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Original Research Article

External Quality Assurance Scheme in a State Reference Laboratory for HIV Testing In Hyderabad

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ABSTRACT

Background: The Department of Microbiology of Osmania General Hospital / Osmania Medical College functions as the State Reference Laboratory (SRL) for HIV testing, covering 84 Integrated Counseling & Testing Centers and 24 Blood Banks of the Hyderabad, Ranga Reddy & Nizamabad districts of Telangana State. It's a first NABL accreditation lab in Government Sector of Telangana State. The External Quality Assurance Scheme (EQAS) in SRL implements Quality Control (QC) Testing, Proficiency panel testing and training programs.
Materials & Method: 7158 samples (5951 HIV negative/ 1207 HIV positive) were tested for QC. All the samples were tested using HIV 1st kit rapid test (Comb Aids), HIV 2nd kit rapid test (S.D Bioline), HIV 3rd kit rapid test (Trispot) and HIV Microlisa (Elisa). The QC samples consisted of 5% negative and 20 % positives, all the 84 ICTC were provided with 4 coded plasma samples (2 reactive & 2 negative) for proficiency testing using rapid tests. The aliquot panels (500µl) were provided twice a year for testing to monitor the laboratory performance.
Results: Out of 7158 samples tested for QC, 7156 (99.9 %) reported correct results and 2 (0.027%) discordant results. Out of 2 samples 1 (0.01%) were false negative and 1 wrong labeling (0.01%), transcriptional errors, tests that were not performed correctly were identified. For proficiency testing 100% reported test results. 3048 (100%) reported correct results. Hands on training were provided for the discordant centers and reported correct results on retesting.
Conclusion: Significant progress in establishing a well-coordinated HIV Laboratory network of SRL and ICTCs had been developed. However the HIV testing and Quality Assurance needs to be strengthened towards certification.

Keywords: Quality Assurance, HIV Testing in Hyderabad, SRL, ELISA, Trispot.

INTRODUCTION

Quality Assurance is the series of procedures that ensure that a correct result is achieved in a standard, reproducible and traceable manner. [1] It is achieved by a series of processes that assure the most accurate and highest quality result. To achieve a high quality result input from all members of every laboratory in testing network is required. All laboratory personnel should be aware of the necessity for quality performance. This requires

continuous education throughout the testing system.

The Department of Microbiology of Osmania General Hospital / Osmania Medical College functions as one of the State Reference Laboratory (SRL) in the External Quality Control Scheme (EQAS) under the National AIDS Control Organization (NACO), Government of India for HIV testing. The SRL is in charge of 84 Integrated Counseling and Testing Centers (ICTC) and 24 Blood Banks (BB) of Hyderabad, Rangareddy & Nizamabad Dist's of Telangana State.

The SRL tests Quality Control (QC) samples i.e., every 20th HIV negative (5%) and every 5th HIV positive samples (20%). Apart from QC testing, preparation of panels, hands on training and training programs on EQAS and QC testing for sentinel surveillance are the other activities.

The Diagnostic tests to detect antibodies to HIV have sensitivity and specificity which are not absolute. In all these tests we have false negative and false positive results which are inherent and cannot be avoided. Thus the validity of diagnostic test results is dependent to a very large extent on the quality of the technical conditions under which the tests are performed. Meaning thereby, consistent reproduction of reliable results requires a stringent overall Quality Assurance program which would control technical conditions before, during and after each assay.

The Center for Disease control (CDC) developed guidelines for implementing and operating Quality Assurance programs. It recommends testing sites to participate in external quality assessments i.e., Proficiency testing of panels, competency assessments and QA monitoring by outside organization observing the testing. [2] The State Reference Laboratory (SRL) implements the External Quality Assurance Program for HIV testing to provide standard quality results.

