, anti HCV, and HBS_{Ag}
 - i. SOPs
 - ii. Quality control
 - iii. Quality Assurance

■ Hands on training on Gen APTIMA COMBO 2 Assay for amplification and detection of NG and CT. (5 days training including testing of a panel of positive and negative samples)

2. Continuous Laboratory mentoring during supportive supervision visits.

During supportive supervisory visits, mentoring was provided in quality control of Gen APTIMA Combo 2 assay, adherence to product information for interpretation of results and validity of the test (HSV 2 ELISA).

c. Preventive Maintenance and Calibration Protocol- Biosafety cabinet, ELISA reader and washer, eppendorf pipettes, freezers, Gen Aptima etc

Table 10: Preventive Maintenance Check list

Equipment	Activity	Frequency	Preventive Maintenance	
1. Bio-safety cabinet	Airflow	Daily	Y	N
2. ELISA Washer	Calibration		Y	N
3. ELISA Reader	Calibration			
4. Gen APTIMA	Monthly check up	Monthly	Y	N
5. Freezer -20C	Temp	Daily	Y	N
6. Freezer -70C	Temp	Daily	Y	N
7. Thermometer	Calibration	Yearly	Y	N
8. Eppendorf pipettes	Calibration	Yearly	Y	N

e. Selection of Test Kits (TPHA, Treponostica, HIV- J Mitra, Genedia, HBV, HCV, HSV2, Gen Aptima for GC/CT, BED Assay)

A technical panel from NARI, FHI, UNC, WHO and Avahan reviewed published data on sensitivity, specificity, availability of kits, and suitability for use on specimens collected from the participants and made recommendation for use at the state and national reference laboratories.

Table 12: The characteristics of test kits used in the survey

Name of the Disease	Test System/Kit Name	Name of the Manufacturer	Sensitivity	Specificity	Reference
Syphilis	TPHA Syphagen TPHA Kit	Bio kit, Spain www.biokit.com	98.2%	99.6%	MHRA Evaluation Report 2004 (UK)
Herpes Simplex Type 2	Herpes Select 2 ELISA IgG	Focus Diagnostics			
HIV	1 st Test: J Mitra EIA Kit 2 nd Genedia HIV ½ EIA kit	J Mitra			Approved by NACO for sentinel surveillance
N.gonorrhoea /C,trachomatis	Gen Probe APTIMA Combo -2	Roche Molecular Systems	95-98*	98-99*	Two multi center clinical studies on urine samples (male and female) Roche Diagnostics
HIV Incidence	BED CIEA HIV incidence assay	Calypte Biomedical Corp.USA	NA	NA	Only one Assay available.
Syphilis /Chancroid /Herpes	m- PCR	NARI In-house	NA	NA	Orle et al 1996 modified by U.Monitoba
Syphilis (DBS)	Treponostika EIA Kit				
HBV (DBS)	Murex HBV ELISA	Abbott Diagnostics			
HCV (DBS)	Murex HCV ELISA	Abbott Diagnostics			

NA: Not applicable

f. Bio-safety policies and Procedures: Based on the Good Clinical Laboratory Practices, each state laboratories developed a bio safety policies and procedures for implementation. All laboratories staff was trained in safety procedure to be adopted while performing tests and documentation of laboratory related incidents/accidents.

Adherence to Universal Precautions: All staff reported adhering to universal precautions

Hepatitis B Vaccination given to staff: All staff received three doses of HBV vaccination prior to starting testing procedures

Accident/Incident Reports: No reports received on needle stick injuries or laboratory related accidents

g. Sample Receipt, handling, storage protocol

1. Biological Samples (Serum and Urine/ swabs): Serum samples received at 4C and were stored at -20C until shipment to IBBA Reference Laboratory (NARI) once a month on dry ice. Genital ulcer swabs received are also stored at -20 until shipment to NARI.

2. Dried Blood Spots

The state laboratory monitored the collection of DBS and recorded on improperly collected specimens by using Schleicher and Schuell Bioscience Simple Spot Check document to determine valid and invalid DBS collection.

f. Data collection and storage systems-

The State Laboratory forms developed for each testing procedure and results were recorded. Results were also entered in to EXCEL format developed by the IBBA Reference Laboratory (NARI). Data back up protocol developed for each state laboratory.

