GUIDELINES

for Assuring the Accuracy and Reliability of HIV RAPID TESTING:

Applying a Quality System Approach

WHO

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CENTERS FOR DISEASE CONTROL AND PREVENTION

Guidelines for Assuring the Accuracy and Reliability of HIV Rapid Testing:

Applying a Quality System Approach

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Table of Contents

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Preface

Serologic tests to detect the presence of antibodies to the human immunodeficiency virus (HIV) in individuals play an increasingly important role in the efforts to address the global AIDS epidemic. With the exponential growth of programs for prevention of mother to child transmission, patient care and therapy, and with increasing emphasis on prevention of HIV infection and on blood safety, these tests will be an essential tool for diagnosis. It is estimated that 100 million people will have to be tested annually to meet the objectives of recent major HIV/AIDS initiatives. Some of this testing will be performed using the standard enzyme immunoassay assay (EIA) technology, but in many situations, rapid tests for HIV will be the most efficient, and perhaps the only feasible, way to provide information about HIV status.

Accuracy and reliability of diagnostic/laboratory testing will be critical to the success of HIV/AIDS programs. In order to ensure this reliability and reduce errors to a minimum, a quality system that addresses all aspects of the testing is essential. The quality system is important in any laboratory or testing site, and applies to all testing and activities, including simple-to-perform tests. In sites conducting only HIV rapid tests, it is also essential to have in place all the elements of a quality system.

The rapid, simple HIV tests that are addressed in this document are single-use, disposable devices that may be used to directly test whole blood specimens, serum, plasma, and/or oral fluids. These single-use devices present unique challenges – testing is often performed by persons without formal laboratory training, there may be no residual specimen that can be checked or re-tested, conventional quality control methods cannot be used, and there are special problems associated with efforts to provide conventional proficiency testing.

The purpose of this document is to establish guidelines for applying quality system essentials to HIV rapid testing. It is intended to provide assistance to all persons involved in policy development, planning, and implementation of HIV rapid testing. The document should be useful to government health officials, those responsible for HIV/AIDS programs and for managing voluntary testing and counseling sites. It also provides information useful for testing personnel, both trained laboratory technologists and those with no laboratory training.

As HIV rapid testing is initiated and/or expanded in large numbers of testing sites, it will be very important to implement these guidelines, including the important monitoring processes, to assure quality and reliability of test results.

Background

All laboratory testing, including rapid/simple testing for HIV, consists of a series of processes and procedures that must be carried out correctly in order to obtain accurate results. An approach that monitors all parts of the testing system is needed to ensure the quality of the overall process, to detect and reduce errors, to improve consistency between testing sites, and to help contain costs. This approach to laboratory quality, called a **quality system**, is defined as the organizational structure, resources, processes, and procedures needed to implement quality management of the laboratory or testing site. The quality system includes policies, quality assurance, quality control, and quality improvement, and in this document is divided into twelve essential elements as described in the Clinical and Laboratory Standards Institute (CLSI) document HS1-A, "A Quality System Model for Health Care."

The performance of HIV rapid testing presents special challenges when undertaking measures to improve test reliability and accuracy. These need to be considered when developing a plan for implementation of a quality system.

In many instances, HIV rapid testing will be conducted by health care professionals who do not have specific laboratory experience, or by lay counselors with no formal health care training. The training program for these non-laboratory staff members must provide all needed laboratory skills, including specimen collection and laboratory safety. Sufficient time for practical hands-on work, as well as some measure of competency on completion of the training, will be very important.

Quality control traditionally includes testing specimens of a known value using the same reagents and equipment that are used for the specimens being measured. Since rapid/simple HIV test kits are single-use devices, this approach is not possible. Quality control specimens must therefore be used in a manner that monitors the correct performance of the test by the operator and the ability of the test kits to work properly. While it is not possible to test each kit, quality control specimens can be used to detect damage of an entire batch or lot number of kits by improper storage or handling, or through manufacturing defects. The recommendations in these guidelines take these factors into account.

External quality assessment (EQA) for HIV rapid testing has frequently been conducted by re-testing a sub-set of the specimens using EIA. Several models for this re-testing are in use. Some sites are using a random sampling of 5% or 10% of all specimens; others are re-testing all positive specimens and 5% or 10% of all negatives. A careful statistical analysis reveals the limitations of this kind of re-testing except in sites with large numbers of specimens. In addition, the re-testing process can present a considerable burden to the reference laboratory charged with the responsibility of conducting these tests. These guidelines emphasize on-site monitoring as another means of performing external quality assessment.

QUALITY SYSTEM GUIDELINES

1.0 Organization and Management

A strong commitment from top-level managers is essential for success of the overall quality program. This commitment is important at all levels, and national laboratory leaders will need to both provide strong leadership from the national level, and also to motivate and help laboratory managers throughout the country to understand the system and commit to its success.

1.1 Responsibilities at the national level

1.1.1 Establishing a laboratory quality system

Implementing a quality system requires commitment from the top levels of management. The Ministry of Health, including national laboratory leaders with appropriate government authority, should:

- Establish a national quality system. This should include
  – a national office of quality assurance or quality management
  – identification of a national quality officer or manager
  – identification of a multisectorial working team, in order to extend the quality system to all aspects of testing practices and to avoid vertical decisions and assessments
- Extend the quality system to all tiers (central, regional, district, point of service) of the laboratory network, and to involve all service providers at all levels.
- Extend the quality system to all laboratory testing, including HIV serology testing.

1.1.2 Planning for management of HIV rapid tests

There must be an overall, country-wide plan for the management of HIV testing, including the roles of HIV rapid tests. The following steps will be needed in establishing this plan:

- National policies must be established for the use of HIV rapid tests. Issues to be addressed include:
  – Use of rapid testing as an alternative to EIA. When and where is this appropriate?
  – Personnel issues. Who will be allowed to perform HIV rapid testing, and what training and certification will be required? How will appropriate supervision be provided?
  – Legal requirements that might apply to testing. Examples include country requirements for existing personnel certification as well as existing national laboratory and safety standards.

  - Evaluation of HIV rapid test kits that will be used in the country and establishing an algorithm to be used for testing.
  - Linkages between EIA or other confirmatory methods and rapid tests.
  - Development of a standard operating procedure to be used in all testing sites.
  - Confidentiality issues.
  - Requirement for corrective or remedial action.

- A strategic plan for implementation of HIV rapid testing should be developed. This plan will include provision for training of personnel and for continuously monitoring and improving the testing process. It is important to establish a timeline as well as processes to deal with many elements of the quality system.
- Monitoring processes should be established which can identify problems and confirm that the system is working. There must be a plan for solutions to the problems, and a record kept of corrective actions taken.

1.2 Responsibilities at the site where testing occurs

At the laboratory or point-of-care facility such as a voluntary counseling and testing center (VCT) there must be provision for oversight of the testing, and for ensuring that the necessary staff and supplies are available, and that confidential record systems are established and maintained. Steps in this process include the following:

- Management of the HIV testing quality system program at each site must be assigned to one person; this person may be designated as a quality officer. The responsibility should be assigned to someone with authority to make and implement decisions, with strong knowledge of HIV testing procedures, and with a complete understanding of the quality system essentials that need to be addressed. In some settings, one quality officer might serve several sites. The quality officer should have a clear channel of communication to the Ministry of Health or policy body, as well as with all staff who are performing testing, so that any changes in procedure or other important information can be shared in a timely fashion.
- A standard operating procedure must be available at each site. This step-by-step set of instructions that outlines all the processes for conducting the testing, including on-site rapid tests, must be accessible to everyone who performs tests.
- Local management must ensure that all testing is performed by staff who are trained and certified according to the national requirements. The quality officer must also have a plan for evaluating the competency of personnel performing testing, both initially and on an on-going basis at appropriate intervals. If there is no national certification program, local management must ensure that staff are trained and are competent to perform HIV rapid tests according to these guidelines or national requirements.
- Oversight of the recordkeeping system must be provided.
- The quality officer should ensure that all other components of the quality system are in place before testing is begun at a site. No testing should be conducted until the site can be demonstrated to be properly prepared.

2.0 Personnel

The most important resource in any health care setting is the working staff. It is essential that this valuable resource be given the tools needed to perform testing so that accurate and reliable test results are obtained. Direct support for the testing staff will include initial training for the testing and some method for periodic evaluation of each person. This periodic evaluation is conducted to ensure that all protocols, including SOPs, are being followed and that testing can still be performed accurately. Unlike other laboratory testing, HIV rapid tests will frequently be performed by individuals who do not have training in laboratory technology. Therefore, careful attention must be given to the training provided, especially for non-laboratory personnel.

2.1 Training for persons who will be performing HIV rapid testing

2.1.1 Initial training programs

Frequently, training of large numbers of staff will be accomplished at the time of broad implementation of the testing, for example the establishment of a new group of VCTs or the expansion of Prevention of Mother to Child Transmission (PMTCT) programs. A standardized training program for non-laboratory staff should be developed and implemented at all levels of service delivery. Experience in some countries suggests that the training program can be conducted over a two to five day period, depending on previous education and training of those being trained. The training for test performance is most often integrated into a program that includes all aspects of counseling and testing.

Training should include:

- How to perform the test, including all parts of the process – from specimen collection to the reading and interpretation, recording, and reporting of results. The appropriate use of external control materials must be included.
- A hands-on component, with all participants actually carrying out specimen collection and testing procedures.
- How the test is used in the program at the site
- The importance of the quality essentials
- The importance of biohazard safety procedures, including waste disposal.
- An assessment of ability to safely and accurately perform the testing.

During the period immediately following training, when new staff first begin testing of clients, provision for continuous mentoring is an important tool.

Appendix A provides additional details on the content of the core curriculum for HIV rapid testing training. Also available are training workshop materials, including a participant manual, a trainer's guide, and other tools prepared by WHO and CDC. The use of these materials may require a different approach depending on the skill level of the audience.

2.1.1.2 On-going plan for training of new staff

The national training plan must make provision for the training of new staff as they are hired and added to the workforce. It will be critical to the maintenance of quality, reliable testing, and may sometimes be a challenge, as the training may have to be done for one or two persons. However, all new staff should be provided the same training program that is used for initial training.

2.2 Additional training needs

2.2.1 Training for those who will provide oversight and monitoring

In many settings, oversight and monitoring of the quality elements for HIV rapid testing will be performed by local laboratory staff. When choosing personnel to carry out these tasks, consideration should be given to good communication skills, an ability to organize and carry out jobs, motivation and reliability, and skill in problem solving. Additionally, the tasks involved require a set of skills and knowledge not always included in the initial laboratory technology training for these persons. Provision must be made for training in how to monitor, perform on-site evaluations, and provide advice and help to testing personnel.

