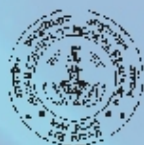


GUIDELINES FOR GOOD CLINICAL LABORATORY PRACTICES (GCLP)



INDIAN COUNCIL OF MEDICAL RESEARCH, NEW DELHI
2008

GUIDELINES FOR GOOD CLINICAL LABORATORY PRACTICES (GCLP)



Indian Council of Medical Research
New Delhi

2008

Published by:

Director-General

Indian Council of Medical Research

New Delhi-110029

www.icmr.nic.in

September, 2008

© Copyright Indian Council of Medical Research

Production Controller :

J.N. Mathur, *Press Manager, ICMR, New Delhi*



भारत सरकार
स्वास्थ्य एवं परिवार कल्याण मंत्रालय
निर्माण भवन, नई दिल्ली - 110011

Government of India
Ministry of Health & Family Welfare
Nirman Bhawan, New Delhi-110011

Naresh Dayal

Health & FW Secretary
Tel.: 23061863 Fax : 23061252
e-mail : ndayal@nic.in

FOREWORD

As a developing country India is making advances in the field of technology, research, infrastructure and skilled manpower. However, much of these advances are limited to metropolitan cities and urban areas. In healthcare too, such disparity exists. The Indian Council of Medical Research has the mandate to foster inter-sectoral coordination in health research between scientific departments, private sector, academia; strengthen their network; develop trained manpower for research and develop systems that will bring useful advances in health research available to all sections of society.

Guidelines for Good Clinical Laboratory Practices (GCLP) outlines the principles and procedures to be followed by medical laboratories involved in clinical research and/or patient care so as to provide quality data which can be used for health research and patient treatment. As the use of laboratory tests (often expensive) are increasingly becoming a part of medical diagnosis and research, generation of quality data would be a cost-effective and ethically sound strategy.

Adoption of GCLP guidelines by laboratories of public sector, private sector and research institutions will be a step forward in the betterment of health care services and health research.

I congratulate the team of Scientists from ICMR for bringing out Guidelines for Good Clinical Laboratory Practices.

(Naresh Dayal)

July 2008



सम्पर्क से पहले सोचो, एच आईवी/एडस से बचो **HIV/AIDS : Prevention is better than cure**

PREFACE

Laboratory tests are used to support diagnosis in patient care as well as medical research. The test results therefore should be reliable, accurate and reproducible. Generation of such 'quality' results involves a step wise process of meticulous planning, perfect execution and thorough checking of results by the whole team involved. International guidelines lay down the principles of Good Clinical Laboratory Practices (GCLP) to be followed by medical researchers to generate 'quality data' but do not address the issues of reporting 'quality' test results for day-to-day patient care.

The Council carries out intramural and extramural research through a number of institutes which are also engaged in day-to-day patient care. Though most laboratories follow some measures to ensure generation of reliable results, there is a need for a uniform procedure to be adopted by the laboratories to allow intra and inter-laboratory comparisons. Keeping this in view, the Indian Council of Medical Research constituted an Advisory Committee to adapt the existing international guidelines to the Indian context. These guidelines can be followed by laboratories engaged in medical research as well as public and private laboratories involved in patient care. We gratefully acknowledge the guidance and support of all experts of the Advisory Committee and the Reviewers in preparing these guidelines.

ICMR laboratories are expected to play a lead role in adoption of these guidelines as well as provide support for consultancy, training and surveillance activities which will promote adoption of these guidelines by non-ICMR public and private sector laboratories. It is also expected that over a period of time a network of GCLP compliant laboratories can be established which would improve the credibility and authenticity of their test results. This would save enormous time, money and resources.

Vasantha Muthuswamy
Scientist 'G' & Head
Div of BMS & RHN
ICMR

S. K. Bhattacharya
Addl. Director General
ICMR

ACKNOWLEDGEMENTS

We gratefully acknowledge the valuable contribution of the Chairperson and members of the Advisory Committee for providing guidance and support in drafting and giving a final shape to the guidelines. The members enthusiastically participated in discussions and provided immensely useful inputs drawn from their vast experience in the subject. We also thank the Reviewers for their suggestions and timely advice.

We are grateful to Dr S K Bhattacharya for envisioning the need for GCLP guidelines in the Indian context and encouraging us to take the initiative to draft them. We also thank Dr V Muthuswamy for being a constant guide and support in drafting the guidelines.

Our special thanks to Mr Raj Kumar and his staff for smoothly organizing the Advisory Committee meetings and Mr Mohan Kant for secretarial assistance.

New Delhi
July 2008

Shalini Singh
A.S. Kanagasabapathy
Sambhu Chakraborty

CONTENTS

	PAGE NO.
1.0 INTRODUCTION	1
2.0 SCOPE	2
3.0 LEVELS OF LABORATORIES	3
4.0 INFRASTRUCTURE	4
5.0 PERSONNEL, TRAINING & DEVELOPMENT	4
6.0 EQUIPMENT	5
7.0 REAGENTS AND MATERIALS	7
8.0 SPECIMEN COLLECTION	8
9.0 REQUISITION FORM	8
10.0 ACCESSION LIST	9
11.0 WORKSHEET	9
12.0 REPORTING TEST RESULTS	10
13.0 SPECIMEN REJECTION RECORD	10
14.0 DATA MANAGEMENT	10
15.0 STANDARD OPERATING PROCEDURE (SOP)	11
16.0 SAFETY IN LABORATORIES	13
17.0 ETHICAL CONSIDERATIONS	17
18.0 QUALITY ASSURANCE	18
19.0 QUALITY ASSURANCE PROGRAMME	18
20.0 INTERNAL QUALITY CONTROL	19
21.0 EXTERNAL QUALITY ASSESSMENT	21
22.0 INTERNAL AUDIT	22
23.0 SUMMARY OF QAP ACTIVITIES	22

BIBLIOGRAPHY	23
ANNEXURES	
Annexure 1. Equipment Log Book	25
Annexure 2. Requisition Form	26
Annexure 3. Accession List	27
Annexure 4. Worksheet	28
Annexure 5. Requisition Form cum Worksheet	29
Annexure 6. Reporting Format	30
Annexure 7. Specimen Rejection Record	31
Annexure 8. Steps to Initiate IQC in Laboratories	32
Annexure 9. Calculation of Mean, Standard Deviation and Tolerance Range	35
Annexure 10. Corrective Actions	38
Annexure 11. Basics of EQA	39
Annexure 12. Understanding Accreditation of Clinical Laboratories	41
ADVISORY COMMITTEE MEMBERS	43
DRAFTING COMMITTEE MEMBERS	45
REVIEWERS	46

GOOD CLINICAL LABORATORY PRACTICES (GCLP)

1.0 INTRODUCTION

Laboratory services are an integral part of disease diagnosis, treatment, monitoring response to treatment, disease surveillance programmes and clinical research. The World Development Report 1993, regarded provision of Essential Health Technology as an important ingredient of Essential Clinical Services. Use of diagnostic techniques aid early diagnosis enabling appropriate and prompt intervention thereby reducing overall disease burden and promoting health. All laboratories are not equipped with facilities for carrying out complex investigations. The structure and function of a clinical laboratory varies according to the level of health care facility. Peripheral laboratories carry out simple tests like urine analysis and haemoglobin estimation whereas higher centers are equipped with sophisticated technology and trained manpower to carry out complex investigations. Establishing a network between peripheral and higher laboratories allows collection of specimen at periphery and their storage and transport for testing at higher centers and communicating report to the peripheral center efficiently without actually having to transfer the patient. In the event of patient transfer, the higher centers do not need to repeat investigations carried out at the peripheral health center, thereby saving crucial time as well as cost and providing continuity in patient care. Networking between laboratories is also essential in disease surveillance programmes and outbreak investigations in order to obtain quick and reliable results.