Analytical

a. **SOPs for each testing procedure** was developed as a component STATE LAB SOP. The testing procedure includes

- i. Materials and Reagents
- ii. Specimen/sample Preparation

Dried Blood Spots (DBS): DBS of all specimens or controls must be at room temperature before they are punched. Specimens or controls stored cold or frozen in sealed bags with dessicant were allowed to equilibrate at room temperature before the bags were opened. The DBS was placed in the bottom of the micro well plate, in the order to be tested, and in a configuration to match the plate as directed by the manufacturer's assay protocol. The laboratory form **was** used to document the elution and testing sequence. Prior to testing, the filter paper was examined visually for completeness elution of sample.

- iii. Steps in Procedure
- iv. Quality control
- v. Reading and interpretation
- vi. Repeat testing of Equivocal samples
- vii. Recording

The tests performed at the state laboratory are;

Serum Samples: TPHA, HSV2, HIV J Mithra and HIV Genedia, and 10% of randomly selected reactive RPR in district laboratory.

Urine Sample: NG/CT Gen APTIMA COMBO 2

DBS: Trepanostika EIA, HSV2, HIV J Mithra and HIV Genedia, Anti HCV (Murex) and HVBs Antigen (Murex)

Only 10 percent of randomly selected serum and DBS samples are tested for anti HSV type 2 antibodies.

b. Quality Control (TPHA, HSV2, HIV JMithra/Genedia, HBS Ag and anti HCV). Each test run, kit controls, blinded known positive and negative controls (supplied by the IBBA Reference Laboratory) were included in the assay.

The validity of the run was determined based on the controls (kit and blinded samples). The QC data was recorded and filed for on going monitoring.

C. External Quality Control Program: Ten percent of the samples from each designated QC cryostorage box sent to NARI from the state laboratories were chosen for external quality control by a designated QC staff (at the IBBA Reference Laboratory NARI).

Table 13: External Quality Control (Retesting) results

<i>Test</i>	<i>No. Tested</i>	<i>Sate Lab Results</i>		<i>Ref Lab Results</i>	
		<i>No Positive</i>	<i>No Negative</i>	<i>No Positive</i>	<i>No Negative</i>
<i>TPHA</i>					
<i>Anti HSV 2</i>					
<i>Anti HIV (J Mithra)</i>					
<i>Anti HIV (Genedia)</i>					
<i>Anti HCV</i>					
<i>HBS Ag</i>					
<i>NG/CT</i>					
<i>Trepanostika EIA</i>					

Rate of Concordant and Discordant for each test was determined and recommendations for improvement suggested.

External Program: During the training period for each state level laboratory prior to the start of the project, proficiency panels (blinded samples) for each assay were sent to the state laboratories for proficiency evaluation. This is to assure that each laboratory is performing the assays adequately. This exercise was implemented and monitored by NARI. Proficiency panels were sent once in every 4 weeks by NARI to the state laboratories, and the results once completed were reported to NARI. Annex 6 shows the results of the State Laboratories.

External Proficiency Results

Laboratory	Panel 1		Panel 2	
	Test System		Test System	
Ref Lab Result				
NIN				
NIE				
RMRC				
National Highways				

d. Supportive supervision: A supportive supervision visit of each state laboratory will occur once a month to audit the GLP record keeping and results performance for the duration of the study period. The inspection will be conducted by a NARI representative or project consultant.