2.2.2 Training trainers and program staff

Training plans should make provision for training those who will have the responsibility of conducting training sessions. Also, it will be important to make available training for program staff who may not be performing the testing but who need information about how the tests are conducted and how they work.

2.3 Certification and early monitoring of performance

The Ministry of Health should develop a program for individual certification of non-laboratory staff. An important first step is to establish criteria for the successful completion of a standardized training program. This should

include a written examination on material presented during the training and a demonstration of competency to conduct HIV rapid tests by testing a proficiency panel.

In some countries certification is awarded only after successful completion of a recognized, standardized training course, followed by another test of competence upon assignment to a testing site. This competency check could be performed by:

- Before assignment to a new test site, testers perform an internship at an established site under direct supervision
- Testers performing rapid testing at new test site where test performance is observed by experienced and trained personnel who are temporarily on-site. This direct observation should occur for at least 50 specimens, or until the individual has demonstrated competency based on site checklist.

Persons without formal laboratory training who are not certified through an official program should not be allowed to perform HIV rapid tests.

In the first few months following training, new staff should receive routine mentoring. On-site monitoring visits should be frequent, since these give opportunities to help new staff improve skills and recognize problems when they occur. New staff will need to know how to seek help from laboratory professionals in solving the problems identified.

2.4 Evaluation of personnel competency on an on-going basis

A method for continuous monitoring of personnel competency needs to be developed. Important elements to evaluate for both newly trained personnel and for those staff who have been performing testing include:

- Check on ability of the personnel to set up a testing environment consistent with that described in the SOP, including temperature and storage conditions, labeling and recording instruments, and safety considerations.
- Observe test performance to ensure that all steps are performed correctly and that the correct result on known specimen(s) can be obtained.
- Provide known samples for analysis by staff who perform testing to ensure that the correct result can be obtained.
- Evaluate ability to interpret quality controls and patient/client results, including the ability to interpret HIV status based on the established testing algorithm.
- Check on ability to properly carry out all record keeping procedures.

Initial and on-going competency evaluation is required for all staff performing testing. There should also be a mechanism for correcting any problems or deficiencies identified during the monitoring visits. The on-site visit can also be used to identify training needs and provide important refresher training.

3.0 Equipment

One of the great advantages of using rapid, simple technology for HIV testing is that little or no equipment is required. However, in some settings the use of whole blood or serum may require a centrifuge and pipetting devices. In this case, a plan for calibration and maintenance should be developed. If refrigeration is required for storing either HIV rapid test reagents or specimens, temperature checks with documentation and a maintenance plan must be in place.

4.0 Purchasing and Inventory

Availability of dependable and reliable test kits and supplies is essential. This requires a national plan for procurement and distribution, as well as careful management of supplies and reagents at the testing site.

4.1 Responsibilities at the national level

- Most countries use a tender process for procurement of reagents and supplies for all laboratories and testing sites managed through the Ministry of Health. It is important that supplies and reagents be carefully selected and that they are ordered in sufficient quantity to last until the next tender.
- Purchased kits must have an expiration date far enough into the future to allow for efficient use and to prevent waste. A policy of "first expired, first out" will also help to assure minimum waste.
- The Ministry of Health/national reference laboratory must have some means of assessing the quality of the kits, reagents, and supplies as they are received by the central purchasing body, to ensure that standards and specifications are met. It is recommended that each lot number should be checked by the national reference laboratory before distribution.
- A distribution plan that allows these reagents and supplies to reach all testing sites within the appropriate time frame and prior to expiration will be needed. The plan must take into account emergency or unexpected needs.

4.2 Responsibilities at the testing site

- An inventory record for kits and supplies should be maintained. Each site should determine re-order levels for each item in the inventory based on workload and usage. This will allow for ordering in a timely manner, so that the testing site always has the necessary reagents and supplies and no interruption in testing will occur. A method for calculation may be found in Appendix B.
- On receipt of new supplies and reagents, the inventory record should be updated and all of the new material stored under the appropriate environmental conditions. Appendix B provides examples of forms that can be used for inventory records.
- To avoid waste, sites should follow the concept of "first expired, first out". Kits that expire earliest must be used before kits with a longer expiration date.

5.0 Process Control

Process control refers to the activities and techniques that are carried out to ensure that the testing procedures are correctly performed, that the environment is suitable for reliable testing, and that the test kit works as expected to produce accurate and reliable results. Steps in the testing process follow the path of workflow, and begin with tasks done before testing, followed by tasks done during and after testing. The path of workflow is frequently described as those steps done before testing (*pre-analytic)*, those done while testing (*analytic)*, and the steps that follow testing (*post-analytic)*. When using HIV rapid test kits, there are a number of steps in these three parts of the path of workflow that are essential in order to assure accurate and reliable test results.

5.1 Evaluation of methods

Evaluation of HIV rapid test kits should be performed by the national reference laboratory or other appropriate body. In order to assure availability of kits for testing, it will be necessary for each country to have checked the validity and have approved several HIV rapid test kits for use in the country. The Ministry of Health should establish a national policy regarding appropriate tests to use and an approved algorithm for testing. A document developed by WHO/AFRO, CDC, and APHL provides guidelines for this evaluation; *Guidelines for Appropriate Evaluations of HIV Testing Technologies in Africa*.

5.2 Standard operating procedure

A standard operating procedure (SOP) must be developed that provides detailed instructions on all aspects of the testing, to include transport of specimens, storage and inventory information, test requesting, environmental requirements, specimen collection and management, test performance, quality control instructions, test interpretation, reporting and recording results, appropriate use of the testing algorithm, and any external quality assessment requirements. Each testing product will need its own SOP. A written SOP should be available at each testing site, and should always be followed when conducting tests. A chart showing a simplified version of the procedure steps (work instructions) is very useful, and should be provided at the point of testing performance. The test site must have written instructions on all policies and procedures that need to be followed when conducting tests, including such things as personnel training and certification requirements, competency checks, confidentiality policies and safety.

The work instructions would lay out steps to be taken during the work process, and would include any pre-analytic, analytic, and post-analytic information needed, such as the following steps:

Pre-analytic:
1. Check storage and room temperatures daily.
2. Check inventory and test kit lots as needed.

3. Receive requests for testing.
4. Set up test area.
5. Record all needed data, such as kit lot number, operator identity.

Analytic:

1. Follow biohazard safety precautions.
2. Perform external quality control (see discussion under 5.3.1) according to instructions.
3. Correctly identify person to be tested if pre-counseled by someone else.
4. Collect the specimen, including specimen for EQA if needed.
5. Perform the test as directed by the manufacturer.
6. Interpret the test results.

Post-analytic:

1. Re-check patient identifier and report results.
2. Clean up and dispose of biohazardous waste.
3. Package and transport EQA re-test specimens to referral laboratory, or appropriately store until next shipment to referral laboratory, if needed.

An example of a standard operating procedure for an HIV rapid test kit is shown in Appendix C.

5.3 Quality control

Quality control procedures are essential to ensure that the testing process has been carried out properly and that the kit reagents are performing as intended.

5.3.1 Definitions of quality control materials

For this document the terms internal quality control and external quality control will be used as follows:

Type of quality control	Description /Definition
Internal quality control	A control is either provided or built in to each test kit or device. It generally provides information about adequacy of quantity of specimen and whether the kit is working properly.
External quality control	Known positive and negative specimens or control materials. These may be purchased from a manufacturer or made locally (usually by a reference laboratory). These positive and negative specimens are used to evaluate the accuracy of the test and to check if the person doing the test performs it correctly.

The internal control in most cases is built into the testing device and will always be a part of the testing process. In some kits, these controls may be provided as a separate material, but will still be used with each test. Internal controls usually do not check the entire testing process.

External controls cannot be used with each test, but must be tested periodically in order to assure that the test kits are accurately detecting HIV antibodies.

5.3.2 Frequency of running external quality controls

The Ministry of Health should establish policy for how and when external quality control material should be used. This information must be provided and described in the standard operating procedure.

Frequency of use of quality control material is dependent on several factors. The condition of the kits must be evaluated over time. In areas where environmental conditions are sometimes extreme, difficult to control, and where transportation can be challenging, it will be important to check kits fairly often. Workgroup discussants for this document have made recommendations based on experience with local problems.

When running controls for HIV rapid testing, it is important to use both a negative and a positive control. Whenever possible, a weakly reactive positive control should be included that has been validated to yield weakly reactive results on all HIV rapid test kits used.

Recommendations

Run a negative and a positive control (and weakly positive when possible) at the following times:

1. At least once weekly, preferably at the beginning of the week.

2. When a new operator (a trained staff member who has not been doing testing for awhile, or a newly trained operator) is performing testing.

3. When beginning use of HIV rapid test kits with a new lot number.

4. Whenever a new shipment of test kits is received.

5. If rapid test kits are exposed to environmental conditions that fall outside the range needed for stability as defined by the manufacturer.

5.3.3 Recording and monitoring quality control results

The national policy should provide guidance on how to record and monitor quality control results. Standardized methods of record keeping will make it easier to compile data, monitor performance of the quality control material, and monitor test performance. Information provided to the testing sites should include:

- A standard worksheet that includes spaces for recording of QC results.
- A separate register for QC results, to which information will be transferred from the worksheet. This allows for quick review of QC data.
- A flow chart for corrective action, showing steps to follow when the QC results do not read as expected. This should include information on how to refer problems back to the reference laboratory.

All quality control data should be regularly reviewed by the quality officer with responsibility for the testing site.

Examples of forms for recording QC data can be found in Appendix D.

5.3.4 Sources of quality control materials

Quality control materials are available commercially, sometimes from the manufacturer of the HIV rapid test kit. They can also be prepared within the country, usually by the national reference laboratory. In some countries, the capability and capacity for preparing QC materials may be present in regional or provincial laboratories. These steps are needed for reliable materials to be used in testing sites:

- Materials for QC use should be prepared using a standard protocol. Two options for preparation of QC material are presented in Appendix E. These include 1) aliquoting and storing known positive or negative specimens, and 2) preparation of weakly positive controls. In the latter case, the weakly positive controls should be evaluated with all nationally approved HIV test kits.
- The reference laboratory should provide information on expiry dates for the material.
- The reference laboratory should monitor the performance of the material by reviewing data from the testing sites.
- It is recommended that QC materials be distributed with the test kits, assuring that each site has sufficient QC material for use with the kits.

5.4 External quality assessment

Through external quality assessment (EQA), the performance of a testing site can be evaluated from outside the laboratory or testing site. Methods for EQA include traditional proficiency testing, re-testing of specimens, and careful on-site monitoring using a checklist and knowledgeable assessors.