The expert committee on Revamping of Public Health System identified the surveillance and control of diseases as an important function of public health system. This formed the conceptual framework of Integrated Disease Surveillance Programme (IDSP) which was started in 2004 in a phased manner. The key components of IDSP are coordination and decentralization of disease surveillance activities, improvement of laboratory support and strengthening data quality. Inclusion of private laboratories to act as sentinel sites and improve community participation are some other important features. The Reproductive and Child Health - II programme of Government of India in 2005, indicated the need for developing public-private partnership in health care including outsourcing of health care, laboratory services and others. It outlines the need for accreditation of service quality including protocols for quality assurance and certification. In India, medical laboratories can volunteer for accreditation of one or more services offered by them. The National Accreditation Board for Testing and Calibration Laboratories (NABL) has been providing accreditation services to medical laboratories since 1998 and is currently following ISO 15189; 2007 standards.

In biomedical research too, achieving a set standard of quality produces credible results and allows comparison between studies carried out at different institutes nationally and internationally. This saves enormous time, money and resources and prevents duplication of research work. The International Conference on Harmonization (ICH) provided Good Clinical Practices (GCP) Guidelines which describe standards

to be followed by researchers while designing, conducting and reporting trials involving human participants. Realizing the rapid pace, wide spectrum and potential for clinical research in our country, the Indian Council of Medical Research (ICMR) launched the Ethical Guidelines for Biomedical Research on Human Subjects in 2000 (revised in 2006) and Central Drugs Standard Control Organisation (CDSCO) released the Indian Good Clinical Practices (GCP) guidelines in 2002 to guide biomedical research in the country. To harmonize practices and generate mutually acceptable data for non-clinical health and environmental **safety studies the Organization for Economic Co-operation and Development (OECD)** evolved Good Laboratory Practice (GLP) guidelines. India is a signatory to OECD and **National GLP Compliance Monitoring Authority** established in the year 2002 by the **Department of Science & Technology**, Government of India, provides GLP compliance certification to the test facilities involved in conducting safety studies on chemicals (viz. industrial chemicals, pharmaceuticals, veterinary drugs, pesticides, cosmetic products, food products, feed additives, etc). To ensure reliability of data quality, WHO/TDR (Research and Training in Tropical Diseases) has developed good practice guidelines for laboratories involved in clinical trials. However, standards have not yet been developed in India for this purpose. The proposed ICMR **guidelines for Good Clinical Laboratory Practices (GCLP)** aim to elucidate step wise procedures which should be followed by laboratories to strengthen the quality of test results. **These guidelines should be adopted by all ICMR laboratories engaged in research as well as patient care. ICMR** carries out research activities through its own institutes as well as through approved research centers in the public and private health systems. It also provides financial support to projects submitted by individual researchers and institutes. Adopting these guidelines will lead to generation of uniformly acceptable and good quality laboratory data. **Subsequently, a checklist will be prepared to monitor these laboratories for compliance with these guidelines.**

2.0 SCOPE

Good Clinical Laboratory Practices should be used by all laboratories where tests are done on biological specimens for diagnosis, patient care, disease control and research such as:

- Microbiology & Serology
- Hematology & Blood Banking
- Molecular Biology and Molecular Pathology
- Clinical Pathology
- Clinical Biochemistry
- Immunology (Immunohematology and Immunobiochemistry)
- Histopathology/Pathology and Cytology

3.0 LEVELS OF LABORATORIES

In India, the laboratory services are integrated with the 3-tier public health system at the primary, secondary and tertiary levels. Besides these, there are Reference Laboratories, Research Laboratories and Specific Disease Reference Laboratories to provide services for complex and special tests. The private sector provides laboratory support at all levels of health care both in rural and urban areas. Each laboratory should identify the scope, functions and the capacity of the services offered by it and appropriate infrastructure with requisite biosafety measures should be planned. Qualified and trained staff should be employed with periodic up-gradation of their skills.

3.1 Primary Level

Simple laboratory tests such as haemoglobin estimation and urine examination for albumin and sugar are carried out at Primary Health Centers (PHCs) and Urban Health Centers (UHCs) by laboratory technicians. Most PHCs and UHCs also have microscopy facilities and trained technicians for examining blood smear for malarial parasite and sputum for acid-fast bacilli (AFB) and a cold chain system. The Community Health Centers (CHCs) receive referrals from PHCs and the laboratory technicians are trained and equipped to handle additional laboratory investigations for the management of medical and surgical emergencies and making etiological diagnosis of RTIs/STIs. Facilities for screening of G6PD deficiency, sickle cell anaemia and thalassemia are also available for vulnerable communities. Under IDSP, training will be provided for diagnosis of typhoid using kits, detection of chlorination levels and fecal contamination in water samples.

3.2 Secondary level

The district hospitals have facilities and manpower for carrying out pathology, clinical pathology, biochemistry, serology, and microbiological investigations. They also carry out tests of water quality and receive referrals from primary level facilities. The laboratory staff includes pathologists, microbiologists, cytotechnicians, laboratory technicians, blood bank technicians and laboratory attendants.

3.3 Tertiary level

The medical college hospitals and non-teaching large hospitals are equipped with sophisticated diagnostic and investigative facilities to provide tertiary level health care. These hospitals receive referrals from the primary as well as the secondary levels.

3.4 Reference Laboratories, Research Laboratories and Specific Disease Reference Laboratories

The Reference Laboratories, Research Laboratories and Specific Disease Reference Laboratories provide services in a specialized field or area of importance. These may be located in a medical college, research institution or a private institution. They set and should maintain high standards of quality in one or more particular area and therefore receive referrals specific to that field. They also offer consultancy, standardize diagnostic tests and carry out training pertaining to that specific area.

4.0 INFRASTRUCTURE

Infrastructure of laboratories should be planned according to the services provided by the laboratory. The basic infrastructure facilities include:

- Reception room/area where requisition forms are received and reports disbursed
- Specimen collection room/area, toilets, privacy for special purposes eg. semen collection, facilities for disabled persons, toilet for staff
- Quality water supply for analytical purpose
- Uninterrupted power supply
- Analytical work area
- Specimen/Sample/slide storage facility including cold storage where applicable
- Record room/area
- Facility for cleaning of glassware, sterilization /disinfection
- Waste disposal facility including biomedical wastes
- Fire-safety equipment
- Ventilation, climate control and lighting arrangements
- Separate room/area for meetings/administrative work
- Separate facilities/area for staff for hand washing, eating and storing food, drinks etc.
- Communication facility with referral centers
- Transport of specimen/samples to referral centers
- Additional infrastructure facilities may be added for special tasks as and when needed.

5.0 PERSONNEL, TRAINING AND DEVELOPMENT

- Each laboratory should designate a **Head** of the laboratory who should be overall in-charge of the daily functioning of the laboratory including administration. A **Quality Manager** should be designated for monitoring and maintaining of day-to-day quality management system.

- The qualifications and experience of the staff outlined in NABL document 112 (2007) should be followed unless specified by the health care providers.
- The strength of staff employed should be appropriate to the level of facility and the workload.
- The roles and responsibilities of the staff should be clearly outlined. The staff should also understand the nature of work assigned to them and must be capable of performing the tasks independently beyond routine working hours if the need arises.
- A programme for technical training and updating of skills on a regular basis should be in place. The laboratory management should be committed for providing continuing professional development and training opportunities to staff. Action plan for improvement in the laboratory should be determined and revised according to the feedback received from previous trainings and experiences.
- Laboratory should organize or conduct periodic staff evaluation, preferably once a year; frequency and method of evaluation should be decided by the laboratory.
- The laboratory should maintain a personal file of all the technical and non-technical staff employed. Personal file should contain all information on:
 - Personal bio-data including educational qualification and experience
 - Copy of degree/diploma and registration with state authority if applicable
 - Copy of appointment letter
 - Duly verified health information (physical fitness including color blindness, immunizations received etc.) prepared at the time of employment and its regular updates
 - Performance appraisal
 - Training certificates, awards/recognition received
 - Disciplinary action if any taken by the management
 - Reference letter from previous employer if applicable

6.0 EQUIPMENT

- Each laboratory should prepare an exhaustive list of equipment and consumables required and available for general functioning of the laboratory and specialized equipment for special tests.
- Laboratory equipment should be of adequate capacity to meet work load requirement.