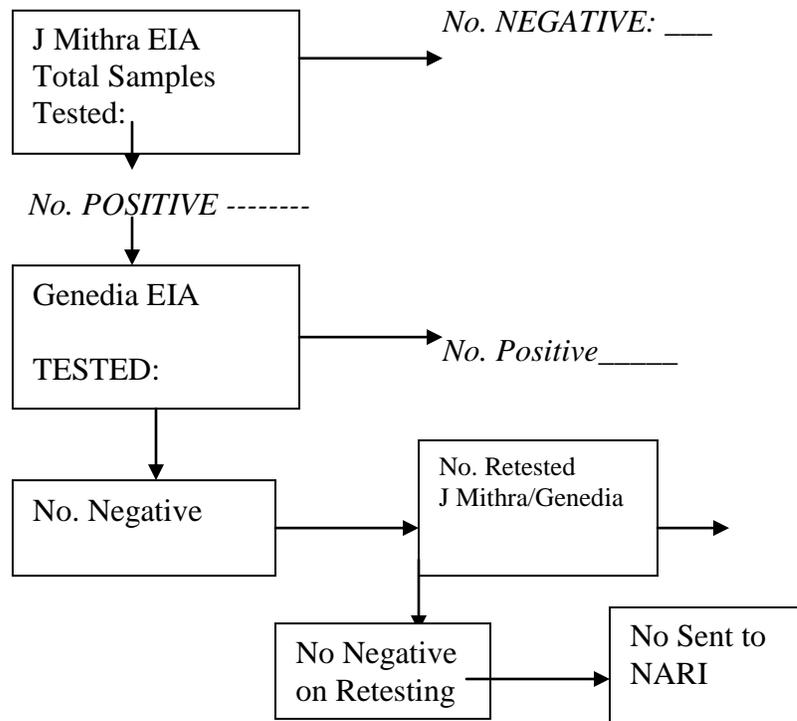
These visits cover

- Review of test procedures and protocol
- Review of test results and kit control performance
- Review of data management (results files and shipment logs)
- Review of QC management (temperatures, maintenance logs, etc)
- Review of oversight and coordination between state and district laboratories

State Laboratory	No Visits	Recommendations/ Corrective Actions
NIN	1	1.Invalid Test Run- HSV2 ELISA and NG/CT Recommendations: Repeat Tests and check reagents and Equipments 2. Labeling of samples: Demonstration on how to label samples
NIE	2	1. Focus on laboratory tests- GLP was reiterated
RMC		
NARI	2	1.Focus on Gen APTIMA COMBO -2 test/ Recommendation: Modification of specimen handling Specimen processing, prevention of cross contamination

Post Analytical

a. HIV Testing Algorithm:



Samples with discordant results were retested by NARI using WB method. Ten percent of negative for HIV will be screened for quality control at NARI

b. Data management.

- Laboratory technicians were conducted tests as recommended in the guideline and the test results were recorded in the designated laboratory forms and register. The laboratory ledger with results was kept in a locked drawer/cabinet.
- The Laboratory Manager and the Senior Laboratory Technician have given the access to specimens and the results.
- The electronic result data sheet was password protected

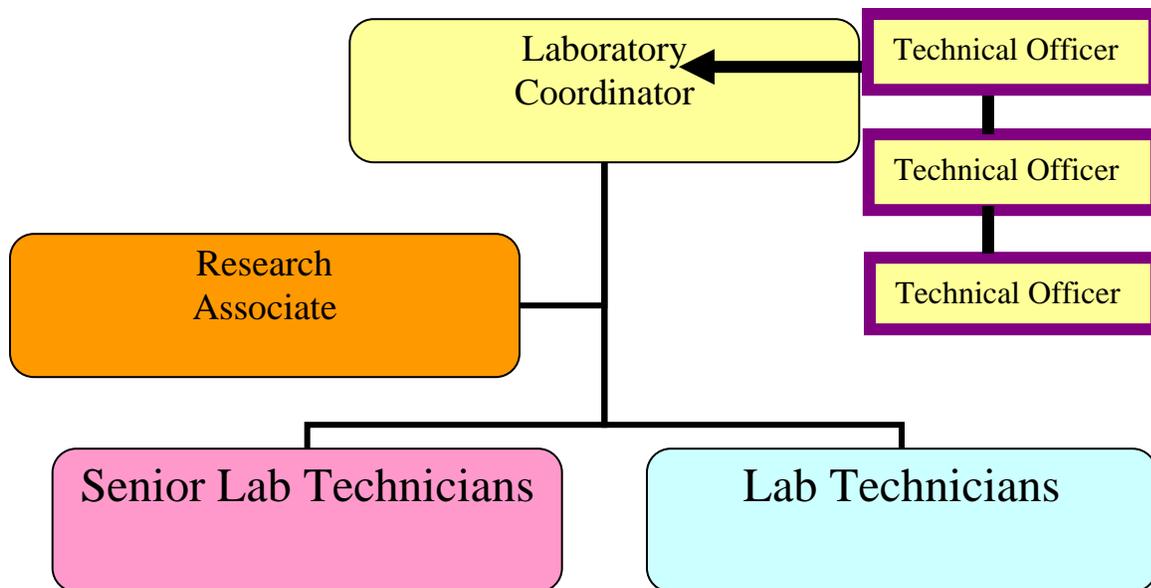
Level 4: IBBA Reference Laboratory (NARI)