5.4.1 Re-testing of specimens

With this EQA technique, serum or dried blood spots are collected from the client at the time of rapid testing. The serum or the dried blood spots are tested using EIA, at a reference laboratory, and the results of this test, or "re-test," are compared with that obtained from the final HIV rapid test result. A common model in use is the re-testing of 5% - 10% of rapid test specimens, randomly selected.

The ability to perform EIA on dried blood spots (DBS) has made it easier to collect specimens for re-testing in areas where personnel

who can perform venipuncture are not available, or where reliable transport of serum specimens is not available. However, the dried blood spots must be carefully collected, dried, and properly stored in order to produce reliable results. The EIA testing performed on these specimens requires an elution step prior to specimen analysis, thus requiring additional time and introducing a new source of error.

Re-testing of specimens has limitations. In many countries there is lack of capacity at the national reference laboratory for re-testing the large number of samples and for conducting the needed analysis of data. Long delays in completing the re-testing results in delayed identification of problems. Appendix F outlines the operational issues that need to be considered if a re-testing program is to be implemented. Finally, statistical analysis reveals that for low-volume sites, a very large percentage of samples would have to be re-tested in order to detect errors. The following table summarizes the statistical information:

(Note: The number of specimens tested is independent of time.)

Re-test size (and %) needed to provide 95% confidence of detecting at least one discrepant result, when the underlying error rate is 1%or 5%E

Volume (per site)	1%* error	5%* error	Re-testing feasibility
Very low 50 specimens	Re-test 48 specimens (96%)	Re-test 31 specimens (62%)	No
Low 500 specimens	Re-test 225 specimens (45%)	Re-test 56 specimens (11%)	Possible
High 5000 specimens	Re-test 290 specimens (5.8%)	Re-test 59 specimens (1.2%)	Yes

* 95% confidence

A full discussion of the statistical models is presented in Appendix G.

Note these statistical models show analysis of site-specific data. Aggregate data could be obtained by combining data from several or many sites, and could have some usefulness in looking at the overall program. However, aggregate data is of no value for evaluating site error and for taking corrective action.

Recommendations

- If on-going re-testing is performed, it must be based upon statistical considerations and a recognition that it will only be feasible in high-volume test sites
- The outcome of re-testing must be analyzed for effective and timely feedback
  - in order to determine cost-effectiveness
  - to determine if corrective action can be taken if problems are identified
- If errors are not found as a result of re-testing, established sites should consider discontinuing re-testing.

5.4.2 On-site monitoring

EQA can be accomplished by a careful on-site observation of the testing processes and procedures, carried out by a knowledgeable person or team. A checklist that allows for assessment of all parts of the quality system is an important tool for such an on-site visit. Appendix H provides information on implementing and conducting on-site monitoring, including a sample checklist.

Recommendations

- On-site monitoring be given major emphasis in the EQA plan. In low-volume sites this may be the only EQA tool that is used.
- On-site monitoring should include all aspects of the quality system, including personnel competency and training, equipment policies, inventory control, quality control practices, records and documents, and facilities and safety.
- If other testing is performed at an HIV rapid testing site, an integrated approach to on-site visits should be taken to assess all aspects of testing practices
- The site visit should include observation of testing with specimens of known reactivity (proficiency panels).
- When possible, direct observation of interaction with a client is useful. Other means of assessing performance of testing personnel could include exit interviews with clients and use of "mystery clients" (persons with known serostatus who present anonymously).
- A standard checklist must be used for all visits.
- The on-site assessment should occur at least twice yearly in established sites, and at least quarterly for new sites or sites with new personnel. Frequency should be based on initial finds and need for corrective action.
- The on-site visits should be instructional and provide a mentoring experience. The experience should not be punitive.

A plan must be established for corrective action related to findings during the on-site visit. All problems should be discussed immediately with on-site staff, and any needed follow-up activities including training should be undertaken in a timely manner.

5.4.3 Proficiency testing

Traditional proficiency testing is organized and conducted by a reference laboratory or center. At regular intervals, a panel of specimens with known reactivity is sent to all participants who test the specimens and return results to the reference laboratory. The data is analyzed and information is provided back to the participant testing sites.

Proficiency testing for HIV rapid tests has some limitations. The panel of specimens sent to the testing site will not necessarily be tested by all staff, so it is not a good measure of individual performance. The sample size is small, so the ability to detect errors is impaired. Also, preparing and distributing specimens for proficiency testing may be burdensome for national reference laboratories. Proficiency testing is provided in some locations, and when available it is a useful tool in combination with on-site monitoring.

6.0 Documents and Records

Standardized documents and records should be developed at the national level in order to assure conformity to national standards and for ease in collecting national data. Documents and records must be maintained in such a way that they are always up-to-date and accurate, readily accessible by laboratory staff, and protected from damage and deterioration. Retention times for documents and records should be established. Policies should be developed to ensure confidentiality when appropriate.

6.1 Documents

6.1.1 Definition

Documents are the written policies, process descriptions, procedures, and any blank forms used in the testing process. Documents developed within the quality system include the written standard operating procedures, safety policies, personnel policies, and all standard blank forms, such as reporting forms.

External documents will also be used and can provide important information. Examples of external documents used in an HIV rapid testing site could include information from the kit manufacturer, references from journals, and any service manuals, such as for centrifuges or refrigerators.

6.1.2 Management of documents

Documents should be consistent with national policy, to assure uniformity and adequacy of data. All documents need to be managed with a tracking system, to assure that all testing sites have current information on hand, and that outdated documents are archived and ultimately discarded to avoid confusion at the worksite.

6.2 Records

6.2.1 Definition

Records result from carrying out processes and procedures within the testing process. Examples include worksheets, test result reports, labels, temperature and maintenance charts, quality control results and charts, EQA activities with results and corrective action, and inventory lists. Records are everything used to capture information, activities, or results when performing a procedure. They may be kept on paper, or electronically using a computer system. Records allow for the continuous monitoring of the quality system.

6.2.2 Management of records

Records for HIV testing sites should be standardized and distributed from the national level. Worksheets should include at a minimum: space for the date and time, client identifiers, name of the person performing the test, name and lot number of the kit used, and quality control results. A separate quality control chart should be maintained to allow for analysis and quick review of quality control results. Personnel records on training, competency evaluation, and work injury should be kept. All adverse occurrences, including any corrective action taken, should be recorded.

Appendix I provides examples of records useful in an HIV rapid testing site.

7.0 Information Management

Records may be kept either manually or using computer technology. When computer systems are available, laboratories are afforded many useful tools for managing client data as well as quality control and external quality assessment information. For example, a system for tracking specimens (either serum or DBS) collected for EQA purposes would make it much easier to manage this aspect of the quality system function. If there is country-wide networking, the ability to correlate an individual's clinical data and laboratory results country-wide will be valuable.

When computerized information management systems are available:

- Processes to ensure accuracy and reliability of data, and to protect data from damage and loss, must be put in place.
- Privacy and confidentiality of data must be strictly observed.
- Staff will need training to develop competency in use of computer tools, including use of the specific laboratory system, as well as in word processing, use of spreadsheets, and databases.

8.0 Occurrence Management

Errors and problems occur in the most carefully conducted and monitored testing environments. The purpose of a quality system is to reduce and minimize errors in the total testing process. In order to meet this goal, each testing site should have a method to detect and resolve problems. It is important to understand root causes and to take corrective action.

The following steps are important when adverse incidents, errors, and problems occur:

- Investigate the error or problem to determine cause.
- Take action to address the cause of the problem. Corrective actions may result in changes in policy or procedures to help ensure that the error will not re-occur.
- Communicate appropriately with all those affected by the error or problem, for example, nursing staff, physician, and/or client.
- Keep a record of all circumstances related to the error or problem. Also keep a record of corrective action taken and any communications with affected persons. This information is useful for those monitoring the testing, for any internal audits, and for use if further inquiries from patients or physicians occur.
- The Quality Officer has the responsibility to ensure that this process is followed, including all appropriate corrective actions taken.

9.0 Assessment

The key to a successful quality system is continuous improvement, and an essential component for this process is assessment. Formal assessments may be external, performed by persons outside the laboratory or testing site, or they may be conducted by staff at the site and be internally managed.

External assessment of testing sites in this model will be conducted as on-site visits, and this has been discussed in section 5.42 of this document.

The regular performance of internal quality assessments / audits can yield much important information about how well the laboratory or testing site is following its quality policies and procedures, and can help to identify problem areas. Information on the internal audit process is widely available, and the International Organization for Standardization (ISO) describes an internal audit process that is useful in laboratories. Smaller testing sites could use a more informal process.

10.0 Process Improvement

Process improvement is part of the continuous effort to look for problems and take actions to improve processes. Process improvement is the action of revising a process based on information gathered. As used in this model, process improvement involves identifying an area to study, then collecting information, evaluating the information, and taking corrective action based on the findings. For example, a testing site might decide to study its turn-around time; this would require collecting data for a period of time, analyzing the data, evaluating whether the turn-around time is sufficiently short, and if not, implementing some steps to shorten the time.

All these efforts should be the responsibility of the Quality Officer, who should manage all processes related to assessment and process improvement, and who should communicate results of all projects to both the site staff and to appropriate higher level management.

11.0 Service and Satisfaction

For diagnostic testing, customers include both providers who order the tests and patients/clients who will eventually receive care based on the results. There should be methods to evaluate how well the testing site is serving the needs of these customers. Questions to ask: 1) are customers receiving quality service, and 2) are program needs being met?

Policies and procedures should be established for responding to all suggestions and complaints from customers.

12.0 Facilities and Safety

12.1 Facilities

Each site where HIV rapid testing is performed must have physical space that is appropriate for the testing. This should include:

- an adequate working surface that can be easily cleaned and maintained
- assurance of an environmental temperature that does not exceed that required by the testing kit,
- refrigeration if needed,
- facilities for hand washing and cleaning.

12.2 Safety

Sites must have available, and personnel must follow, procedures to safely handle biohazardous material. This includes:

- Instructions on use of gloves, closed footwear, hand washing, handling and disposing of sharps, and spill containment and disinfection must be provided.
- Basic safety procedures should be clearly posted or visibly available in the laboratory.
- General policies such as "no eating, drinking, or smoking," "no unauthorized persons in the testing area," must be enforced.
- Procedures for safe disposal of all specimens and materials used in testing must be available and must be observed at each site. This is essential for protecting those performing the tests as well as others who might be exposed to discarded materials. All specimens and materials must be handled as if they are capable of transmitting an infectious disease.
- A procedure must be developed for workers to follow if there is accidental exposure of staff to biohazardous material. This procedure, as well as a list of persons to contact in an emergency, must be readily accessible to all staff in the facility. It is recommended that all persons performing HIV rapid tests should know their serostatus.