- Equipment should be suitably located in the laboratory so as to allow accessibility and sequential utilization thus minimizing the need for frequent movement of specimens or reagents.
- All equipment should be in good working condition at all times. Periodic inspection, cleaning, maintenance of equipment should be done. An equipment log book should be maintained for all major equipment. Laboratories should maintain necessary instructions for operation and maintenance of equipment in the form of Standard Operating Procedures (SOPs). A copy of SOP should be readily available.
- Maintenance contracts including warranty cards, telephone numbers of staff to be contacted in case of equipment malfunction should be kept safely. User manual should be available readily for reference. The staff should be aware of trouble shooting measures to be adopted for preventing equipment malfunction. A format of the equipment log book provided in Annexure 1 can be used.
- New equipment should be calibrated and validated before routine use. AMR (Analytical Measurement range) should be verified, manufacturer can be consulted for verification and selection of range.
- Periodic performance check/calibration check for all equipment should be done using reference standard/reference material. The frequency of performance check should be based on the day-to-day performance of the equipment.
- Equipment performance should be verified from Internal Quality Control results and External Quality Assessment results. Outlier parameter trend analysis record should be maintained in respect of its effect on the equipment.
- All analytical equipment should be calibrated and calibration certificate provided by equipment company. Non-analytical equipment such as pipette, thermometer, weighing balance and centrifuge should be calibrated by accredited calibration laboratory or done in-house with traceability to National Physical Laboratory (NPL). For in-house calibration, laboratories should use :
 - Calibrated tachometer - for centrifuge
 - Calibrated digital temperature sensor - for checking temperature of refrigerator, incubator etc.
 - Calibrated glass thermometer- for temperature checking of oven, water bath etc.
 - Calibrated weights - for balance

- National Institute of Science and Technology (NIST) buffer - for pH meter. Standard buffer solutions bought from reputed manufacturers with certifiable traceability can be used as alternative.
- Equipment measuring pressure, temperature, humidity etc. should be checked by using suitable reference standards.

7.0 REAGENTS AND MATERIALS

- Standard reagents of certified quality must be used for the purpose of analysis. The batch number of reagents must be recorded. The quality of the reagent viz. Analar grade, HPLC grade, etc. to be used for in-house procedures should be defined in SOP
- The reagents, chemicals and consumables should be stored under appropriate environmental conditions.
- Quality of newly purchased reagents should be validated against suitable control/reference material prior to use. Validation data should be properly documented. In-house prepared reagents should also be checked periodically for stability and a record of the same should be maintained.
- Reagent label should contain name of reagent, concentration, date of preparation/opening, date of expiry, storage conditions and warnings eg. 'do not use if solution is turbid' where applicable. When individual bottles are small, this information can be recorded in a goods received ledger.
- Microbiology laboratories should check activity/potency of each lot of antibiotic sensitivity discs before using and at least weekly thereafter with reference strains. Other microbiological consumables such as strips etc. used for identification should be checked against reference strains. Laboratories testing microbiology specimens should check the quality of media by using appropriate reference strain and pH of the media.
- All batches of culture containers should be checked for sterility before issuing to patients for collection of specimen.
- Water quality should be checked for its grade and presence of interference elements. Reagent grade water according to IS1070 : 1992 of Bureau of Indian Standards (BIS) should be used for testing.

8.0 SPECIMEN COLLECTION

- Specimen collection is the first phase of interaction between the patient and the laboratory. Appropriate counselling should be done before specimen collection and consent taken whenever needed. Attention should be paid to patient's sensibilities during the entire process. Any error in specimen collection can lead to erroneous results. It is therefore considered an important step of good clinical laboratory practice and is referred to as "**preanalytic control**".
- Specimen collection can be done at the patient's bedside, in the laboratory or in the field.
- Trained manpower/phlebotomist should be employed for specimen collection. Other staff such as doctors, nurses and others who are involved in specimen collection should also be trained periodically.
- Laboratory should have a "primary specimen collection manual", containing information on patient preparation before specimen collection (if any), exact methodology of specimen collection, labelling, handling, transportation and storage of the specimens. In addition, the laboratory should provide adequate and appropriate information/instructions to patients wherever necessary. All preanalytical factors that may influence the test results should be identified. The manual should include guidelines on specimen collection including preservation for histopathological examination. These manuals should be available for reference and should be used for training of staff engaged in specimen collection.
- Specimen should be secured properly so that there is no leakage, spillage or contamination. A Biohazard symbol should be used on the containers during transportation. Appropriate specimen transportation kit (such as use of dry ice, etc.) to be used wherever required. Specimen should be sent to the laboratory along with the requisition form.

9.0 REQUISITION FORM

- The requisition form should be completed by the doctor requesting the tests and sent along with the specimen/patient to the laboratory.
- It should contain the patient's identity, age, location, date of specimen collection and the investigations requested. The referring doctor should be encouraged to mention the provisional or working diagnosis and relevant clinical and treatment history in the space provided (Annexure 2).

10.0 ACCESSION LIST

- Accession list is a record of all the specimens received by the laboratory for analysis and is prepared by the laboratory at the time of specimen receipt (Annexure 3).
- It records the patient's identity including name, age, sex, location in the hospital/ medical facility, name of referring physician, investigations requested, date and time of receipt of specimen and condition of the specimen at receipt. The laboratory assigns a unique laboratory number to register each specimen received, which can be used to trace the specimen in the laboratory. The test results and remarks if any are also entered in the accession list.
- In laboratories handling a very large number of specimens, the accession list may be computer generated and the condition of specimen at receipt may not be recorded unless it has been rejected.

11.0 WORKSHEET

- Worksheet is essentially a form provided to the analyst along with the specimen (Annexure 4). The following details should be recorded on the worksheet:
 - Date of analysis
 - Condition of the specimen before starting analysis (should be entered in the laboratory notes)
 - Findings and result
 - Name and signature of the analyst (In case of electronically generated and maintained worksheets, appropriate control, validation and access procedures should be built in the system)
- Laboratory number assigned to the specimen should be mentioned in the worksheet before sending the specimen to the analyst.
- The specimen should be analyzed according to the plan mentioned in the SOP. Any deviations from the analysis plan should be mentioned giving reasons. Wherever applicable the laboratories can use requisition form cum worksheet (Annexure 5) instead of two separate forms. However, laboratories using electronic transmission of patients' details and test results may not require individual worksheets.

12.0 REPORTING TEST RESULTS

- Test results approved and signed by the designated authority should be made available to authorized person(s) only.
- Results should be reported clearly, without any errors, specifying measurement procedure where appropriate and units of measurement as recommended by professional societies such as International Council for Standardization in Haematology, International Society of Haematology & International Federation of Clinical Chemistry and Laboratory Medicine. A format for reporting results is given in Annexure 6.

13.0 SPECIMEN REJECTION RECORD

- Laboratories should maintain a record of specimens which were rejected prior to analysis. Rejection statistics (eg. number of hemolyzed specimen etc.) along with reason for rejection and person responsible for rejection should be maintained (Annexure 7).
- Specimen rejection statistics can be used by laboratories to identify the need and areas for staff training. For example, if specimen contamination is detected, the containers should be checked for sterility prior to collection as well as specimen handling procedure. This information should be shared with the medical, nursing and other staff engaged in specimen collection and transportation.

14.0 DATA MANAGEMENT

- Laboratory data management includes recording details of the patient, findings of analysis, reporting of results and archiving the data for future reference. Recording data allows smooth functioning of the internal quality control measures, internal audit and external quality assessment. From the point of view of management, **absence of record implies that the work was never done.**
- The format of recording and reporting results should be described in the Standard Operating Procedures (SOPs).
- Data entry should begin as soon as registration number is assigned to the specimen. Further entries should be made in the accession list and worksheet. The final report should be recorded after approval/signature of the designated authority.
- All auto analyzers should be connected with printer and uninterrupted power supply (UPS). If printing option is not available or semi auto analyzers are

used, laboratory should maintain manual raw test data counter-checked by two persons.