The National AIDS Research Institute (NARI), a leadership ICMR organization was established in 1992 with the mission to provide leadership in biomedical research on HIV/AIDS in India and also to compliment and strengthen the National AIDS Control Programmes. The institute has been perusing international research programs in prevention, treatment, care & support and behavioral strategies. NARI has been conducting clinical trials in compliance with Good Clinical Practices. The Institute has been promoting GCLP and has been instrumental in developing national schemes on quality assurance for HIV diagnosis.

The National AIDS Research Institute was given the responsibility of coordinating and implementing the first phase IBBA study in five high prevalent states and highways in India in 2005. The novel component of this survey is an attempt to collect biological indicators of STI (syphilis, antibodies to herpes type 2, NG/CT).

Pre analytical Phase

a. Organization of Reference Laboratory Structure



The role of NARI Scientific Officers is to perform tests on randomly selected QC samples received from the state laboratories. The role and responsibilities of IBBA Reference laboratory are;

1. Coordination of IBBA Laboratory activities in collaboration with the ICMR state laboratories
2. Conducting train the trainer workshop on IBBA laboratory procedures
3. Performing high complexity laboratory tests (BED ELISA, mPCR and HIV-WB
4. External Quality Control and Proficiency testing
5. Long term storage of samples
6. Data analysis and feedback.

b. Capacity Building and training on Good Laboratory Practices

The University of North Carolina, FHI and NARI developed training program on Good Laboratory practices and performance of tests selected for IBBA survey and implementation of Quality systems in the state laboratories.

c. Sample receipt, handling and Storage:

Two cryovials per participant were received on dry ice once a month for long term storage and performance of external quality control tests for HIV, HSV as well as performance of BED ELISA for HIV incidence. Sample is checked against cryo- submission form and stored at -20C until testing. All QC vials were logged into Storage form and placed at -70C.

d. Preventive maintenance of equipments:

NARI maintenance Department provided support for monitoring of all laboratory equipments as per the preventive maintenance guidelines.

e. Data storage and retrieval system:

IBBA Laboratory data was recorded in a pre determined laboratory forms and then transferred to computerized excel data sheets.

f. Policy on correction of wrong labeling

g. Guidelines on Changing labels and numbers in data forms (Annex : Guidelines on Changing Labels and PID No)

An operational guideline on changing labels on cryovials and PID No in the form as well as computer was developed and implemented. The guidelines consist of

1. Procedure for labeling cryo-storage vial
2. Changing PID NO in documents
 - a. field submission form
 - b. Transport sheet
 - c. All Lab forms including registers
3. Data modification on the long-term storage forms

Analytical Phase

a. Retesting Protocol (sample selection criteria, resolving discordant results)

Ten percent of randomly selected (SPSS Program) samples (serum and DBS) received were the state laboratories were re tested using the same method and kit as a part of external quality control for;

1. HIV ELISA (J Mithra and Genedia EIA)
2. Anti HSV2 antibodies EIA (Focus Diagnostics)
3. Anti T. pallidum antibodies EIA (Treponostika)
4. Anti HCV antibodies EIA (Murex 4.0)
5. HBV S antigen (Murex3.0)
6. Gen Aptima Combo-2 NG/CT Assay

The concordant and discordant rates were calculated. The result obtained by the reference laboratory was considered as the final and entered in to the computer database and laboratory forms.

b. External Quality Assessment Schemes (CAP and other institutions)

NARI microbiology and molecular diagnostic laboratories participate in Proficiency testing (EQAS) conducted by the College of American Pathologists USA. The following panels are being tested at NARI

1. HIV EIA
2. HCV and HBV
3. HSV
4. CD4
5. TPHA
6. NG/CT

Table 13: IBBA (NARI) EQAS Proficiency Results

Type of Test	2005			2006			2007		
	1 st	2 nd %	3 rd	1 st	2 nd %	3 rd	1 st	2 nd %	3 rd
HIV									
HCV									
HBs Ag									
TPHA									
NG/CT Assay									

c. BED CIEA Test for Estimation of HIV incidence:

The Calypte HIV-1 BED Incidence EIA is an in vitro quantitative enzyme immunoassay for the determination of the proportion of HIV-1 specific IgG in a given serum or plasma specimen with respect to total IgG as an aid in determining the elapsed time since HIV-1 infection occurred. The Calypte HIV-1 BED Incidence EIA is recommended to be used for public health purposes

only, such as for population incidence estimates to assist in prevention programs, targeting resources, monitoring and evaluation, and identifying high risk cohorts for prevention research, including vaccine trials. The assay has been evaluated for use with the dried blood spots by CDC. An algorithm for interpretation of test result was developed and used in IBBA survey.

d. mPCR for T.pallidum, H.ducrei and Herpes simplex virus type 2

This method was established based on Orle et al, 1996, and as modified by Ian Maclean (the Department of Medical Microbiology of the University of Manitoba).The GUD m-PCR assay permits simultaneous PCR amplification of *H. ducreyi*, *T. pallidum*, and HSV-1 and 2. The Master Mix reagent contains biotinylated primers that are pair specific for each of the targets. The detection of amplified DNA is performed using target-specific oligonucleotide probes that permit the independent identification of *H. ducreyi*, *T. pallidum*, and HSV-1 and 2 amplicons.

Post Analytical Phase

- a. Laboratory Reporting:*
- b. Turn around time*
 - a. Testing for NG/CT by Gen APTIMA COMBO -2 within 90 days of sample collection*
- c. Long term Sample Storage and Access policy (to be developed)*
- d. Guidelines on use of repository of specimen for research*

References

Annex 2. : Infrastructure and Supplies for Collection of Specimens in the field

Infrastructure	Lab Supplies
1. Chairs (Participant, MO and LT)	1.Vacutainer tubes
2. Working table	2.Needles and needle holders
3. Examination table/cot	3.Tourniquet
4. Privacy(curtain)	4. Alcohol swabs
5. Examination light	5. Containers for collection of urine (50ml)
6. Running water (6. Urine transport tubes with disposable pipettes
7. Soap	7.Dacron swabs and tubes for Dacron swabs
8. Waste disposal bins (three colour coded if possible)	8.Sterile cotton balls
9. Needle destroyer	9. Sterile normal saline (0.9%)
10.Cabinets for keeping stocks of supplies and stationery	10Thermacol boxes with sponges and thermometer
11. Small Desk for thermocol box for transport of specimen	11.Lab forms
	12. Scissors, cello tape and package tape
	13.Permanent markers
	14.Tissue roll for specimen packing
	15. Gel packs (frozen)
	16.Zip lock bags (small and big)
	17. Disposable gloves
	18.Labels with participants ID No.
	19. Disposable bags and Autoclave bags
	20. Puncture proof container for needle disposal
	21.DBS paper (No. 903, Schleicher and Schuell) with envelopes
	22.Lancets
	23. Large low gas permeable zip lock bags
	24. Desiccant pouches
	25.Humidity indicator card