Full safety requirements for testing HIV specimens are very detailed. Any site performing testing should have available a complete set of guidelines used in the country. Useful references include: ISO, WHO Biosafety guidelines, CDC Biosafety Guidelines.

Useful References

CLSI HS1-A2 “A Quality Management System Model for Health Care”

CLSI GP26-A3 “Application of a Quality Management System Model for Laboratory Services”

ISO 19011:2002 - Guidelines for Quality and / or Environmental Management System Auditing

Guidelines for Appropriate Evaluations of HIV Testing Technologies in Africa

Appendix A - CDC/WHO Training Program for HIV Rapid Testing

Implementation Guide:

Establish national policies that will address:

- who will be trained
- who will serve as trainers
- scheduling of training programs to coincide with national program implementation
- budget for supplies, kits, printing of training materials, participant perdiems, etc.

Training Curriculum:

	Module	Learning Objectives	Content Outline
1	Overview of HIV Infection Duration: 30 min	• Describe the difference between HIV infection and AIDS • Discuss the HIV epidemics globally, regionally, and locally in terms of number of people affected • Define the terms antibody and antigen • Explain how "window period" may affect HIV testing results • Describe the progression of HIV infection	• What is HIV? • What is AIDS? • The HIV pandemic • Window period • Stages of HIV infection
2	Integration of HIV Rapid Testing in HIV Prevention and Treatment Programs Duration: 30 min	• Recognize the need for HIV testing and counseling (T&C) in HIV prevention programs • Describe the role of HIV rapid testing in supporting prevention and counseling programs • State the advantages of using HIV rapid tests in specific settings (e.g., VCT and PMTCT programs) • Describe the programs/ settings where HIV rapid tests are used in your country	• Need for expanding access to HIV testing • Testing and counseling as an integral part of HIV prevention, care and support services • Client counseling and HIV rapid testing are a formidable combination in any HIV prevention strategy

	Module	Learning Objectives	Content Outline
3	Overview of HIV Testing Technologies Duration: 60 min	• Describe key issues around expansion of HIV testing • Discuss the process for developing a national testing strategy • Discuss the spectrum of testing technologies for HIV • Describe WHO/UNAIDS Testing Strategies • Explain HIV Testing Algorithms	• Implementing national testing strategy • Challenges with HIV testing • Spectrum of HIV diagnostic tests: EIA, HIV Rapid Tests, Western Blot, Viral Load, p24 Antigen, CD4 • WHO testing strategies • HIV testing algorithms
4	HIV Testing Strategies and Algorithms Duration: 1 hour	• Explain the benefits of HIV rapid tests • Accurately recognize individual test result as positive, negative, or invalid • Describe the approved testing algorithm (process) • Interpret HIV status using the testing algorithm (process)	• Benefits and challenges of HIV rapid testing • Three formats of HIV rapid tests • Reading individual test results • Testing algorithm • Interpreting HIV status
5	Assuring the Quality of HIV Rapid Testing Duration: 1 hour 15 min	• Explain the systems approach to lab quality and its benefits • Identify the essential elements of a lab quality system and how they apply to HIV rapid testing • Recognize key factors that may compromise the quality of HIV rapid testing • Describe your responsibilities in preventing and detecting errors before, during, and after testing	• The approach we take to achieve quality • Essential elements of a lab quality system • Quality assurance procedures at the HIV rapid testing site • How you can contribute to quality before, during, and after testing

	Module	Learning Objectives	Content Outline

6	Safety at the HIV Rapid Testing Site
	
	Duration: 45 min
		• Adhere to personal health and safety practices
		• Maintain a clean and organized workspace
		• Disinfect and dispose of infectious materials
		• Take appropriate actions following accidental exposure to potentially infectious specimen
		• Follow written safety procedures and keep proper safety records
			• General safety practices
			  • Work habits (personal, work space, material)
			  • Proper disposal of sharps and waste
			  • Disinfection of work areas
			• Safety documentation

7	Preparation for Testing – Supplies and Kits Duration: 50 min	• List and identify all the supplies required for HIV rapid testing • List and identify all the components of test kits for HIV rapid testing	• Supplies & materials • Test kits
8	Blood Collection: Finger prick Duration: 2-2.5 hrs	• Explain the preparation tasks required for rapid tests • Put a client at ease while collecting blood • Collect blood from a finger prick accurately and confidently	• Preparation for testing • Educating your client • Performing finger prick
9	Performing HIV Rapid Tests: Demonstration and Practice Duration: 5.5 hrs	• Perform 3 HIV rapid tests according to SOP • Perform multiple tests simultaneously • Accurately interpret individual test results • Accurately determine HIV status	• Overview of testing procedures • Workstation setup • Demonstration • Practice session with known specimens • Practice session with blinded specimens

	Module	Learning Objectives	Content Outline
10	Inventory: Managing Stocks at the HIV Rapid Testing Site Duration: 1 hour	• Maintain proper records • Maintain proper level of consumables • Use first-expiry-first-out concept when managing stocks • Inspect delivery of supplies before acceptance • Identify lot numbers and expiry dates • Keep kits and supplies in proper storage	• What is stock management? • Record keeping • Re-order levels • Receipt of consumables • Storage of consumables
11	Use and Care of Equipment at the HIV Rapid Testing Site Duration: 55 min	• Specify your responsibilities related to equipment • Routinely monitor the temperatures of refrigerators and freezers • Confirm that auto pipettes deliver specified volumes • Properly use and maintain centrifuges	• Rationale for using properly maintained equipment • Your responsibilities for equipment • Use and care of equipment at the HIV rapid testing site o Refrigerator and freezer o Pipette o Centrifuge
12	Quality Control Duration: 45 min	• Differentiate between internal and external controls • Use external quality controls at designated frequencies • Analyze common problems associated with invalid test results	• What is Quality Control (QC)? • Benefits of QC in rapid testing • Internal versus external quality control • Troubleshooting invalid results • Quality control records

	Module	Learning Objectives	Content Outline
13	EQA: On-site Evaluation and Re-testing Duration: 1 hour 30 min	• Assess operations at test site to determine if quality requirements are met • Take corrective actions following External Quality Assessment (EQA) • Keep appropriate records related to EQA • Avoid common problems associated with EQA specimen management	• What is EQA and why is it important? • EQA Responsibilities • EQA Methods o Proficiency Testing o On-Site Evaluation o Re-testing • How to implement EQA
14	Blood Collection and Handling: DBS Duration: 1.5 hours	• Collect dried blood spots (DBS) • Package and store DBS in a way to maintain specimen integrity • Maintain DBS records • Distinguish between valid and invalid DBS	• Required supplies • How to collect and dry DBS • How to package and store DBS • Valid and invalid DBS • Hands-on practice
15	Documents and Records Duration: 30 minutes	• Tell the difference between a document and a record • Explain the rationale for maintaining documents and records • Provide examples of documents and records kept at a test site • Follow the procedures as prescribed in SOPs • Describe how to properly keep and maintain test site documents and records • Describe the types of information typically not found in a manufacturer's product insert	• What are documents and records? Documents • Why are they important? • What documents should you keep? • Why is it important to follow SOPs? • What is the proper way to keep and maintain documents? Records • Why are they important? • What records should you keep? • What is the proper way to keep and maintain records?

	Module	Learning Objectives	Content Outline
16	Professional Ethics Duration: 45 minutes	• Describe ethical issues related to HIV rapid testing • Explain the importance of professional ethics • Apply ethical conduct to HIV rapid testing • Take appropriate actions to maintain client confidentiality	• What is ethics? • Why is ethics important? • Who is responsible for ethics? • How is ethics applied to HIV rapid testing? • Maintaining confidentiality • Code of conduct

Appendix B - Stock Card

Item Name: _____ Unit: _____

Manufacturer: _____

Kit Lot Number: _____ Expiration Date: _____

**Minimum Stock (Re-Order Level): _____

Date	Received From	Issued to	Quantity Received	Quantity Issued	*Balance	Lot #	Signature

**Minimum Stock = Maximum lead time in weeks times (x) maximum usage
Maximum lead time is 2 weeks
Maximum usage / 3 months = 3 kits
Minimum Stock = 2 x 3 = 6 kits

*Balance – Perform physical inventory count each month. If physical count does not equal balance, consider causes for discrepancies, e.g., theft, expired kits

Stock Book

Item Name	Qty (units) Requested	Date Requested	Qty Received	Date Received	Lot #	Expiry Date

Appendix C - **Example of a Standard Operating Procedure

**This SOP has been provided to illustrate the kinds of information that need to be captured and followed to ensure accurate performance and interpretation of test procedure. The site in which testing is performed may have specific requirements that will need to be incorporated in this SOP and other collective documents.

****

Test Procedure using Uni-Gold HIV Rapid Test Kit
Check the expiry date and use kits within expiry date

Intended Use
The Trinity Biotech Uni-Gold™ HIV test is a single reagent assay for the detection of antibodies to human immuno deficiency virus types 1 and 2 in serum, plasma or whole blood.

Principle of the Procedure
Synthetic peptides of diagnostic relevance representing the highly immunoreactive sections of the envelope proteins of HIV-1 and HIV-2, glycoprotein gp41, gp120 (HIV-1) and glycoprotein gp36 (HIV-2) respectively are immobilised at the test region of the nitrocellulose strip. The peptides are also linked to colloidal gold and impregnated below the test region of the device. A narrow band of the nitrocellulose membrane is also sensitised as a control region.

During testing one drop of serum, plasma or whole blood is applied to the sample port, followed by two drops of wash buffer and allowed to react. Antibodies to any immunoglobulin class, specific to the synthetic HIV-1 or HIV-2 peptides, will react with the colloidal gold linked antigens. The antibody peptide-colloidal gold complex moves chromatographically along the membrane through the test and control regions of the test device.

A positive reaction is visualized by a pink/red band in the test region of the device.
A negative reaction occurs in the absence of human immunoglobulin antibodies to HIV in the analyzed specimen. Consequently no visually detectable band develops in the test region of the device.

Excess conjugate forms a second pink/red band in the control region of the device. The appearance of this band indicates completion of the test as well as proper performance of the reagents in the device.

Kit contents:

- 20 Test Devices
  Each test device contains colloidal gold labelled with synthetic HIV peptides, synthetic HIV peptides as test zone and a control line.

- Wash Reagents (4ml)
  - Serum/Plasma Wash - Tris buffered wash containing detergent and preservative (0.1% sodium azide).
  - Whole Blood Wash - Saline wash containing preservative (0.1% sodium azide).