- The laboratory should maintain raw test data preferably for one month in non-editable format or signed printed copy.
- Procedure for adequate data protection and security including data editing and deleting should be developed and maintained by the laboratory. Authorization for amendment procedure should be specified in the SOP. The laboratory should also record reason for editing or deleting data.
- Facilities sending reports electronically should include electronic signature of the authorized signatory. Laboratories should be able to provide critical information required by a physician on telephone eg. frozen section biopsy report is required while operating a patient with suspicion of cancer or growth of a particular organism in culture can aid in early diagnosis and treatment.

15.0 STANDARD OPERATING PROCEDURE (SOP)

- SOP is a document, which contains detailed, written instructions describing the stepwise process and technique of performing a test or procedure in the laboratory.
- SOP helps to ensure uniformity, consistency and control over the processes carried out. It ensures that the procedures are done in exactly the same way each time irrespective of the operator.
- SOP should contain information on who can perform the test, their qualification and training, how to carry out the test including pre-analytical, analytical and post-analytical stages of test/procedure, laboratory conditions required for the test/ procedure, routine care and maintenance of equipment, precautions and safety instructions, trouble shooting measures, waste disposal and linkage with reference laboratories.
- SOP should be simple and written in an easy to understand language.
- The procedure described in the SOP must be followed exactly by all staff members to ensure high quality results.
- It should be titled along with version number, dated and signed by an authorized person and updated regularly.
- It is important for the SOP document to be readily available in the working area and is therefore also referred to as '**laboratory bench work manual**'.
- SOPs are **controlled documents** and can be changed only with approval of the laboratory quality manager and/or Head of the laboratory.

15.1 Format of SOP

The header of SOP should display the following information on all pages:

- Title of SOP and Document number
- Version number with dates of revision
- Issue number and date of issue of the document
- Page number/Number of pages

15.2 The text of SOP for test procedure should contain information on:

- Name of test
- Author's name and approving authority
- Scope of test
- Principle of the test
- Equipment and materials required
- Detailed test procedure including type, quantity and condition of specimen required; sample processing and preparation. Alternative procedure for test in case of breakdown of equipment should also be stated.
- Documentation of results including calculations
- Limit of detection (Analytical sensitivity)
- Analytical Measurement Range (AMR)
- Reference range
- Clinical significance, Inference and limitation of the test
- Critical alert values
- References of test procedure
- Precautions & Safety
- Quality Control procedures
- Specimen preservation and storage before analysis and after analysis
- Data management

15.3 Types of SOP include:

- Staff appointment, training, evaluation
- Maintenance of laboratory conditions including work space, lighting, ventilation, temperature regulation, noise control, designated eating and smoking area
- Cleaning, sterilization & disinfecting procedures
- Equipment care, operation, calibration, validation and maintenance of equipment
- Data Management
- Precautions & Safety measures including treatment if required and appropriate vaccination of staff
- Handling and disposal of waste including bio-wastes
- Documentation of laboratory's reference ranges (*In the absence of laboratory's own reference ranges, data generated officially for Indian subjects or failing that reference range on the manufacturer's guidelines contained in the kit brochure may be used*).
- Internal quality control procedures including procedure for reporting abnormal test results and corrective action procedure for quality control outliers
- Internal audit procedures
- Participation in external quality assessment programmes

16.0 SAFETY IN LABORATORIES

Personnel working in laboratories may be exposed to risks from various chemicals, infectious materials, fire hazard, gas leak etc. The environment is also at risk of being contaminated by hazardous materials used and wastes generated in the laboratory. Safety in laboratories therefore includes **protection of both the staff and the environment** from hazardous materials.

16.1 General Safety Measures

- Documentation of Laboratory Safety Policies and Procedures.
- All laboratory personnel should be aware about the laboratory safety policies and procedures and follow these at all times.

- List of hazardous materials used in the laboratory should be prepared. All hazardous materials should be accounted for on a continuous basis.
- Laboratory personnel should follow safe hygienic practices which include hand washing, wearing protective clothing, gloves, eye protection etc.
- Eye wash facility should be available as "stand-alone" facility or attached to sink. Portable, sealed, refillable bottles should also be available.
- Biohazard symbol should be used on all container/equipment containing biohazardous material.
- Laboratories should ensure proper preservation and security of specimens.
- Destruction/disposal of hazardous material should be authorized, supervised and handled according to standard procedures.
- Laboratory personnel should be thoroughly trained in managing fire, and non-fire emergencies such as large spillage, gas leakage etc.
- Adequate fire extinguishers should be readily available in the laboratory
- Periodic checking of all safety equipment and accessories should be ensured.
- Accident/incident/injuries record of laboratory personnel should be maintained and reported to the designated authority. The report should include description of the event, factors contributing to the event and information on first aid or other health care provided. This information can be analyzed periodically towards effectively controlling and preventing future events. The records should be checked periodically by the laboratory safety officer even in the absence of fresh entries.

16.2 Biosafety Precautions

- Laboratory personnel are at risk of exposure to a variety of infectious agents and need to observe special precautions for safe handling of pathogenic organisms in the laboratories.
- The World Health Organization (WHO) has classified pathogenic organisms into 4 risk groups - WHO Risk groups 1, 2, 3 and 4, based on the infectivity of an organism to cause disease in an individual and/or community (higher risk group indicates higher infectivity). Risk group classification takes into account the pathogenicity of the organism, mode of transmission, host factors such as immunity, hygiene etc., local availability of effective preventive and treatment measures.

- Four levels of biosafety laboratories (BSL) - 1, 2, 3 and 4, have been designed for handling biohazardous material. Usually higher level of biosafety is required while carrying out procedures using higher risk group organisms. However, certain procedures which generate high concentration of a low risk organism, may also necessitate the use of higher level of biosafety eg. generation of aerosols. Laboratory facilities such as equipment, containment facilities, ventilation, design etc. should be planned according to the risk group of organisms which the laboratory is handling and also the test procedures adopted.
- The management should facilitate the setting up of biosafety precautions in the laboratory by providing adequate manpower, resources, infrastructure and policy. A biosecurity and/or biosafety officer should be overall in-charge of biosecurity/biosafety in the laboratory.
- The laboratories should have their own biosafety manual written in consultation with the head of the laboratory. Policies should outline the use of sharps, disposal of bio-waste, reagents, sharps and other wastes generated in the laboratory in accordance with **Bio-Medical Waste (Management & Handling) Rules, 1998 (amended in 2000)**.
- All personnel working in the laboratory should be aware of these biosafety precautions and trained to follow them always.
- Laboratory staff should also realize that they may be inadvertently exposed to organisms of higher risk groups and therefore must observe safety precautions laid down by the laboratory at all times.
- It is desirable to train all staff in good microbiological techniques (GMT) wherever applicable.
- Laboratory staff cleared for working in Level-3 and Level-4 laboratories should be specially trained and their clearances updated at regular intervals (e.g. annually).

16.3 Levels of Biosafety Laboratories (BSL)

- **BSL 1** can handle biological materials with **minimum biohazard** to the laboratory personnel and environment eg. laboratories at PHC level, side laboratories in labour rooms or wards. It is considered a **cold zone**. Access to the laboratory should be limited to laboratory personnel and the staff should use personal protection such as gowns, gloves, eye protection eg. glasses, footwear, use a separate area for hand washing, storing food, drinks, etc. The laboratory work can be carried out on open bench tops and the surface should

be decontaminated as per the safety requirements, arthropod and rodent control measures should be followed, mouth pipetting should be replaced with mechanical procedure and techniques which minimize splashes and aerosol formation.