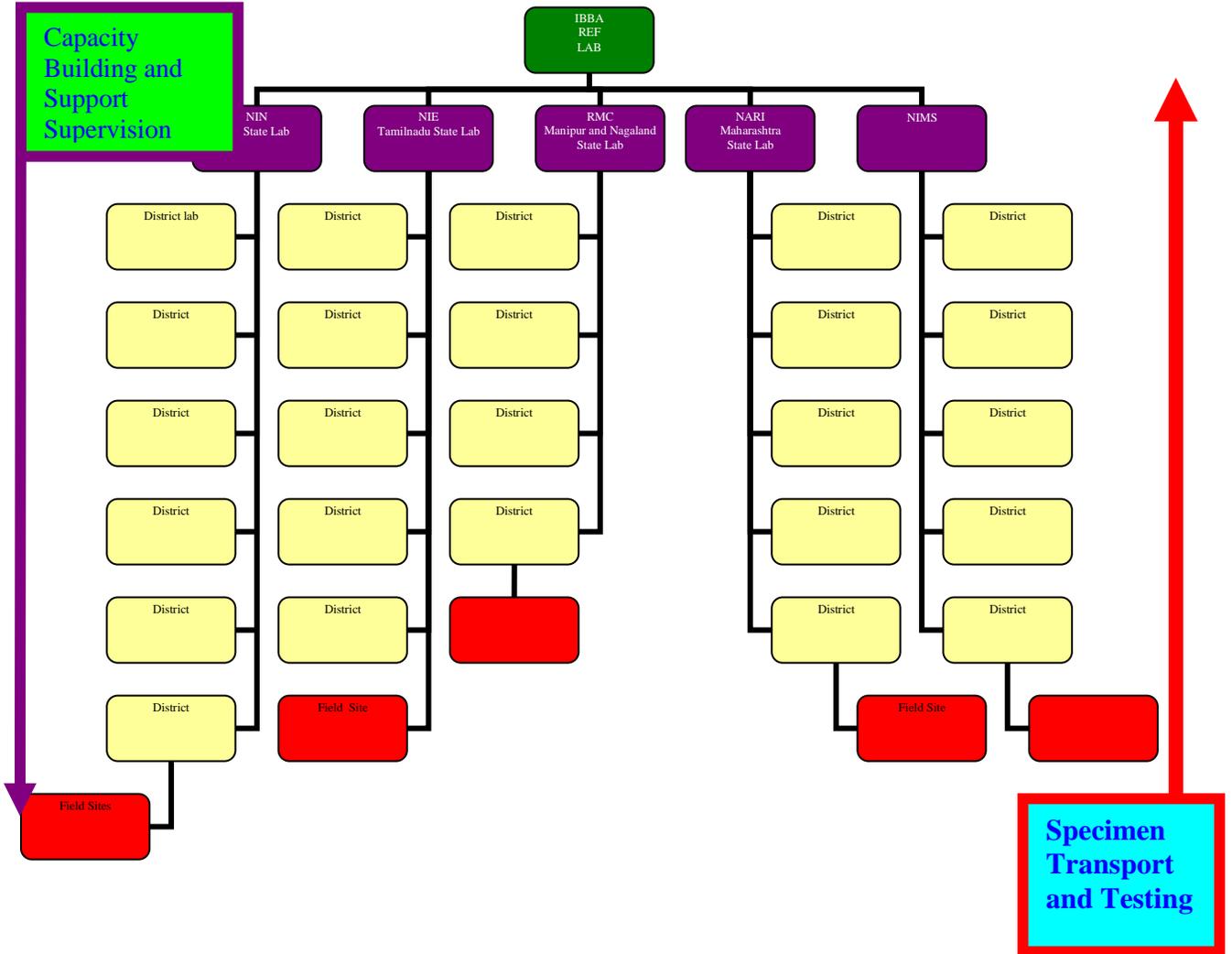
Annex 3: Standard Operating Procedure Manual for the District Laboratory and State Laboratories

CHAPTERS
CHAPTER 1: INTRODUCTION
CHAPTER 2: SPECIMEN RECEIVING AND SORTING
CHAPTER 3: SPECIMEN PROCESSING
CHAPTER 4: SPECIMEN STORAGE
CHAPTER 5: LABORATORY TESTING
CHAPTER 6: TRANSPORTATION OF SPECIMENS TO STATE
CHAPTER 7: STORES AND SUPPLIES
APPENDICES

Annex 4: Minimum stock maintenance and order sheet at district laboratory

SN	Item	Minimum	Reorder
1	Vacutainers	130	260
2	Vacutainer holder	2	4
3	Alcohol swabs	130	260
4	Band aids	130	260
5	Tourniquet	2	4
6	Pre-printed label	For 130 subjects	For 130 subjects
7	Markers	5	10
8	Small zip-lock bags	500	1000
9	Gloves	1000 pairs	2000 pairs
10	Urine container	130	260
11	USTT	130	260
11	Cello tape	10 rolls	20 rolls
12	Packaging tapes	10 rolls	20 rolls
13	Gel-pack boxes	50	100
14	Large zip-lock bags	100	200
15	Dacron swab with container	50	100
16	Scissors	5	10
17	Sterile cotton balls	50	100
18	Disposable pipette	130	260
19	Cryo storage vials	1000	2000
20	Cryo storage boxes	20	40
21	Bleach solution (1 liter bottle)	10	20
22	Sterile saline 500ml bottle	2	4
23	RPR test kit	1	2
24	200 ul tips	250	500
25	Discarding beaker	5	10
26	Sharp container	2	4
27	Autoclavable bags	50	100