- 20 Disposable Pipettes

- Package Insert

Materials required but not provided with the kit:

- Timer or stopwatch
- Blood collection devices (i.e., lancets, capillary tubes/ test tubes)

Storage and Stability

The Uni-Gold™ HIV test devices can be stored at 2-27°C. No kit components should be used after the kit expiry date.

Precautions

- The Trinity Biotech Uni-Gold™ HIV Test is intended for in vitro use.
- Do not use the kit past the expiration date.
- Do not smoke, eat or drink in areas in which specimens are handled.
- Dispose of all specimens, used devices and pipettes as though they are capable of transmitting infection. The preferred methods of disposal are by autoclaving at 121°C for a minimum of 60 minutes or by incineration.
- All spills should be wiped thoroughly using a suitable disinfectant such as a sodium hypochlorite solution.
- Use a separate disposable pipette and device for each specimen tested.
- Sodium azide is a toxic compound and solutions containing same should be handled with care. If the wash solution is disposed through the drain it should be flushed thoroughly with water.
- Controls where supplied have been certified virus free. As with all screening assays, any results should be considered presumptive until confirmatory assays have been performed according to the testing algorithm.

Specimen Collection and Storage

Whole blood, serum or plasma may be used.

Whole Blood: If fingerstick whole blood is used, drops of blood produced should be taken up from the finger-tip by the pipette supplied and dropped from the pipette onto the device. Blood droplets **should not** be dropped directly from the fingertip onto the device as their size may vary. Whole blood specimens should be used within ten minutes of collection for optimum performance.
If a specimen has started to clot, do not remix before testing. In such instances, the clear serum should be pipetted off the clotted specimen and used for analysis. If an anticoagulant has been used in the blood sample, whole blood can be used directly on the device using the pipette supplied. If testing is not to be carried out immediately, samples should be stored at 2-8°C for up to three days, or preferably, the sample should be centrifuged and the plasma retained for future testing.

Serum or Plasma: Serum or plasma may be kept for seven days at 2-8°C. Samples should be frozen for longer storage. Avoid repeated freezing and thawing of samples.

Quality Control
Good Laboratory Practice necessitates the use of control specimens to ensure proper device performance at least once daily.

A built in procedural control on the test device indicates that the test is functioning correctly. A pink/red band should always appear at the control window.

Internal and External controls should be run daily prior to analyzing patient/client specimens. Results should be recorded on the QC log. Patient/Client results are reported only if QC results are acceptable.

Test Procedure

1. If any reagent /sample have been refrigerated remove and allow to stand for 20 minutes to reach room temperature.

2. Remove the required number of test devices from their protective wrappers.

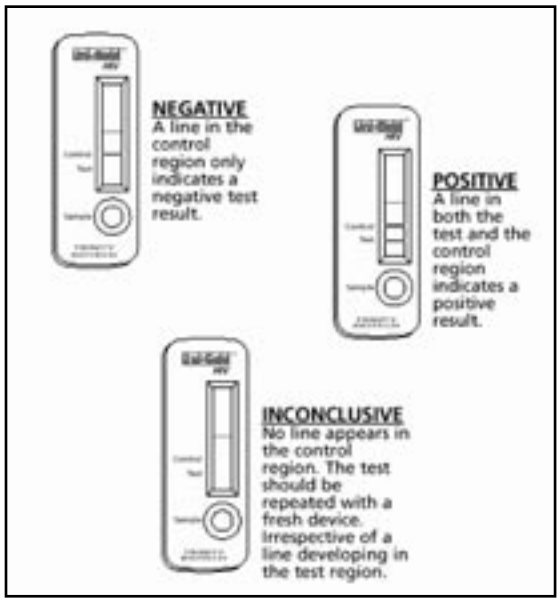
3. Label each test with the appropriate patient identification.

4. Using one of the disposable pipettes supplied, fill with sample (serum/plasma/ whole blood).

5. Holding the pipette over the sample port add 2 drops of sample (approx. 60µl) carefully.

6. Add 2 drops (approx. 60µl) of the appropriate wash reagent to sample port.

7. Allow 10 minutes for reaction to occur. The result should be read at the end of the 10 minute incubation time. Results are stable for at least 20 minutes after addition of sample to the device



Interpretation of Test Results

- One black line of any intensity only in the control window report as negative/ non-reactive.
- Two black lines of any intensity in both the patient and controlwindow report as positive or reactive.
- No black line in control window in control window report as invalid

Limitations

Uni-Gold™ HIV test procedure and interpretation of results must be followed closely when testing for the presence of HIV antibodies in serum, plasma or whole blood.

The Trinity Biotech Uni-Gold™ HIV test is intended for the testing of undiluted samples only. Samples should not be diluted before testing.

Immunosuppressed or Immunocompromised individuals infected with HIV-1 or HIV-2 may not produce antibodies to the virus.

Testing with any kit designed to detect antibodies may give negative results and would not be a reliable test method for such patients.

Infants may receive antibodies from an infected mother or they may not produce antibodies in response to an infection. Therefore, it is necessary to exercise great care in interpreting their results.

AIDS and ARC are clinical syndromes and their diagnosis can only be established clinically. Uni-Gold™ HIV test results alone cannot be used to diagnose AIDS. A negative result does not preclude the possibility of exposure to HIV or infection with HIV.

Appendix D - Quality Control Form

Daily Record of Quality Control Results

Kit: _____ Lot #:_____ Expiration Date: _____

Date	Negative Control Result	Neg Control Lot #	Acceptable? Y / N	Positive Control Result		Pos Control Lot #s		Acceptable? Y / N	Initials	Reviewed by & Date
				Low Pos	Pos	Low Pos	Pos			

Corrective Actions

Date	Action Taken	Initials	Reviewed by & Date

Appendix E - Protocol for Preparing HIV Positive Quality Control Materials

PURPOSE This procedure provides instructions for making a supply of HIV positive samples at a desired reactivity level to be used as a daily control, or as part of a proficiency testing panel.

EQUIPMENT AND MATERIALS:

Equipment

- Magnetic Stirrer, Non-Heated
- Single channel Pipettes (0.05 – 20 µl, 50-200 µl)
- Multi-channel Pipettes (0.5 – 20 µl and 50-300 µl)
- Vacuum Pump
- Tubing for Vacuum Pump
- Water bath or incubator
- Thermometer
- ELISA equipment, e.g., reader, washer

Materials

- Unit (Blood Type Group O) of HIV antibody positive serum
- Unit (Blood Type Group O) of HIV antibody negative serum
- Sterilizing Filters, .22 micron
- Cryogenic vials, polypropylene, 1.0 ml (for storage of aliquots)
- Cryovial storage boxes
- Brain Heart Infusion Broth (ready to use in tubes) or Brain Heart Infusion (BHI) powder and materials to prepare tubes of broth.
- Sterile screw cap tubes, 16x125 (for BHI broth, if needed)
- Non-sterile plastic tubes, polypropylene 12x75 (for serial dilutions)
- Glass Stir-rods
- Pipette tips
- Individually Wrapped Sterile Pipettes (1.0, 5.0, 10.0, 25ml)
- Discard or waste containers
- Disinfectant
- Autoclave bags
- Gloves and labcoats

HANDLING CONDITIONS:

1. Units of HIV serum should be stored at 2 -8°C.
2. Follow good laboratory safety practices when handling all samples
3. Properly dispose of contaminated waste according to established waste disposal procedures.

OVERVIEW OF PROCESS

1. Calculate sufficient volume required for one year supply of ready-to-use aliquots, and an additional bulk volume for freezing and storing for future use, e.g., 250ml of serum will yield 500 aliquots of 0.5 ml.
2. Obtain HIV positive and HIV negative sera. Consider the National Blood Transfusion Service as one potential source of sera.
3. Heat inactivate positive sera and negative sera
4. Filter and Sterilize positive and negative sera
5. Titrate HIV positive sera
6. Perform HIV ELISA test to select desired titered sample
7. Prepare bulk volume of selected titer
8. Validate results of bulk volume
9. Aliquot, label, and store
10. Perform homogeneity and stability testing
11. Maintain data logs and records

STEPWISE PROCEDURE:

Calculate volume required

1. Calculate the total volume of sample required before beginning production to ensure that sufficient materials / reagents are available. The volume required may depend on a number of factors:
    - How long the pooled serum is needed , e.g., 12 months
    - How often the HIV test is performed
    - The sample volume required by the test
    - The number of participating laboratories in your Proficiency Testing Program
    - Approximately 10% overage for determining homogeneity and stability testing

Obtain HIV positive and HIV negative sera

2. Obtain a unit of sterile HIV antibody positive and HIV antibody negative serum from Type O donors. *Note: One unit yields approximately 400 ml of serum*.
    - Both units should be negative for HBsAg and HIV antigen.
    - Both units should be non-haemolysed, non-laepemic, and free of particulate material.
    - The HIV positive unit should have a high HIV antibody titre (6 – 8X ELISA test cut-off).

Heat inactivate HIV positive and negative sera

3. Heat-inactivate the HIV antibody positive and negative units at 62°C for 20 minutes in a water bath.
    - If a water bath is not available, place the unit of serum inside a large glass container of water in an incubator set to 75°C for 20 minutes.
    - Place a thermometer in the water and monitor a rise in temperature to 62°C.
    - Place the unit of HIV antibody positive serum in the water.
    - Continue to monitor the temperature of the water and when it again reaches 62°C, time for 20 minutes.

Filter and Sterilize sera

4. Using sterile technique, filter the heat-inactivated serum through a .22 micromole size sterile filter into a sterile enclosed polypropylene container.

5. Using sterile pipettes and sterile technique inoculate 3 – 4 tubes of Brain Heart Infusion Broth with 100µl of the heat-inactivated, filtered serum and incubate at 37°C for 7 days.

6. Store the remainder of the HIV antibody positive heat-inactivated, filtered serum at 2 - 8°C.

7. Using sterile technique, filter the HIV antibody negative serum through a .22 micromole size sterile filter into a sterile enclosed container.

8. Using sterile pipettes and sterile technique, inoculate 3 – 4 tubes of Brain Heart Infusion Broth with 100 µl of the filtered HIV antibody negative serum and incubate at 37°C for 7 days.