- **BSL 2** laboratories are equipped with facilities to handle biomaterial which pose **moderate hazard** in the event of injury to skin or exposure to mucous membrane or ingestion. It is also considered a **cold zone**. All diagnostic and healthcare laboratories in public or private sector should have BSL-2 facilities. In addition to BSL-1 precautions, a biohazard warning sign should be displayed at the entrance. Special care should be exercised while handling sharps and an autoclave should be available for decontamination. Biohazardous material should be handled in class I or II biological safety cabinets (BSC) within the laboratory. Mechanical circulation of air with inward flow is preferred. Chemical, fire, electrical safety measures should be followed. Eyewash station should be conveniently located. All laboratory personnel should undergo pre-employment health check-up and a record of their illnesses and immunization received should be available to the laboratory managers.
- **BSL 3** laboratories are located in teaching and/or research institutions, production and clinical testing facilities. They can handle pathogenic agents which can cause **potentially lethal** disease when inhaled. They are containment laboratories and are considered a **warm or neutral zone**. Access to the laboratory should be determined by the laboratory in-charge. Higher degrees of personal protection of the laboratory staff is needed including respiratory protection in certain instances. The staff should undergo baseline and periodic serum testing for the agent being handled in the laboratory. Laboratory design in addition to BSL-2 facilities should have a ducted exhaust air ventilation system installed with negative airflow into the laboratory. Heating, ventilation, air-conditioning (HVAC) control systems may be installed to avoid positive pressure in the laboratory. High-efficiency particulate air (HEPA) filtration should be used to re-circulate air into the laboratory. Handling of biohazardous material should be done in BSC class I, II or III. Laboratory staff should be trained to work in BSL-3 laboratories.
- **BSL 4** laboratories or maximum containment laboratories are suitable for handling dangerous and exotic agents with high risk of **life threatening disease** associated with aerosol transmission. These laboratories should be located in isolated areas. All biosafety precautions for BSL-3 laboratories should be followed with additional safety practices including complete change of clothing and shoes prior to entering and exiting from the laboratory. **No individual should work alone: two-person rule should be followed.** Staff

should be trained to handle personnel injury or illness. All work should be conducted in class III BSC or class II BSC with personal one-piece positive pressure suits fitted with ventilatory support. Ventilation system should be non re-circulating with unidirectional airflow from the area of least hazard to area(s) of greatest potential hazard. HVAC control systems should be installed to monitor airflow in the supply and exhaust systems. The facilities should be negatively pressurized to prevent contamination if airflow system fails. High Security Animal Disease laboratory (HSADL) is India's only BSL-4 laboratory at Bhopal. It conducts research on all kinds of zoonotic diseases and emerging infectious disease threats.

17.0 ETHICAL CONSIDERATIONS

Personnel working in clinical and/or research laboratories or engaged in biomedical sciences or research should be aware of their ethical responsibilities and comply with the ethical code of conduct which are governed by the following principles :

17.1 Principle Of Non-Maleficence, whereby it is ensured that activities, discoveries or knowledge of personnel engaged in biomedical sciences 'do no harm' by -

- i) refraining to engage in any activity or research that is intended or likely to cause harm to plants, animals, humans or environment.
- ii) refraining from contributing to the development, production or acquisition of microbial or other biological agents or toxins, whatever their origin or method of production, of types and/or in quantities that have no justification for prophylactic, protective, therapeutic, or other peaceful purposes.

17.2 Principle Of Beneficence, whereby it is ensured that legitimate benefits are being sought and that they out-weigh the risks and harms. The personnel should work for the ethical and beneficial advancement, development and use of scientific knowledge.

17.3 Principle Of Institutional Arrangement, whereby reasonable care is taken to ensure that all procedures are required to be complied and all institutional arrangements are required to be made to assure bio-safety and security. Access is allowed to biological and other chemical agents that could cause harm, only to bonafide scientists in a transparent manner who, there are reasonable grounds to believe, will not misuse them. Appropriate committees to oversee the activities to be put in place.

17.4 Principle Of Risk Minimization, whereby due care and caution is taken to provide all bio-safety precautions and restrict the dissemination of dual use of information and knowledge where there are reasonable grounds to believe that there are serious risks that information or knowledge could be readily misused to inflict

serious harm. Bring to the attention of the appropriate persons/authorities, activities including unethical research, that there are reasonable grounds to believe are likely to contribute to harm.

17.5 Principle Of Ethical Review, whereby research activities are subjected to ethics and safety reviews and monitoring through appropriately constituted committees to establish their ethical acceptability. If human or animal participants are involved, to ensure that such involvement is ethical and essential for carrying out highly important research by following the relevant national and international guidelines.

17.6 Principle Of Transmission Of Ethical Values, whereby (the duties and obligations embodied in this code) the ethical principles upon which it is based are transmitted faithfully to all who are, or may become, engaged in the conduct of biomedical activities or research.

17.7 Principle Of Voluntariness, whereby researchers are fully apprised of the research and the impact and risk of such research, and whereby scientists retain the right to abstain from further participation in research that they consider ethically or morally objectionable.

17.8 Principle Of Compliance, whereby personnel engaged in biomedical activities and research abide by laws and regulations that apply to the conduct of science, duties, and obligations embodied in this code, and disseminate the same to all concerned.

18.0 QUALITY ASSURANCE

Quality Assurance (QA) is the total process whereby the quality of laboratory reports can be guaranteed. Incorrect Laboratory results may be due to errors occurring during specimen collection (pre-analytical stage), testing (analytical stage) and/or while reporting and interpreting (post-analytical stage) test results. Whereas, Internal Quality Control (IQC) refers to the process of minimizing analytical errors, QA encompasses procedures adopted for minimizing errors that may occur at any stage. Provision of precise and accurate laboratory results optimize medical management. Inappropriate test selection, unnecessary investigations and incorrect test results not only have serious health implications but are also a financial burden to the individual and community. Reports from a laboratory with a high level of QA will help the physician to arrive rapidly at a correct diagnosis. To provide QA, all laboratories must have a Quality Assurance Programme (QAP) in place.

19.0 QUALITY ASSURANCE PROGRAMME (QAP)

- QAP is a managerial process of maintaining high standards of performance and of improving standards where necessary. The concept is best illustrated

by Deming's cycle of Planning (P), Doing (D), Checking (C), and Acting (A). If any of the four components of the cycle lag behind then the quality declines. While planning a QAP it is important to put effort at each step to prevent, detect and correct errors.

- Quality Manager or designee or competent authorized person should review the quality control data and maintain record of evaluation.
- The two important tools toward maintaining laboratory quality are
 - Internal Quality Control (IQC) - for detection and minimization of immediate errors
 - External Quality Assessment (EQA) - for monitoring long term precision and accuracy of results.
- **The laboratory should treat IQC/EQA samples and patients' specimens alike and use same procedures for analysis.**

20.0 INTERNAL QUALITY CONTROL

20.1 Practice of IQC

This includes the following:

- Recognition of errors which arise within the laboratory during analytical stage (testing).
 - Taking steps to minimize errors.
 - Equipment & method calibration, method validation (Annexure 8).
- Laboratories should perform IQC
 - Every day on tests run daily
 - Every time the tests are run in case of infrequently used tests.
- Quality control checks should be employed for both quantitative and qualitative tests.

20.2 IQC for Quantitative Tests

- Levy Jennings's (LJ) chart should be used to plot daily QC values (Annexure 9). It indicates the changes in trends and shifts of the laboratory performance. Westgard rules should be used to interpret daily QC values. The level of QC applied in the laboratory varies according to the number of specimens analyzed

per day. The following protocol may be adopted by the laboratories according to the total number of specimens analysed per analyte :

- Less than 40 per day - apply at least one level QC once a day.
- Between 40-80 per day - apply two level QC at least once a day.
- More than 80 per day - apply two level QC at least twice a day for such analytes.
- For haematology, 2 level QC (using normal & high **OR** normal & low controls) should be analyzed at least once a day although it is preferable to run 3 level QC (using normal, high & low controls) once a day.
- The following guidelines will be useful to the laboratories in the practice of IQC using either one level or two level QC materials.

When one level QC is used :

Reject test run if following errors occur:

- Value is outside 3 SD (1_{3s})
- 2 consecutive values are outside 2 SD on the same side, but within 3 SD (2_{2s})
- 4 consecutive value are outside 1SD on the same side, but within 2SD (4_{1s})
- 10 consecutive values are above or below the mean, but within 2 SD (10_x)

When two level QC are used :

Reject test run if following errors occur:

- Either QC value is outside 3 SD (1_{3s})
- Both QC values are outside 2 SD on the same side, but within 3SD (2_{2s})
- Difference between the two level QC values is >4 SD i.e. one level QC is >2 SD and other level QC is < 2 SD (R_{4s})
- 10 consecutive values of the same level QC are above or below the mean, but within 2 SD (10_x)
- 5 consecutive values of one level QC and 5 consecutive values of the other level QC are above or below the mean, but within 2 SD (10_x)
- **Laboratories need to establish guidelines for responding to out-of-control situations (refer to Annexure 10 for corrective actions).**

Note: The laboratory personnel performing the test should determine the appropriate corrective action to be taken for QC data that fall outside the established tolerance limits. Corrective action should be documented with the technician's initials and date.