MATERIALS & METHODS

This is a retrospective study. The Quality Control (QC) samples were received from the 84 Integrated Counseling and Testing Centers (ICTC) and 24 Blood Banks (BB) of Hyderabad, Ranga Reddy & Nizamabad districts of Telangana State, by hand delivery from 2010 to 2015. The ICTC/ Blood Banks test the samples in their respective centers. After testing the QC samples alone are sent to the SRL. All the samples are received at SRL after rechecking for cold chain maintenance temperature with a digital thermometer and screening for quality of samples, every 20th HIV negative sample (5%) and every 5th HIV positive samples (20%) from the ICTC are tested as QC samples in the SRL. 7158 samples were received in the SRL for QC testing (1207 positive and 5951 negative). In QC testing all the samples are retested using Comb Aids (Span Diagnostics Ltd, Surat, India) as the 1st rapid kit. It employs the same principle as EIA thereby the immobilized antigen antibody complex is visualized by means of colour production (chromomeric reaction). Each comb was observed for the control and the test results. The HIV positives alone were retested by the 2nd kit HIV S.D Bioline ½ 3.0 Rapid test (Bio Standard Diagnostic Pvt. Ltd) and the 3rd rapid HIV ½ Aidscan Trispot (Bhat Bio-Tech India (p) Ltd). NACO strategy of using 3 rapids for HIV testing was followed. [3] Insufficient and hemolysed samples, samples that were not properly labeled were excluded and informed to the testing centers. Apart from the routine samples and kit controls Internal Quality Control samples in house prepared samples basing on L.J Charts were used during each inter quality control preparation. The test kits were provided by the APSACS (Andhra Pradesh

State AIDS Control Society). All discordant samples were discussed with the testing center and were followed up with hands on training for the laboratory technicians. It is a continuous process where the SRL plays a key role in monitoring and evaluating the ICTCs & Blood Banks.

The SRL conduct the EQAS program every quarterly i.e. on January, April, July & October Apart from QC testing two training programs were conducted twice a year for the ICTCs. One laboratory technician represented each ICTC. The training comprises lectures on Q.C scheme, hands on training in HIV testing and panel distribution. The SRL provide the ICTC's with 4 panels (2 HIV positives & 2 negative coded samples) during the training program. Panels were prepared by SRL procuring plasma from NRL. Using ELISA and HIV rapid tests, the panels were validated by sending them to the National Reference Laboratory before the training program. A report was sent from the NRL to SRL informing the results and samples that had concordant results were used for proficiency panel testing. Aliquots were further prepared and stored in at -20˚C. The SRL conducts a one day training program for ICTCs & Blood Banks and provide the panels. ICTC/ Blood Banks to submit the Proficiency Panel reports to SRL within 7 working days The reports of the 84 ICTC's provided a feedback and this enable the SRL to train them periodically and improve their performance in HIV testing.

RESULTS

Seven thousand one hundred fifty eight (7158) Quality control (QC) samples were re-tested at the State Reference Laboratory during a period of 5.7 years. Out of 7158, 1208 samples were HIV positives (16.87%) and 5948 were HIV negative samples. The samples were retested using three different HIV rapids test kits. The 7158 samples were tested using Comb AIDS kit as the first rapid kit. Out of 7158 samples tested, 7156 reported concordant results. 5948 QC samples were HIV negative and 1208 QC were positive for HIV. 2 samples were reported discordant results, 1 false Negative, 1 wrong labeling (Table 1).

Out of 1207 QC positive samples reported from the SRL, were HIV positive. Similarly out of 5951 QC negative samples, 1 tested false negative and one sample with wrong label the remaining 5949 were negative (Table 2). The QC positive samples, 1 false negative sample were retested using the second HIV rapid test SD Bioline & Aidscan Trispot. 1207 samples were positive, reported 2 false negative in HIV rapid test SD Bioline & Aidscan Trispot. The above QC samples were again retested with the third HIV rapid test kit and similar results were observed. 2 discordant samples reported the same results in all the three rapid kits. The QC sample result & discordant samples were reported to the respective centers. The main reasons for discordant samples were, labeling error and transcriptional errors.