Annex 1: IBBA Laboratory Network



IBBA Ref Lab State Lab District Lab Field Site Lab

Annex 5 : State Laboratory Assessment Tool



PPD_Lab_Audit_Shell
_106_doc.pdf

Annex 6: Field and district training programme for biological component of the IBBA

Duration of Training: 3 days

Day 01:

S. No.	Time	Session
1	10.00 to 10.15 AM	Distribution of Materials and Pre-assessment Welcome note
2	10.15 to 11.00 AM	Introduction & Over view to IBBA project
3	11.00 to 11.45 AM	Tests to be conducted in IBBA project
4	11.45 AM to 12.00 Noon	Tea break
5	12.00 to 12.30	Field Activities: Discussion on work flow and responsibilities in the field
6	12.30 to 1.15 PM	Bio Safety Practices in field and Laboratory
7	7.15 to 2.00 PM	Lunch
8	2.00 to 2.30 PM	Collection of Urine Specimens
9	2.30 to 3.00 PM	Collection of Blood Specimens
10	3.00 to 3.30 PM	Collection of Ulcer Swabs
11	3.30 to 4.00 PM	Filling of Laboratory submission form proper packaging and transportation
12	4.00 to 4.15 PM	Tea Break
13	4.15 to 4.35 PM	Proper Waste Disposal in the field
14	4.35 to 5.00 PM	Queries and Discussion

Day 02

S. No.	Time	Session
1	10.00 to 10.45 AM	Good Clinical Laboratory Practices
2	10.45 to 11.00 AM	Specimen receiving and sorting in the district lab
3	11.00 to 11.15 AM	Tea Break
4	11.15 to 12.15 PM	Specimen Processing in district lab
5	12.15 to 12.30 PM	Specimen storage and transportation in district lab
6	12.30 to 12.45 PM	Stores and supplies at District lab
7	12.45 to 1.15 PM	Post TEST Assessment
8	01.15 to 02.00 PM	Lunch Break
9	02.00 to 04.00 PM	Hands on training on sample collection in field and processing in district lab (division into three working groups)
10	04.00 to 04.15 PM	Tea Break
11	04.15 to 05.00 PM	Discussion and Post-Test assessments

Day 3: RPR Test

1	Lecture on RPR Test
2	Demonstration of the test
3	Bench training
4	Quality Control
5	Interpretation
6	Reporting
7	Specimen Storage

Annex : 6

PRE ASSESSMENT TEST

- 1) What is your work experience in drawing the blood by vacutainer?
- 2) Are you aware of the universal precautions while handling bio-hazardous waste?
- 3) Are you aware of bio-hazardous waste disposal policies
- 4) Do you understand the meaning of sexually transmitted diseases?

POST ASSESSMENT TEST

Please tick mark the correct choice. There is only one correct answer per question.

- 1) The study is going to detect the following diseases except
 - a) Chlamydia trachomatis and Neisseria gonorrhoeae
 - b) HSV-II
 - c) HIV
 - d) Treponema pallidum
 - e) All of the above
- 2) The following have to collected from the participants
 - a) urine
 - b) Venous blood
 - c) Ulcer swabs
 - d) Dried blood spots in IDUs
 - e) All of the above
- 3) The urine to be collected from participants should be
 - a) 24 hrs. urine collection
 - b) Mid stream urine
 - c) First void urine
 - d) Random urine
 - e) Any of the above
- 4) The first void urine means that the participant should have not urinated for
 - a) 30 minutes
 - b) 2 hrs
 - c) 8 hrs
 - d) 5 hrs.
 - e) 6 hrs
- 5) Swabs to be taken from females should be
 - a) Low vaginal swab
 - b) High vaginal swab