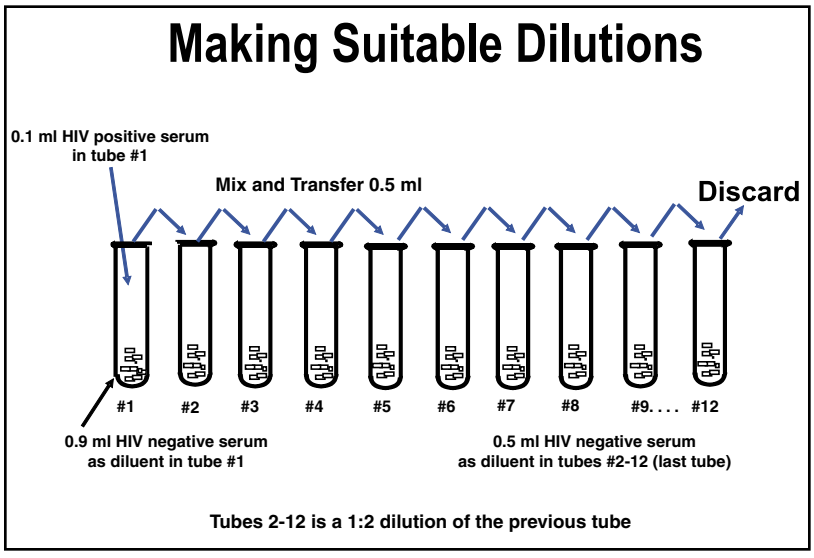
9. Store the remainder of the HIV antibody negative filtered serum at 2 - 8°C.

10. At the end of 7 days, check the broths for turbidity. If no turbidity exists in any tubes, begin titration of the HIV antibody positive serum.

Titrate HIV positive sera (Determine desired dilution)

11. A titration is conducted as follows:

    Make an initial 10-fold dilution of the HIV antibody positive serum by adding 0.1 ml of the HIV positive sera, and 0.9 ml of HIV negative serum to tube 1. Pipette 0.50 ml of HIV antibody negative serum into tubes 2-12. Make 2-fold dilutions in tubes 2-12 by mixing and transferring 0.50 ml of from tube 1 to tube 2. Continue mixing and transferring 0.50 ml through the last tube, ending with a dilution of 1:20,480.

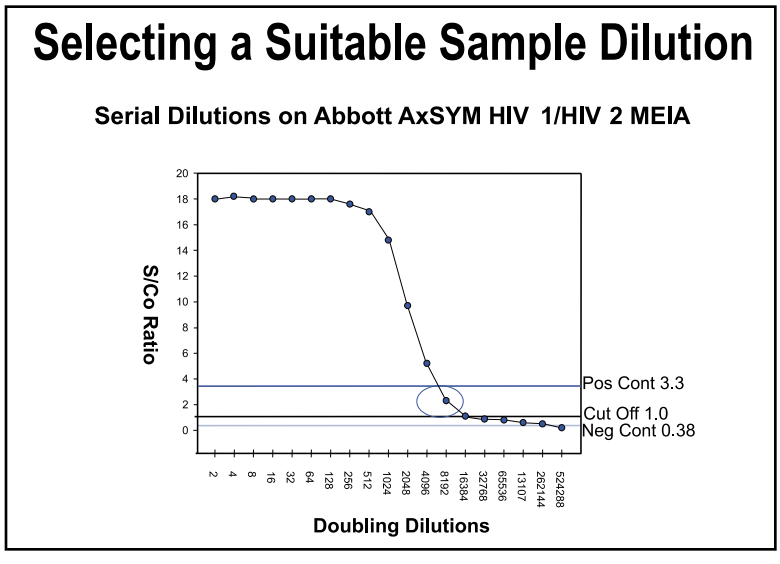


Perform HIV ELISA test to select desired titered sample

12. Perform a HIV1/2 ELISA following the manufacturer's instruction and your standard operating procedure. In accordance with the manufacturer's instructions, include kit positive and negative controls in the test run. Test each titrated sample, and the original sample from which dilutions were made, in triplicate.

13. Based on the Sample/Cut-off (S/Co) ratios calculated for the HIV antibody positive diluted samples, select dilutions that represent a high titered and a low titered sample. Generally, a low titered positive has an S/Co ratio of 2 – 3 and a high tittered positive has an S/Co ratio of 5 – 6.

14. Plot the S/Co ratio results against the sample dilutions. For example, a dilution of 1:40,960 will yield a cut-off value of approximately 3;



Prepare Bulk Volume

15. Choose the appropriate dilutions for high titered and low titered positive samples and determine the volume of sample suitable for your purpose. For example, if 500 ml was needed and the 1:10,240 dilution was selected as the high titered sample, add 50 µl of the bulk HIV antibody positive serum to 512 ml of the HIV antibody negative serum. Use a sterile container with a lid to contain the dilutions for the bulk samples.

16. Add a preservative such as Bronidox (0.5%), to the final diluted serum, e.g. 0.5mls to 500mls diluted control. Check the package insert of the assay for which the QC/PT sample is to be used, to ensure that the preservative is appropriate and will not interfere with the performance of the assay.

17. Place the diluted serum on a magnetic stirrer in a biohazard cabinet and mix for at least one hour to ensure a homogenous batch.

Validate results of pooled serum

18. Retest using the same HIV 1/2 ELISA assay to validate the results of the diluted batches of serum. Compare these results of the batch with the initial EIA results.

Aliquot, Label, and Store

19. If results of the ELISA test are acceptable, aliquot appropriate (small working) volumes, e.g. 0.5-1.0 ml from the HIV antibody high positive and the HIV antibody low positive batches into sterile internal threaded, polypropylene vials with silicon O-rings in the caps.

20. Store the vials and remaining bulk volume in well labeled containers at -80˚C until needed. Aliquots stored at 4˚C should be discarded after one week.

Perform homogeneity and stability testing

21. To ensue that the pooled serum has been well mixed and homogenous, randomly select approximately 10% of total aliquots. Test these samples, and compare results with target S/C ratio. Investigate the variability of the samples by calculating the CV for the selected samples. The batch of pooled serum is acceptable if the CV of the results is less than 15%.

22. To validate the stability of the level of reactivity of the pooled serum, place aliquots of the sample at -20ºC , 4˚C, and room temp (15-25˚C), Test the samples at 7, 14, 21, and 28 days. Review the results obtained at each temperature. The batch of pooled serum is considered stable if the S/Co ratio is falls within ± 2 standard deviations of the original results.

Appendix F - External Quality Assessment of HIV Rapid Tests: Operational Issues for Retesting

If retesting is used as a way to conduct EQA for HIV rapid testing, there are a number of aspects to be considered when determining and implementing national (or local) policy.

Collection of samples for retesting

Appendix F provides tables and guidance for selecting the appropriate sample size for retesting. The numbers shown in the tables can be applied to any time period, and countries will need to select the appropriate period, balancing the need for a reasonable retest sample size against how frequently performance needs to be evaluated. For example, selecting a time interval of a year may make it feasible for a reference laboratory to test the required number of specimens, but may seem unreasonably long for purposes of detecting potential performance problems. An interval of three months may, in smaller sites, result in a very large number of repeat specimens, but would provide a more frequent look at the performance of the site. In summary, the retest sample size required for statistical validity and the time period to be used for measurement must be determined based upon practicality and sustainability.

Whenever possible the samples collected for retesting should be randomly selected and distributed throughout the testing period. This will allow the retesting EQA to be more representative of the testing process.

In a site with multiple staff performing testing, it is logistically difficult to attempt to have a separate sample for each individual. However, if the retesting is to be representative of the performance of the entire site (therefore all persons performing testing), it will be important to assure that samples collected for retesting represent as widely as possible the entire staff.

Two methods are possible when collecting samples for the retesting:

1. Collection of dried blood spot (DBS). When using whole blood testing with a fingerprick, a specimen for DBS can be collected at the same time. A truly random sampling would involve collecting DBS on every client and then retrospectively selecting a random sample of all DBS specimens. However, this may be impractical and requires considerable resources (DBS filter paper, space for drying and storing DBS, etc.). Most sites will need to prospectively identify those clients who will have a DBS specimen collected, rather than collecting on all clients.

2. Venous blood collection. In some areas, a venous specimen is collected for testing with an HIV rapid test kit. In this instance, there can be retrospective selection of specimens for a random sample. When this method is used, all specimens may be aliquoted and stored, and then a random selection process used to determine which specimens to retest. Alternatively, the selection may be done first, and only the selected specimens aliquoted and stored.

Venous blood collection may also be carried out at sites that perform HIV rapid testing with a finger prick, but do not have available (or choose not to use) DBS procedures for the retesting. In this case, prospective identification of the clients to be tested is required.

Recording information and transporting retesting samples

It is important to maintain patient confidentiality in the retesting process. It is recommended that a laboratory register or specimen number be used when sending samples from the original testing site to the reference laboratory for retesting.

Distributing the retesting workload for the reference laboratory will help to avoid work overload and delays in returning results to the testing site. This will require a preplanned schedule so that retest specimens reach the reference laboratory at spaced intervals. Frequent transport, a continuous retesting process, and prompt feedback of results will help to assure timely monitoring of performance and promopt alerts when problems are detected.

Finally, care must be taken to assure that specimens are transported in such a way that the reference or retesting laboratory receives them in good condition.

Retesting of samples

Generally, retesting of samples will be carried out by one or several reference laboratories within the country. The reference laboratory, or laboratories, should assure its quality of testing by appropriate validation of the EIA technology employed, both for DBS and for venous blood. In addition, all laboratories performing this reference testing should participate in EQA for HIV testing.

Following retesting, an approach is needed for investigating discrepancies between the original rapid testing and the re-test by EIA. Errors or differences in results may occur for a variety of reasons. Operator error in test performance is one cause of discrepancy, requiring additional quality assurance and training at the site. A common source of error is a transcription mistake at some point in the process. Errors may be produced if the samples for re-testing are improperly stored and/or transported. Very slight differences are also observed between rapid tests and EIA tests, with neither being more accurate than the other. All discrepancies should require investigation. A policy must be developed for how to resolve discrepancies, and an acceptable level of discrepancy must be determined.

Reporting and corrective action

The results of the retesting should be reported to the original testing site and to the designated quality officer where applicable. The testing site and its quality officer should evaluate all results received from retesting, and take appropriate corrective action when performance goals are not met.

In most countries, the Ministry of Health will also be a recipient of information on the results of retesting. The results should be collected systematically and used to evaluate testing performance on a national basis, as well as to initiate appropriate corrective action when needed.

Appendix G - External Quality Assessment of HIV Rapid Tests: Statistical Models for Re-testing

Re-testing of samples has been used for monitoring HIV rapid testing in lieu of conventional EQA/PT, which often is not available for laboratories and testing sites. This document looks at the statistics that apply to this re-testing, and provides information that will be useful in determining appropriate models for EQA re-testing.