Twice a year proficiency testing panels (8coded samples) & bulk validated plasma were provided by NRL to SRL, SRL Osmania General Hospital will test & submit panels report to NRL, NRL issues concordance report of Proficiency Panel to SRL,

Aliquots were further prepared and stored in –20˚C. The SRL conducted a one day training program for ICTCs & Blood Banks and provided the panels (4 coded samples). ICTC / Blood Banks to submit the Proficiency Panel reports to SRL with 7 working days.

Table – 1 EQAS retesting results from ICTC- SRL – NRL

Total No of QC Samples	Concordant samples	Discordant samples	
7158	7156 (99.97%)	2 (0.02%)	
		False Negative 1(0.01%)	Wrong labeling 1(0.01%)

Table – 2 EQAS retesting results from ICTC- SRL – NRL

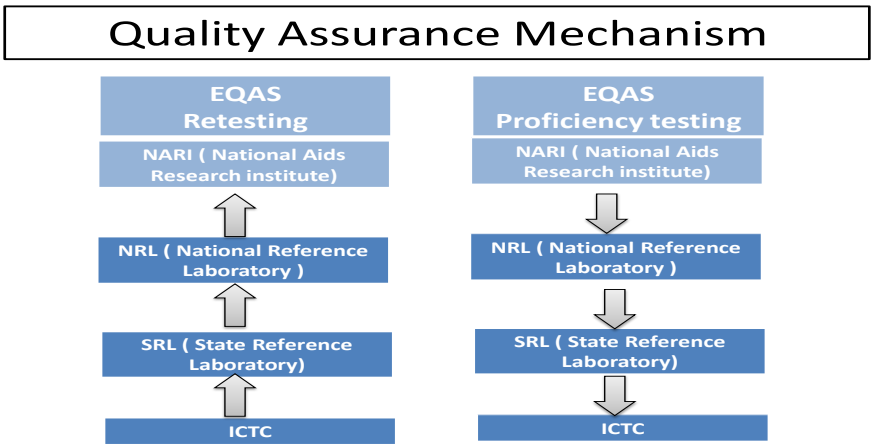
Year	Total Number of samples received from ICTC	Reported HIV Positives at ICTC	Reported HIV Negative at ICTC	HIV Positives at SRL	HIV Negatives at SRL	Discordant results	Total no of samples sent NRL for EQAS retesting	Total no of HIV Positives	Total no of HIV Negatives	Discordant sample sent to NRL	% Concordance results
2010 - 2015	7158	1207	5951	1207	5949	2	243	117	124	2	100%

Table 3 Testing: Re testing results

No. of samples	HIV 1st kit: Combaids results for		HIV 2nd Kit SD Bioline results	HIV 3rd Kit Aidscan Trispot results
	Negatives	Positives	Positives	Positives
7158	5949	1207	1207	1207

Table 4 Proficiency testing results From NRL – SRL – ICTC

Proficiency testing NRL – SRL (2010 – 2015)						Proficiency testing SRL - ICTC (2010 – 2015)					
No. of Proficiency samples received from NRL	No. of Samples tested at SRL	Negatives	Positives	No. of samples discordant	% of Concordance	No. of Proficiency samples Issued to ICTC	No. of Samples tested at ICTC	Negatives	Positives	No. of samples discordant	% of Concordance
88	88	45	46	0	100%	3048	3048	1547	1501	0	100%



SRL distributed to ICTC's / Blood Banks during the training program. Out of 74 centers, 63 presented in proficiency panel programme (85%) reported the panel results to the SRL. 6 (9.5%) centers did not send the report. 57 Out of 63 centers, (100%) reported correct results.

DISCUSSION

The SRL is involved in Quality Control testing, proficiency panels, conducting training programs and providing hands on training for HIV testing. It is also involved in sentinel surveillance QC testing which is conducted annually by NACO. Quality Assurance program ensures high level of performance of HIV assays. [4] As a SRL the Quality Assurance program ensures

the participating centers are correctly testing, getting consistent results and it checks their validity of results.