- c) Cervical swab
 - d) Urethral swab
 - e) External ulcer swab
- 6) The sample to be sent for PCR to the national laboratory;
- a) First void urine
 - b) Ulcer swab
 - c) Urine after centrifugation
 - d) Serum sample
 - e) Dried blood spot
- 7) If participant reports more than one external ulcer then
- a) Swabs should be collected from each ulcer and put each swab in one tube each
 - b) Swabs should be collected from each ulcer and put all the swabs in the same tube
 - c) Patient should undergo internal examination
 - d) Samples should be collected from any one ulcer
 - e) Patient should be referred to hospital
- 8) Specimens should be transported from field to district laboratory at
- a) 4°C - 8°C
 - b) -20°C
 - c) Room temperature
 - d) 2°C - 30°C
 - e) At any temperature
- 9) The thermocol box should be packaged in the following way from top to bottom
- a) Gel pack, Perforated sponge, Gel pack, Sample, Thick sponge
 - b) Thick sponge, gel pack, gel pack, sample, perforated sponge
 - c) Gel pack, perforated sponge, sample, perforated sponge, gel pack, thick sponge
 - d) Perforated sponge, gel pack, sample, perforated sponge, gel pack, thick sponge
 - e) Thick sponge, gel pack, perforated sponge, sample, perforated sponge, gel pack
- 10) Correct storage conditions of the samples in the district before transportation to state lab are
- a) Urine, -20°C
 - b) Urine, 4°C
 - c) Swab, Room temperature
 - d) Blood, room temperature
 - e) Swab, -20°C
- 11) Correct sample processing in district lab
- a) Separate serum and aliquot into three vials, aliquot urine transport media into two vials, store swab without further processing
 - b) Separate serum and aliquot into two vials, aliquot urine transport media into two vials, store swab without further processing
 - c) Separate serum and aliquot into three vials, aliquot urine transport media into three vials, store swab without further processing

- d) Separate serum and aliquot into three vials, aliquot urine transport media into two vials, store swab after suspending it in buffer provided
 - e) Store blood at 4⁰C and no serum separation, aliquot urine transport media into two vials, store swab without further processing
- 12) Urine collected in urine container in the field should be
- a) Stored and transported at room temperature to district lab
 - b) Stored and transported at 4⁰C to district lab
 - c) Transferred to urine transport media and then transported at 4⁰C to district lab
 - d) Transferred to urine transport media and then transported at room temperature to district lab
 - e) Any of the above choices.
- 13) Waste generated at field should be
- a) All waste should be thrown in the municipal bin
 - b) All waste should be packaged in a bag and sent to district laboratory
 - c) Waste should be segregated into biohazardous waste in red autoclave bags and non-biohazardous in black disposal bags and transported to district laboratory
 - d) Waste should be segregated into biohazardous waste in black disposal bags and non-biohazardous in red autoclave bags and transported to district laboratory
 - e) Any of the above.
- 14) Blood collected in the field should be in
- a) Plain red top vacutainer
 - b) EDTA lavender top vacutainer
 - c) ACD yellow top vacutainer
 - d) Plain cryo-storage vial
 - e) Either by using a vacutainer or needle and syringe.
- 15) In the district laboratory specimen should be processed
- a) On the normal working bench
 - b) In a horizontal laminar flow hood
 - c) In a vertical biosafety cabinet
 - d) On a table top
 - e) In any convenient location

Name of the documents:

1. Field lab submission form.
2. Transport sheet from dist lab-
 - a) RPR Negative
 - b) RPR positive
 - c) HIV,HSV2
 - d) QC
 - e) Urine
3. Serum vials of
 - a) RPR Negative
 - b) RPR positive
 - c) HIV,HSV2
 - d) QC
4. Urine QC vial.
5. HIV test protocol sheet.
6. HIV locations register.
7. HSV2 protocol sheet.
8. HSV2 10% list.
9. TPHA test protocols.
10. RPR locations register.
11. Urine test protocol registers.
12. Gene-Aptima test-machine copy.
13. Urine QC location registers.
14. QC HIV test-
 - a) List of No for QC.
 - b) Test protocol.
15. QC of TPHA test
 - a) List of No for QC
 - b) Test protocol.

Computer entries of-

1. List of received samples
2. RPR test results
3. HIV test results.
4. HSV2 test results.
5. Urine test results.
6. Result of HIV QC.
7. Result of TPHA QC.