- Tests for which control material is not available or when running of control is not viable due to low volume of tests, the laboratory should apply alternate quality control techniques such as :
 - Retesting of any randomly chosen specimen/s
 - Replicate test of specimen by different method, different machine and different person, wherever applicable
 - Correlation of test results with other parameters

20.3 IQC for Qualitative Tests

For qualitative tests, positive and negative controls should be included with each run. For staining procedures, gram stains require both Gram positive and Gram negative control organisms to be used once per week. QC should also be run whenever a new lot of the stain procedure kit is used and/or any of the four components of the stain procedure kit is replaced with a new lot.

21.0 EXTERNAL QUALITY ASSESSMENT

- External Quality Assessment (EQA) refers to a system in which the performance of a laboratory is assessed PERIODICALLY AND RETROSPECTIVELY by an independent external agency to indicate to the laboratory staff of any shortcomings in performance. It indicates a need for improving and/or changing IQC procedures. An organizing agency or laboratory sends identical specimens to all participating laboratories for testing by methods routinely adopted by them. The results from the participating laboratories are received by the organizing agency and compared with a "correct" answer retrospectively and a performance score is assigned. All the participating laboratories are identified by a code and reports issued to the participants contain the performance score of all participating laboratories including its own score. This allows a comparison of quality between laboratories and thus describes the "state of the art" for that area of laboratory work encompassed by EQA programmes (Annexure 11).
- In India, participation in EQA schemes is a pre-requisite for clinical laboratories applying for NABL accreditation but is not required for obtaining license. However, in some countries participation in EQA is mandatory for licensure.
- It is desirable that all laboratories take part in one or more EQA programmes. If EQA for a particular analyte is not available, an appropriate inter-laboratory comparison is recommended.

- Benefits of EQA
 - Assesses the overall performance of laboratory
 - Establishes inter-laboratory comparison
 - Serves as an early warning system for problems
 - Identifies systematic kit problems
 - Provides objective evidence of laboratory quality
 - Indicates areas towards which efforts need to be directed for improvement of quality of results
 - Identifies training needs

22.0 INTERNAL AUDIT

Audit is a process of critical review of the functioning and evaluation of services. Internal audit is the systematic, independent and documented process for obtaining audit evidence and evaluating it objectively to determine the extent to which the specific criteria are complied with. Internal audit can be effectively carried out by examining documents, specimens, equipment, environmental conditions, examination procedures and personnel competence. Effective internal audit will identify the problems and weak points in the system and suggest remedial measures.

23.0 SUMMARY OF QAP ACTIVITIES

- Establish QC Policy & QC Procedure
- Secure QC material supply for several months, preferably for one year with same lot number for both normal & abnormal QC
- Construct LJ charts and plot daily QC values
- Scan LJ charts for trends and shifts
- Define "out-of control limits" and corrective actions
- Participate in EQA programmes
- Evaluate IQC & EQA reports once a month towards method / analyzer modifications
- Evaluate the whole QC programme once a year for its effectiveness, cost and areas needing attention

BIBLIOGRAPHY

- Biosafety in Microbiologic and Biomedical Laboratories. 5th ed. Washington: U. S. Government Printing Office; 2007. Available from http://www.cdc.gov/OD/OHS/biosfty/bmbl5/BMBL_5th_Edition.pdf [Accessed on 17 July 2008]
- Bio-Medical Waste (Management and Handling) Rules, 1998 (Ammended in 2000) Available from http://www.searo.who.int/LinkFiles/SDE_SDE_mgmt-bio-medical-framework.pdf [accessed on 07 July 2008].
- Ethical Guidelines for Biomedical Research on Human Participants. New Delhi, Indian Council of Medical Research; 2006. Also available from http://icmr.nic.in/ethical_guidelines.pdf [Accessed on 17 July 2008]
- Ethical Guidelines for Biomedical Research on Human Subjects. New Delhi, Indian Council of Medical Research; 2000.
- Geethanjalai FS, Fleming J, Swaminathan S, Selvakumar R. External quality assurance - role of ACBI /CMC scheme. Indian J of Clin Biochem. 2006; 21 (1): 211-212.
- Good Clinical Practice for Clinical Research in India, Central Drugs Standard Control Organization, Dte.GHS, Ministry of Health and Family Welfare, Government of India Available from <http://cdsco.nic.in/html/GCP1.html> [accessed on 15 May 2008].
- Health Laboratory Services in Support of Primary Health Care in South-East Asia. 2nd ed. New Delhi: WHO Regional Publications, SEARO Series, No 24; 1999.
- ICH Harmonised Tripartite Guideline - Guideline For Good Clinical Practice E6(R1) Current Step 4 version, dated 10 June 1996. Available from <http://www.ich.org/LOB/media/MEDIA482.pdf> [accessed on 17 July 2008].
- Indrayan A, Sarmukaddam SB. Medical Biostatistics. New York: Mercel Dekker Inc; 2001.
- Kanagasabapathy AS, Rao P. Laboratory accreditation-procedural guidelines. Indian J of Clin Biochem. 2005; 20(2): 186-188.
- Kumari S, Bhatia R. Quality Assurance In Bacteriology and Immunology. 2nd ed. New Delhi: WHO Regional Publication, South-East Asia Series No. 28; 2003.
- Laboratory Biosafety Manual, 3rd ed. Geneva : WHO Publication; 2004.

- National Good Laboratory Practice Compliance Monitoring Authority. New Delhi: Department of Science & Technology, Government of India; Available from <http://indiaglp.gov.in/> [accessed on 15 May 2008].
- OECD series on Principles of Good Laboratory Practice and Compliance Monitoring. Paris: Environment Directorate, Organization For Economic Co-Operation And Development:1998. Available form [http://www.olis.oecd.org/olis/1998doc.nsf/linkto/nt00000c5a/\\$file/01e88455.pdf](http://www.olis.oecd.org/olis/1998doc.nsf/linkto/nt00000c5a/$file/01e88455.pdf) [Accessed on 17 July 2008].
- Specific Criteria for Accreditation of Medical Laboratories. NABL Document 112, Issue No. 03, Version 2007. Available from <http://www.nabl-india.org/nabl/asp/users/documentMgmt.asp?cp=2&docType=both> [accessed on 17 July 2008].

Annexure 1. Equipment Log Book

Name & Model of Equipment **Emergency Telph No.**

Date of Installation **Warranty period**

In-house maintenance schedule	Maintenance done on	Company maintenance schedule as per AMC	Company maintenance done with date & time	Problems encountered with date & time	Rectification details with date & time	Parts repaired, replaced with date & time	Remarks, signature

Annexure 2. Requisition Form

Name Age Sex OPD No. /Ward-Bed No.....

Consultant In-Charge Date of specimen collection

Type of specimen and source

Tests requested.....

.....

Provisional diagnosis

Relevant Clinical History including drug treatment

.....

.....

.....

.....

.....

Signed by

Annexure 3. Accession List

Lab.No	Name	Age/Sex	OPD/Wd Bed No.	Referring doctor	Invest. required	Date & time of specimen receipt	Condition of specimen at receipt	Result	Remarks

Annexure 5. Requisition Form cum Worksheet

Lab No Name.....AgeSex

OPD No./Ward-Bed No.

Consultant In-Charge Date of specimen collection

Type of specimen and source

Tests requested

.....

.....

Provisional diagnosis

Relevant Clinical History including drug treatment

.....

.....

.....

Laboratory Notes.....

.....

.....

.....

Result

.....

.....