In India the EQAS program had been implemented by NACO since 2000. It functions with 1 Apex Laboratory, 13 National Reference Laboratories, 118 State Reference Laboratory and thousands of sub centers covering the entire Government Medical Colleges, Hospitals, blood banks and primary health centers in each state. This EQAS program is linked with the respective State AIDS Control Society (SACS) in each state for the effective functioning under the NACO. Out of 7158 samples tested for QC, 2 samples had discordant results: 1 false negative and 1 with wrong labeling. In the follow up training program, they were instructed to follow the Standard Operating Procedures (SOP), use of correct pipetting technique, use of single tip for each sample, avoid transcriptional errors and prevent sample contamination. Participation in proficiency testing is a key component of any Laboratory Quality assurance program, whether available locally, nationally or internationally. [5]

Quality Assurance program should be in place to continuously assess and improve the performance of Laboratory results. It is important for the physicians in guiding the patient for treatment and for further management. All aspects of sample handling right from the arrival to reporting must be monitored, documented and subjected to quality control procedures. It is also suggested that regular audits of laboratory procedures and reviewing incident reports must be carried out by the senior staff who are in charge and should be discussed with the Director or the Head of the Laboratory. [5, 6]

The Standard Operating Procedures (SOP) is an important component of the Quality Assurance program. All the members of the laboratory should be familiar with the procedures. The SOP's are reviewed every year and no deviations from the procedures in order to get correct results. Reagent and equipment performance must be monitored over time to detect any changes in quality and integrity. [7]

The storage of reagents and expiry date too plays an important role in the performance of testing. If controls are not working faint spots or lines may develop. It will lead to misinterpretation of results. Calibrating pipettes and centrifuges annually should be done in the laboratory. The temperatures of the refrigerators are monitored by a digital thermometer where HIV testing kits are stored. Expired kits should not be used.

In Catalonia, Spain a survey on HIV testing was undertaken to assess the quality of HIV testing. HIV specimens were identified by specific labels, extracting new specimens for a second test to confirm, or failing to guarantee the confidentiality of results was found. [8] In a QA program the pretesting, testing and post testing should be monitored, observed and evaluated. The recommendations will play an important role in improving the centers.

In China the National AIDS Reference Laboratory (NARL) provides quality assurance through technical, bio safety and managerial trainings, periodic proficiency testing, on site supervisory inspections and commercial serological kit evaluations. [9] The 7158 samples tested for QC provided a feedback mechanism on the performance of the respective centers. It provided an opportunity to pick up discordant samples or it could have been left undetected. After the implementation of the EQAS program the standards of HIV testing had been improved.

Rapid test with immediate test result are popular in the US and Canada. It requires good laboratory quality control

practices. The laboratory must be backed by a license to provide confirmation of positive results and resolution for indeterminate results. Quality Assurance and participation in HIV proficiency testing ensures the accurate, timely and clinically relevant laboratory results. [7]

The EQAS training twice a year and proficiency testing was aimed to monitor testing at ICTC's who in turn apply the same methods to the sub centers. 74 Out of 63 centers provided with proficiency panel, 57 (90 %) reported the results. 9.5 % did not report. The main reason being the center did not participate in the training program. And 57 centers 100% reported correct results.

In Poland there is no guidelines related to HIV diagnosis quality assurance and control. Hence developing of a National Unified quality control system based with a central institution is highly desirable. This was based on the survey conducted to assess the laboratory performance. It was also suggested certification be made mandatory for all diagnostic laboratories. [10] The aim was to improve the reliability of results among the clinicians and the patients.

The proficiency testing provided an opportunity for the ICTC centers to know their abilities and improve their performance. They were also provided with forms to fill up the results i.e., kit name; lot No, expiry dates, results of controls and samples and the final interpretation of results. These forms were provided by the Apex Laboratory and it provided an opportunity for the Lab personnel to read and fill in the forms. All the 63 ICTC centers were provided with unique codes identifying with the SRL, District and their VCTC, BB centers.