Current Situation

Currently there are a variety of re-testing EQA schemes in place in various countries. Examples include:

1. Re-testing 5% of all samples and the 1st 40 samples tested by each technician that runs tests.
2. Re-resting 10% of all samples.
3. Re-testing all positives and varying percentage of the negatives.

Other considerations are as follows:

- In all cases retesting is with a confirmatory EIA method.
- Current re-testing schemes do not account for numbers of samples tested at each site. This volume can be highly variable; from 50 to 1000 tests performed per month, or 500 to 10,000 per year.
- Current national rates of HIV prevalence vary from low to very high. In a particular site, the rate of positivity may be much higher (or lower) than the national prevalence. Rate of positivity in testing sites is highly variable.
- Data on agreement between rapid HIV methods is becoming more available as national HIV rapid test evaluations are taking place. Algorithms used require confirmation of all positive tests by at least one different method and many negatives also have to be negative on two kits (in some countries initial negatives are not confirmed). In cases where the first two results do not agree, a third kit is used and this is considered confirmatory (several variations exist, but this is sufficient for the purposes of this discussion).
- The sample for re-testing must be obtained at the time of initial testing.
- Errors (disagreement with EIA) can occur for a variety of reasons, including
    - use of outdated kits
    - improper storage of kits
    - lack of technical competence
    - clerical error
    - insensitivity of the kit
- We do not have (for use in this document) information on current agreement rates for re-tested samples. It would be very useful to know the distribution by agreement on positive and negative samples, the agreement rates at different sites, or the agreement rate for new technicians, in addition to the overall agreement rates that are being found.

Objectives:

The model needs to consider the following variables:

- True error rate (unknown)
- Positivity rate
- Population size (number of cases per study period)
- Probability of detection of errors
- Number of re-tested cases, or the proportion of cases to re-test
- Decision rule: act on a single discrepancy or multiple discrepancies?

The recommended re-testing scheme should achieve the following objectives:

- Provide a stated level of confidence that low error rates will be detected.
- Be independent of positivity rate.
- Accommodate different numbers of tests performed in the time period (50-10000).
- Assume that even a single disagreement will lead to investigation.
- Assume re-testing (EIA) is performed without error.

Model Assumptions:

The model presented below could be applied equally to samples that are positive and negative on initial testing, or could be used on all tests no matter what the initial result is. The recommended number of samples could be drawn independently from patients that are initially diagnosed as positive and from those that are initially diagnosed as negative, or could be chosen randomly from all patients. In some instances it would be difficult to base re-testing on the initial result, so the easiest re-testing scheme would come from a random sample of all patients.

The model assumes that any discrepant result is an action signal. That is, whatever the actions are for "suspect" results from test sites, these actions would be initiated on the discovery of a single discrepant result, no matter what the sample size or positivity rate.

The suggested model eliminates the need to consider rate of HIV positivity, but there remain four important dimensions to the recommendations:

- Population size
- Sample size
- True error rate
- Confidence level

Positivity rate would not affect the estimates in the following tables and figures, but it would affect the "power" of the procedure (ability to detect errors) if there are different probabilities for false negatives and false positives. The positivity rate and the clinical impact of false positives and false negatives could lead to different re-testing procedures for positives and negatives.

Model:

The Hypergeometric distribution can be used to predict the probabilities of detection for any given sample size. For this model,

- Eight different sizes of tests were checked (this could be numbers of negatives or positives, or both): 50, 100, 200, 500, 1000, 3000, 5000, and 10000.
- Three different possible error rates were investigated: 1%, 3%, and 5%.
- Three levels of confidence were checked: 90%, 95%, and 99%.
- Three different re-testing rates were checked: 5%, 10%, and 20%.
- For small samples and low error rates it was necessary to assume at least 1 error; for example, 50 samples with 1% error rate was assumed to have 1 error, and a 5% sample produces 3 cases. This can distort the percentages in the tables.

The model was applied in three different ways to answer three questions:

1) For given numbers of cases and given error rates, what sample size is needed to provide a stated confidence of having at least 1 discrepant result? The numbers can then be converted to percentages of the number of cases (**Table 1a-c**)
2) For given numbers of cases, given re-testing rates and given error rates, what is the probability of observing at least one discrepant result? This can also be called the Power of the re-testing and decision rule.(**Table 2a-c**)
3) For given numbers of cases and given re-testing rates, what is the lowest error rate that can be detected with a stated confidence? This is the upper limit of the Confidence Interval for the error rate (the lower limit is zero) (**Table 3a-c**)

TABLE 1a: Re-test size (and %) needed to provide 90% confidence of detecting at least one discrepant result, when the underlying error rate is 1%, 3%, or 5%E

Number	1%E	3%E	5%E
50	45 (90%)	34 (68%)	27 (54%)
100	90 (90%)	54 (54%)	37 (37%)
200	137 (64%)	63 (32%)	41 (21%)
500	184 (37%)	71 (14%)	43 (8.6%)
1000	205 (21%)	73 (7.3%)	44 (4.4%)
3000	221 (7.4%)	75 (2.5%)	45 (1.5%)
5000	224 (4.5%)	76 (1.5%)	46 (.92%)
10000	227 (2.3%)	77 (0.77%)	47 (0.47%)

Example: If there are approximately 1000 cases in the time period, and 90% confidence is acceptable for detecting 5% errors, then a 4.4% re-test will suffice (44 samples)

TABLE 1b: Re-test size (and %) needed to provide 95% confidence of detecting at least one discrepant result, when the underlying error rate is 1%, 3%, or 5%E

Number	1%E	3%E	5%E
50	48 (96%)	39 (78%)	31 (62%)
100	95 (95%)	63 (63%)	45 (45%)
200	155 (78%)	78 (39%)	51 (26%)
500	225 (45%)	90 (18%)	56 (11%)
1000	258 (26%)	94 (9.4%)	57 (5.7%)
3000	284 (9.5%)	97 (3.2%)	58 (1.9%)
5000	290 (5.8%)	98 (2.0%)	59 (1.2%)
10000	294 (2.9%)	99 (1.0%)	60 (0.60%)

Example: If there are 200 cases and the objective is to have 95% confidence in detecting an error rate of 3% or more, then the number of re-tested cases would be 78, or 39% of all cases.

TABLE 1c: Re-test size (and %) needed to provide 99%confidence of detecting at least one discrepant result, when the underlying error rate is 1%, 3%, or 5%E

Number	1%E	3%E	5%E
50	50 (100%)	45 (90%)	39 (78%)
100	99 (99%)	78 (78%)	59 (59%)
200	180 (90%)	106 (53%)	73 (37%)
500	300 (60%)	131 (26%)	83 (17%)
1000	368 (37%)	141 (14%)	86 (8.6%)
3000	425 (14%)	148 (4.9%)	88 (2.9%)
5000	438 (8.8%)	149 (3.0%)	89 (1.8%)
10000	448 (4.5%)	150 (1.5%)	90 (0.90%)

Example: If it is desired to have 99% confidence that an error rate of 1% or more can be detected, in a situation with 50 (or fewer) cases, then 100% of results need to be re-tested.

TABLE 2a: Probability of obtaining at least one discrepant result with re-testing rates of 5% with error rates of 1%, 3%, and 5%E

Number	1%E	3%E	5%E
50	.06	.12	.17
100	.05	.14	.23
200	.10	.27	.41
500	.23	.54	.73
1000	.40	.79	.93
3000	.79	.991	1.0
5000	.92	1.0	1.0
10000	.994	1.0	1.0

Example: If there are 5000 cases and a 1% error rate (50 errors), then if 5% of cases are re-tested (250 cases) there is a .92 probability of selecting at least one of the errors (power). If there are 1000 cases (50 re-test cases), there is a .93 chance of detecting 5% errors.

TABLE 2b: Probability of obtaining at least one discrepant result with re-testing rates of 10% with error rates of 1%, 3%, and 5%E

Number	1%E	3%E	5%E
50	.10	.19	.28
100	.10	.27	.42
200	.19	.47	.66
500	.41	.80	.93
1000	.65	.96	.996
3000	.96	1.0	1.0
5000	.995	1.0	1.0
10000	1.0	1.0	1.0

Example: If there are 1000 cases, an error rate of 3% and a re-test rate of 10%, then there is a probability of .96 that at least one result will be discrepant.

TABLE 2c: Probability of obtaining at least one discrepant result with re-testing rates of 20% with error rates of 1%, 3%, and 5%E

Number	1%E	3%E	5%E
50	.20	.36	.50
100	.20	.49	.68
200	.36	.74	.90
500	.67	.97	.997
1000	.89	.999	1.0
3000	.999	1.0	1.0
5000	1.0	1.0	1.0
10000	1.0	1.0	1.0

Example: If there are 500 cases and a 20% re-sampling (100 re-sample cases), then there is a .67 probability of having at least one failure in the sample when the error rate is 1%.

TABLE 3a: Lowest error rate that can be detected with 90% confidence with the stated re-testing rates (%ReT) and given number of cases

Number	5%ReT	10%ReT	20%ReT
50	54%	36%	20%
100	37%	20%	10%
200	21%	11%	5.5%
500	8.6%	4.4%	2.2%
1000	4.4%	2.2%	1.1%
3000	1.5%	0.73%	<.5%
5000	0.90%	<.5%	<.5%
10000	<.5%	<.5%	<.5%

Example: With 3000 cases, it would require 10% re-testing (300 re-test cases) to detect a 1% error rate (table entry 0.73%), with 90% confidence. If no errors are found, the 90% confidence interval for the error rate is (0 to 0.73)

TABLE 3b: Lowest error rate that can be detected with 95% confidence with the stated re-testing rates (%ReT) and given number of cases

Number	5%ReT	10%ReT	20%ReT
50	62%	44%	24%
100	45%	25%	13%
200	26%	14%	6.5%
500	11%	5.6%	2.8%
1000	5.7%	2.9%	1.4%
3000	1.9%	0.97%	<.5%
5000	1.2%	0.58%	<.5%
10000	0.58%	<.5%	<.5%

Example: If 10% of 50 slides are re-tested (5 re-test cases), and no errors are found in the sample, then the 95% confidence interval for the error rate is: (0 to .44).

TABLE 3c: Lowest error rate that can be detected with 99% confidence with the stated re-testing rates (%ReT) and given number of cases

Number	5%ReT	10%ReT	20%ReT
50	90%	78%	34%
100	59%	36%	19%
200	37%	20%	10%
500	17%	8.4%	4.2%
1000	8.6%	4.3%	2.1%
3000	3.0%	1.4%	0.70%
5000	1.8%	0.88%	<.5%
10000	0.90%	<.5%	<.5%

Example: With a 20% re-test rate and 200 slides in the population (40 cases selected), the error rate has to be at least 10% (20 errors), if we are to have 99% chance of including at least one of the errors in the re-tested cases. If no errors are found in the re-test cases, the 99% confidence interval for error is (0 to .10).