Date of reporting Reported by

Signature

Annexure 6. Reporting format

Lab No		Name		Age		Sex	
OPD No./Ward-Bed No.				Consultant In-Charge			
Date of specimen collection				Date & time of specimen receipt			
Date & time of release of result							
Method used							
Test Result				Normal Value			
.....							
.....							
.....							
.....							
.....							
.....							
.....							
.....							
.....							
Date of reporting				Reported by			
Signature							

Annexure 7. Specimen Rejection Record

S.No	Lab No.	Type of specimen	Reason for rejection	Remarks	Signature

Annexure 8. Steps to Initiate IQC in Laboratories

Check Calibration of the Equipment : Most equipment are factory calibrated and require infrequent recalibration. Recalibration should be performed whenever the equipment undergoes a major repair.

Calibration of Equipment : It is a process which is applied to quantitative measuring or metering of equipment to assure its accurate operation throughout its measuring limits. Calibration of equipment refers to hardware calibration which will be performed by the company engineer and subsequently will be validated by checking QC/reference material. Equipment calibration must include technical aspects such as checking of optical systems, temperature, pipette probe, voltage etc. The company engineer should provide a calibration certificate with relevant details. Correct QC/reference material values indicate that the calibration of the equipment is according to the prescribed standards. If the results are beyond the acceptable limits, the source of error should be identified and the equipment recalibrated.

Validation of test : Analysis of a specimen in a laboratory involves a number of stepwise procedures using different equipments, processes, in-vitro devices, software etc. The “correctness” of results depends upon the correct functioning of all procedures involved. It is therefore essential to validate the equipment, processes, in-vitro devices, software etc. ISO 9001 defines validation as “the attaining and documenting of sufficient evidence to give reasonable assurance, given the current state of science and the art of manufacture, that the process, system and test method under consideration consistently does and/or will do what it is expected to do”.

Validation must be done just after calibration of the system with the use of appropriate control material. This provides reassurance that the system and operator are working correctly. Thereafter, validation is done periodically according to user requirement and as written in the SOP manual. The responsibility of validation usually rests with the Head of Department.

Validation gives information on:

- chemical and instrumental drift
- assay stability

Performance characteristics with reference to validation are:

- Accuracy
- Precision
- Efficiency
- Linearity

Accuracy

Closeness of agreement between true value and the mean of observed value obtained over large number of observations. This can be quantitatively expressed as **Bias**

$$\text{Bias} = \frac{\text{Observed value} - \text{True value}}{\text{True value}} \times 100$$

Good accuracy means minimum Bias.

Precision

Precision is closeness of results with each other of a large number of successive observations in a measurement process, under prescribed conditions. This can be quantitatively expressed as percentage of coefficient of variation (% CV). Good precision means minimum % CV. It can be calculated using the formula given below:

$$\%CV = \frac{SD}{\text{Mean}} \times 100$$

IQC also assesses whether the performance of the laboratory is sufficiently similar to its own previous performance. It thus provides a continuity of patient care over a period of time. Precision can be demonstrated through repeatability and reproducibility.

(a) Repeatability : Closeness of the agreement between the successive measurements of the same specimen and with the following conditions:

- The same measurement procedure
- The same analyst
- The same measurement systems used under the same conditions
- The same location

(b) Reproducibility : Closeness of the agreement between the results of successive measurements of the same specimen and with the following conditions:

- The same measurement procedure
- Different analysts
- Different measuring systems
- Different locations and at different times

Efficiency of test - is the ability of a test to give the correct diagnosis of a disease/condition. The clinician requesting for a particular test must keep this in mind when interpreting the results of tests with low efficiency. This is measured by two criteria:

- a) **Diagnostic sensitivity** : The proportion of subjects with disease who have positive test results. The greater the sensitivity of a test, the fewer the number of false-negative results.

$$\text{Sensitivity} = \frac{\text{True Positives}}{\text{True Positives} + \text{False Negatives}}$$

- b) **Diagnostic specificity** : The proportion of subjects without disease who have negative test results. The ability of a test to give fewer false positive results.

$$\text{Specificity} = \frac{\text{True Negatives}}{\text{False Positives} + \text{True Negatives}}$$

Linearity - It determines the upper and lower limit of reporting range of a test.

Annexure 9. Calculation of Mean, Standard Deviation and Tolerance Range

Each laboratory must set up quality controls for different tests before beginning to analyze specimens of patients. To calculate these values appropriate controls whose value or concentration is known are analyzed under standard conditions. The controls should be stable and of reproducible composition. Analysis of control samples is done in 20 batches (one batch per day). From the results, mean value (assigned or expected value) and standard deviation is calculated using statistical formula.

Mean (x) is calculated by adding up the results obtained and dividing by the number of observations (n) taken.

$$\text{Mean (x)} = \frac{x_1 + x_2 + \dots + x_n}{n}$$

Deviation from mean is the difference between the observation and the mean value. It is calculated for each observation.

$$\text{Deviation from mean} = [x_1 - x] ; [x_2 - x] ; \dots [x_n - x]$$

Mean deviation is average of the deviation from mean of all the observations taken. This can be calculated by adding the deviation from mean for all observations and dividing by the total number of observations.

$$\text{Mean deviation} = \frac{[x_1 - x] + [x_2 - x] + \dots + [x_n - x]}{n} \text{ or } \sum \frac{(x_i - x)}{n}$$

where $i = 1, 2, \dots, n$

As mean deviation can be either positive or negative, to get rid of the minus sign, the deviations are squared up and divided by the number of observations. This gives

$$\text{variance of a measurement. Variance} = \sum \frac{(x_i - x)^2}{n}$$

Standard deviation: The square root of variance is known as the standard deviation (SD)

In case of laboratory tests where the number of observations is less as compared to population surveys, the denominator 'n' is replaced by (n-1) to give better results.

$$\text{SD} = \sqrt{\sum \frac{(x_i - x)^2}{n - 1}}$$

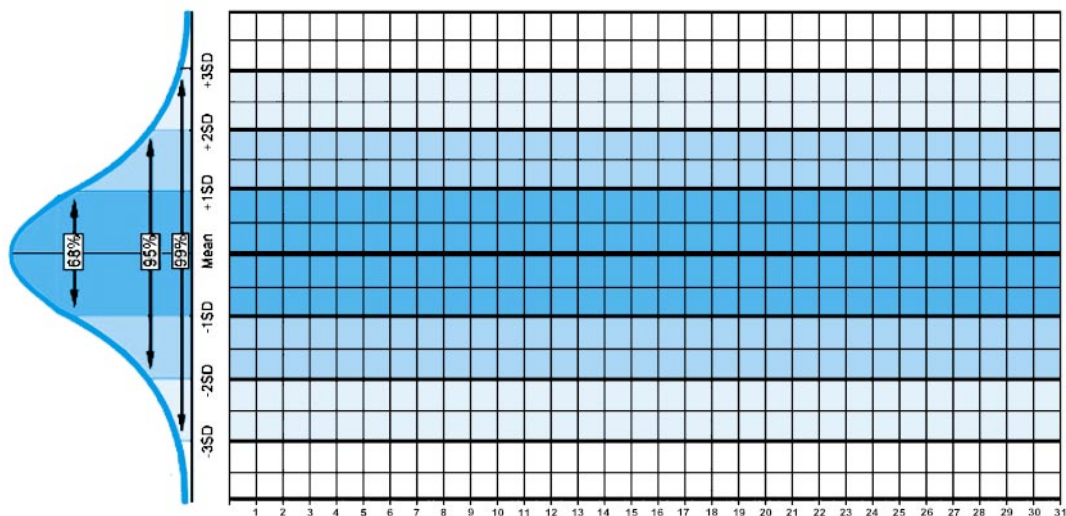


Figure 1. Levy Jennings Control Chart showing mean value, SD, tolerance range

The results are plotted on a Levy Jennings (LJ) Control chart (Fig 1.), with test result on the 'Y' axis and batch number (usually days) on the 'X' axis. The quality control samples demonstrate a Gaussian distribution of results. The **tolerance range** is taken as any value which falls between mean - 2SD to mean + 2SD. Horizontal lines are drawn corresponding to the mean (expected) value at + and -2 SD and at + and - 3 SD values. Accordingly 68.3% of values are within +/-1 SD of the target value, 95.5% are within +/- 2 SD and 99.7% are within +/- 3SD (Figure 1). Westgard rules are applied in interpreting daily QC values.