In India five laboratories were identified as Regional Institutes (RIs) to monitor and supervise the Quality control practices and assurances in the sentinel surveillance testing covering five states. The testing laboratories were adhering to the standard Operating Procedures. Concordance of test result between the RI and laboratories was high. The lacunae and the recommendations were put forward for the future surveillance. [11]

CONCLUSION

Significant progress had been made in establishing a well coordinated HIV Laboratory network through EQAS. In this program it ensures mistakes may be avoided by proper sample handling, labeling, testing and reporting. The consistency of performance is maintained. The Quality Assurance system with proficiency testing, quality control testing, training programs and on-site monitoring strengthens the participating laboratories and provides opportunity for improvement.

SRL – Department of Microbiology, Osmania General Hospital / Osmania Medical College achieves NABL accreditation in 04.08.2014 with accordance ISO -15189:2007, recently we have completed NABL Surveillance visit with accordance ISO-15189-2012 by Dr.Vani Ravi Kumar (Lead Assessor) & Dr.H.Srinivasa (Technical Assessor) from Bangalore on 4.7.2015 with certification is in process. The certification process which covers the entire laboratory performance will provide opportunities for filling the gaps and strengthening their performance.

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REFERENCES

1. E. M. Dax, "External Quality Assurance Scheme in HIV testing Guidelines," State Reference Laboratory, Melbourne, 2000. www.srl.gov.au

2. Centers for Disease Control and Prevention, "Quality Assurance Guidelines for Testing Using Rapid HIV Antibody Tests Waived under the Clinical Laboratory Improvement Amendments of 1988," 2007. http:www.cdc.gov/hiv/topics/testing/resources/guidelines/pdf/QA_Guidlines.pdf
3. National AIDS Control Organization, "Guidelines for HIV Testing," 2007. www.nacoonline.org
4. D. E. Dwyer, "Advances in HIV Laboratory Testing," NSW Public Health Bulletin, 2010; 21(3-4): 86-88.
5. D. W. Tholen, L. M. Berte, D. J. Boone, W. G. Cooper, J. Gun-Munro, M. A. Noble, S. J. Sarewitz and M. L. Williams, "Using Proficiency Testing to Improve the Clinical Laboratory, Approved Guideline," CLSI/NCCLS GP27-A, 1999;27 (8)
6. G. B. Clarke, S. J. Sarewitz, K. J. Aziz, K Castaneda Mendez, L. K. Miller. E. Simson and D. W. Tholen, "Continuous Quality Improvement: Essential Management Approaches: Approved Guideline," CLSI/NCCLS GP22-A,1999;19(13)
7. M. Fearon, "The Laboratory Diagnosis of HIV Infections," The Canadian Journal of Infectious Diseases and Medical Microbiology 2005; 16(1):26-30.
8. M. J. Casado, A. Rovira, C.Blanch and J. Casabona, "Survey of the Performance of Anti-HIV Antibody Detection Tests in Laboratories in Catalonia (Spain)," Gac Sanit Journal 2004; 18(1):68-71.
9. Y. Jiang, M. Qiu, G. Zhang, W. Xing, Y. Xiao, P. Pan, J. Yao, C. Y. Ou and X. Su, "Quality Assurance in the HIV/AIDS Laboratory Network of China," International Journal of Epidemiology 2010; 39 (2):72- 78.
10. M. Parczewski, K. Madalinski, M. Leszozyszyn-Pynka and A. Boron-Kaczmarska, "Evaluation of Quality of HIV Diagnostic Procedures in Poland," Przegi Epidemiol Journal 2010; 64(1): 47-53.
11. S. Kant, K. Goswami, S. K. Rai, L. Dar and P. Misra, "Quality Assurance of Annual HIV Sentinel Surveillance: Experience of Regional Institute, Central Zone," Indian Journal of Public Health 2007; 51(1):28-32.

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