Observations:

1. The estimates above can be applied to any subset of testing situations, or any combined group, including all tests for a year or all tests with a specific kit. That is, the numbers in Tables 1-3 can be used to estimate sample sizes needed to assure levels of performance of specific testing centers, technicians, or kits.

2. Traditional proficiency testing (PT) and re-testing are both useful EQA methods; they serve similar purposes in some ways, but differ in others in their ability to detect errors and in the services they provide. Both systems:
   - Monitor performance to detect systematic errors.
   - Motivate laboratory and technician to pay attention to quality.
   - Assure responsible oversight.

   They differ in that:
   - Re-testing provides more samples than PT and therefore is more sensitive to errors. Since error rates of concern are expected to be <5%, large samples are required to detect errors.
   - PT provides controlled samples and routine interlaboratory communications, with manageable operation.

3. Current field data or re-test data should be mined for additional information, such as agreement between kits and the numbers of "tie-breakers" required by a technician or a facility. These could be important quality indicators. For example, there should be routine recording of all tiebreaker cases, including kit names (and lots) and tie-breaker result.

4a. In sites with low numbers of cases (<500), the likelihood of detecting errors is very poor without re-testing large percentages. This would apply to programs that require re-testing of all positives (low error rate, high power required).

4b. In sites with 500 or more cases there are opportunities for reasonable power for error detection, with feasible but large numbers of re-test cases.

4c. In situations with very large numbers of cases (3000 or more), then re-tested cases can be limited to a maximum number. For example, 200 to 250 cases seem to provide high power for detecting low error rates, so re-testing rates could be set accordingly.

Appendix H. On-site monitoring

Organization and management

The Ministry of Health must establish an organizational structure to assure that on-site monitoring occurs in all locations. This will require a sufficient number of staff who has been trained to conduct the monitoring. Persons with laboratory training and experience are preferred for monitoring the testing process, but an integrated approach that also looks at overall program needs is important; therefore a team approach to involve those with program experience will be helpful.

A decentralized scheme using staff from district or primary hospitals will likely be needed in order to adequately monitor all testing sites. There should be a plan for national evaluation, and a central data collecting point to assess the process.

Visits should be conducted at least twice yearly to established sites with experienced personnel. New sites or sites with new staff should be visited at least quarterly. In sites with demonstrated problems, the number of visits should be increased in order to provide training and technical assistance.

The findings from each site visit should be recorded according to the national policy, and the findings should be reviewed and corrective action taken when required.

Guidelines for visit

The routine on-site visit to the testing site should be scheduled in advance, so that all necessary preparations can be made. It is preferable to have all staff performing testing to be at work on the day of the visit.

During the visit, the assessor or team should meet with hospital, clinic, or testing site management to explain visit objectives, solicit input, feedback or concerns about HIV testing performance.

The visit to the testing site should include the following elements:

- Conduct an initial, brief meeting with staff to explain objectives, review schedule
- During the visit, complete all items on the checklist.
- Observe the testing facilities and look at current inventory of kits.
- Review standard operating procedures, EQA results, algorithm use.
- Observe on-site staff performing rapid HIV testing on client specimens.
- Observe process of recording results on test request forms and log book
- Review internal quality control results (including kit controls and external controls, if available)
- Assess turnaround time (estimate from laboratory staff, review of log books)
- Observe on-site staff performing a proficiency panel of specimens (5-10 specimens)
- Review results with laboratory staff when completed.
- Conduct a closing meeting with laboratory staff and medical officer or health care provider (review findings, solicit additional concerns corrective action plan with timeline).

A report and the completed checklist should be submitted to the relevant authorities for review and corrective action if needed. When report is finalized, copies should be provided to the monitoring officer (quality officer for the site) and to the management of the testing site.

The following is a suggested format for a report, as well as an example of a checklist to be used during the visit. These should be modified as needed to reflect national policies and experience.

HIV TESTING SITE VISIT REPORT

Site:

Date:

Assessment Team:

Major Findings:

Recommendations:

On-site Monitoring Checklist – Assessment of Quality System

Quality System Essential		Yes	No	Assessor's comments
Organization	• Is there a quality policy manual present and accessible? o Does the policy manual address all elements of the quality system?			
	• Does the site have a designated quality officer?			
	• Is the site manager aware of all quality system components?			
Personnel	• Does testing staff possess certificate indicating successful participation in HIV rapid test training?			
	• Has the staff been oriented to the patient/client flow at the test site?			
	• Does staff demonstrate professionalism?			
	• Is number of staff adequate for the site workload? o Approximately how many tests does each staff member perform per month?			
Documents and Records	• Are standard operating procedures for all aspects of the testing process written, up-to-date, and accessible to staff?			
	• Is the handwriting legible?			
	• Do worksheets include appropriate information?			
	• Are external quality control records up-to-date, easily reviewed?			
	• Are corrective actions recorded?			
	• Are results interpreted and recorded according to the SOP and site protocol?			

Quality System Essential		Yes	No	Assessor's comments
Purchasing and Inventory	• Are kits and reagents stored properly?			
	• Is staff following "first in, first out" method when managing inventory stock?			
	• Is there a policy for re-ordering kits and supplies?			
	• Is minimum stock on hand? (Look at re-order levels)			
	• Is the site ever unable to perform testing because there are no kits or supplies on hand? o How frequently does this occur?			
	• If unable to test, how is this information provided to clients?			
	• Does testing site have adequate space for storage of specimens?			
Equipment	• Is refrigerator clean and organized?			
	• Are refrigerator temperatures monitored and recorded?			
	• Are freezer temps monitored and recorded, if present?			
	• If centrifuge is used, are function checks routinely performed?			
	• If non-disposable pipettes are used, are calibration records available?			
Process Control ***Specimen Management***	• Is there a written procedure for collecting, processing and storing specimens?			
	• Are specimens appropriately labeled and packaged for transport to reference laboratory?			
	• Does staff follow universal or standard precautions when collecting or handling patient/client specimens?			
	• Do DBS or other collected specimens meet acceptance criteria?			
	• Observe staff perform rapid test using client specimen. All steps should follow SOP.			

Quality System Essential		Yes	No	Assessor's comments
Quality Control	• Are both internal and external quality control samples analyzed? o How often? o What is the source of QC material?			
	• Are external quality control results recorded and reviewed?			
	• Are appropriate corrective actions taken when controls results do not produce expected results?			
	• Is HIV status reported only when internal and external control is acceptable?			
Information Management	• Are test results properly maintained and accessible?			
	• Is a computer used in recording information and data? If yes, are there procedures to assure accuracy?			
	• Is patient/client confidentiality maintained? o If yes, is there adequate back-up to assure integrity of data in case of equipment (computer) failure?			
	• Is the testing site meeting program reporting requirements?			
Occurrence Management	• Is there a written policy for investigating errors?			
	• How is communication with affected customers handled?			
	• Are all errors, with any corrective action and communication, recorded?			

Quality System Essential		Yes	No	Assessor's comments
Assessment	• What EQA method(s) is used?			
	• Are results of re-testing submitted to testing site in timely manner?			
	• Are appropriate actions taken when re-testing results differ from initial results?			
	• Has the testing site been monitored within last 6 month?			
	• Have corrective actions been taken for previously identified problems?			
	• Are corrective actions taken and recorded?			
	• Are the results of on-site monitoring communicated to staff?			
	• Observe staff perform rapid tests using proficiency panel. (record results for each individual). All steps should follow SOP.			
Process Improvement	• Have any projects been undertaken for process improvement?			
Service and Satisfaction	• Is staff courteous to clients?			
	• Are there efforts to reassure client and/or alleviate client's fear of needle or sight of blood?			
	• When reporting to outside providers, is turnaround time appropriate?			
	• Does the site solicit input and advice from clients?			
	• Are program needs and expectations being met?			
	• Is the testing space adequate in size; clean and well organized?			
	• Is the environment suitable for patient testing (e.g. temperature, electrical supply)?			

Quality System Essential		Yes	No	Assessor's comments
Facilities and Safety	• Is storage space adequate, clean and maintained?			
	• Are gloves available and used routinely?			
	• Are hand washing supplies and facilities available in a convenient area?			
	• Are work areas disinfected?			
	• Is biohazard waste disposed of safely?			
	• Does the laboratory have policy and procedure for addressing accidental exposure to infectious material?			
	• Are emergency contacts located in visible area at test site?			
	• Is staff aware of policy for post exposure prophylaxis?			

Appendix I - Examples of documents and records useful in an HIV rapid testing site

- Example Specimen Transfer Log for Re-testing
- Rapid HIV Test Request Form

Example Specimen Transfer Log for Re-testing

Insert Name of Referring Testing Site,
Contact Name
Address and Phone Number

Date:_____

Referring Testing Site _____

Specimen Tracking Number	Test Subject ID*	Final Result (Testing Site)	Date Specimen Collected	Specimen Type (DBS or Serum)	Collected by	Referral Lab Req† Completed (√)	Date to referral lab	Date Conf Result Received	Result of Re-test

*ID = Identification
†Lab Req = Laboratory Requisition

RAPID HIV TEST REQUEST FORM

Site Code: _____ VCT Number:_____ Age_____Sex M/F

Code name of counsellor/person collecting blood
_____

Origin of sample *(hospital department – please tick)*

Antenatal clinic General VCT Counselling Area Medical Ward

TB Ward Outpatients Paediatric Ward

Purpose of testing *(reason for test - please circle)*

A = Pregnant woman/ANC attendee **B** = male partner **C** = VCT general client

D = clinical care/medical diagnosis **E** = TB patient **F** = Child given Nevirapine/other ARV at birth

Laboratory /Test Site Report

Date _____/_____/_____ **Lab /Test Site No**……

RESULTS

FIRST TEST _____ Lot No._____ Expiry Date_____

SECOND TEST _____ Lot No. _____ Expiry Date_____

REPEAT TEST

1 _____ Lot No._____ Expiry Date_____

2 _____ Lot No. _____ Expiry Date_____

TIEBREAKER

…… EXPIRY DATE _____
LOT NUMBER _____

FINAL RESULT _____

TESTS PERFORMED BY _____
(PRINT NAME AND SIGN)

Client Result Sheet

Site Code: _____VCT Number:_____ Lab Test Site no:_____

Code name of counsellor/person collecting blood _____

Origin of sample *(hospital department – please tick)*

Antenatal clinic General VCT Counselling Area Medical Ward

TB Ward Outpatients Paediatric Ward

Result
HIV Antibody Test Result ……Signed……


9 789241 593564