Manual LJ Chart: When automated counters are not available, quality control charts can be prepared manually. For this, the control is run each day for 10-15 days using the routine methods of the laboratory as carefully as possible. Their mean and standard deviations are calculated and LJ charts prepared with the mean being in the centre and lines drawn at $\pm 1, 2$ and 3 SD. Subsequently, a control sample is analysed daily or on each batch of tests alongside the patients' specimens. LJ chart is plotted using day (or batch) on the 'X' axis and the value obtained on the 'Y' axis. Its position on the graph gives an indication of precision of the test.

Quality Control through Duplicate Testing: Laboratories which do not have a high workload can adopt duplicate testing of all specimens for ensuring quality control. Standard deviation of the test is calculated which is used as a marker for quality control.

The Standard deviation is calculated as follows:

- Test 10 consecutive specimens in duplicate
- Derive SD using the formula $SD = \frac{\sqrt{\sum \text{of } d^2}}{2n}$

where

d= difference between duplicates

n= number of pairs

The duplicate test is performed on every 10th or 20th specimen. Results are interpreted as follows:-

- Difference between the paired results <2SD : results are acceptable
- Difference is between the paired results > 2SD, consider :
 - Random alteration of reagents
 - Alteration in performance

IQC in Haematology

In large hospitals where a large number of blood counts are performed each day using automated cell counters, day-to-day or week-to-week variability in the mean of red cell indices [Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC)] may be used for quality control. When specimens are evenly distributed there should be no significant change in the day-to-day mean value of red cell indices. Any significant change may indicate a change in instrument calibration or a fault in its function. However, results are valid only if the population from whom these specimens are received are from the general population and not from specific outpatient clinics for example thalassemia clinic held on specific days of the week. Results from these clinics are best excluded for calculation of mean red cell indices.

Sophisticated automated cell counters have a computer program incorporated in the system which makes it possible to analyze the mean red cell indices continuously. Alternatively, a programmable calculator or personal computer may be used to analyze results. Laboratories which do not have computer facilities can use manual methods. For this, only the mean MCHC is calculated daily on all the measurements obtained during the course of the day. Mean MCHC value of 11 consecutive working days is used to calculate the SD and a graph is drawn to record daily mean values. Any significant alteration in the daily mean MCHC indicates that a fault has occurred. The component indices Haemoglobin (Hb%) and Packed Cell Volume (PCV) are controlled indirectly by this procedure.

Annexure 10. Corrective Actions

Laboratories should establish guidelines for responding to out-of control situations. Carefully planned and properly implemented IQC minimizes false rejections. The best response to an out-of control situation is to identify the cause of the problem, find appropriate solution that will eliminate the cause, and prevent future occurrences. When it is clear that QC on a particular day is out-of control, the first action to be taken is to withhold patients' results in that batch. Then, the technician should start checking the simplest and most frequent faults and continue further as necessary in a logical order depending on the method and the equipment involved.

It is essential to determine the type of error (random or systematic) in order to specifically correct the problem.

- (i) Random errors (affect precision) – Usually 1_{3s} and R_{4s} errors
- (ii) Systematic errors (affect accuracy) – Usually 2_{2s} , 4_{1s} and 10_x errors

Proposed actions:

It is a good practice to start by excluding gross errors such as mix-up of control materials, reagents, pipettes and failure to follow instructions for analysis. Step wise action may be taken as suggested below:

- Repeat QC from the same vial/fresh vial taken out from the freezer/from a freshly reconstituted QC lot
- Check QC storage conditions
- Check reagents-expiry date/contamination
- Check calibrator storage conditions
- Recalibrate
- Check equipment operation – follow manufacturer's trouble shooting instructions and if necessary call equipment engineer
- Relate causes to any recent changes

Annexure 11. Basics of EQA

EQA is a systematic, time bound ongoing activity in which the roles and responsibilities of the organizing agency (OA) and participating laboratories (PLs) are well defined. The OA consists of a panel of scientists or experts with the knowledge about the processes or tests that are being evaluated. They are assisted by statistical experts, managerial and administrative staff. The participating laboratories may be classified as small, medium or large depending upon the number of tests performed in a year. The participation of laboratories is voluntary and anonymous.

- The OA prepares a list of PLs and assigns a code to each laboratory to ensure confidentiality.
- The test material is homogenous so that all PLs receive a similar material. The material should be stable to allow adequate time for transportation. It is also known as "reference material".
- The frequency of test material distribution depends on several feasibility factors and is planned prior to initiating the process. It is important to intimate the report of the previous EQA to the PLs before starting the next round. Most agencies conduct EQAs between once in a fortnight to once in four months represented in a graphical form e.g. histogram forms. The test results of the PLs should be displayed in the report such that each laboratory can identify and check the correctness of data entry by OA. The statistical methods used for EQA should be clearly understood by the PLs. If more than one method is used for analysis of reference material then results should also be classified according to the different methods used.
- The OA should be in communication with PLs through periodic meetings, newsletters and annual reports. Poorly performing laboratories should be offered help and advice. PLs should be able to refer matters such as error in assessment back to the OA.
- PLs should be encouraged to view the process of EQA as 'their own' and use the opportunity as a learning experience to improve their performance. It is important that the PLs present true picture of the methods used and results obtained.
- The PLs should use the analytical method of their choice and it should be the same used in their routine analytical work.
- The PLs should communicate the results to the OA in a manner which is similar to the one used for the patients.

- The results received by the OA are entered into the computer data base and statistical calculations are done to arrive at performance scores of the various PLs. Commonly used performance scores are 'Z' scores and 'Q' scores.
- The OA issues the report of EQA to the PLs. The report should include the performance score of all PLs in a clear, comprehensive format such that inter-laboratory comparisons can be made. The PLs should plot the EQA performance scores regularly in a graph and display it prominently in the laboratory to motivate staff to perform better.

Agencies Organizing EQA

EQA can be organized by international, national and regional agencies or laboratories. The WHO has established international external quality assessment for its member countries. The Association of Clinical Biochemists of India (ACBI) along with CMC Vellore first started EQA in Clinical Biochemistry in 1978 with 50 participating laboratories. Many private agencies also organize EQA for different types of medical laboratories.

EQA schemes in the following sections are available in India:

- Haemogram - Indian Society of Haematology and Transfusion Medicine (ISHTM); Randox-RIQAS
- Coagulation - ISHTM/CMC, Vellore; Randox-RIQAS
- Clinical chemistry - Association of Clinical Biochemists of India (ACBI), CMC, Vellore; College of American Pathologists; Biorad; Randox- RIQAS; Royal College of Pathologists of Australia(RCPA- QAP); UK National External Quality Assessment scheme
- Microbiology - Indian Association of Medical Microbiologists- CMC, Vellore
- Virology- NACO- CMC, Vellore

Annexure 12. Understanding Accreditation of Clinical Laboratories

Accreditation is the formal recognition of a laboratory by a competent authority to provide credibility to the tasks carried out in the laboratory. It assures the clinician and the patients that the test reports are reliable and also gives a feedback to the laboratory on its performance vis-à-vis international standards. NABL works in accordance with ISO/IEC 17011 and is a signatory to International Laboratory Accreditation Cooperation (ILAC) as well as Asia Pacific Laboratory Accreditation Cooperation (APLAC) mutual recognition arrangements (MRA). This provides international recognition to NABL credited laboratories. NABL assesses the medical laboratories according to ISO 15189:2007 standards. NABL has published "Specific Criteria for Accreditation of Medical Laboratories" which is useful in understanding the process of accreditation.

A laboratory may apply for accreditation of one or more tests which are performed in accordance with NABL criteria. In India, accreditation is not an essential criterion for licensing.

Benefits of Accreditation

- Increases confidence and satisfaction of clinicians and patients in the services provided.
- Promotes business development both nationally and internationally.
- Improvement of operations and services of laboratories in accordance with NABL guidelines
- Reduction in cost and time by eliminating the need for re-testing

Accreditation Process

1. Preparation : The laboratory should be set up in accordance to the requirements of NABL. The relevant documents on procedures must be procured from NABL. The laboratory should also have in place an Internal Quality Control system.
2. Apply : The laboratory has to submit an application to NABL requesting for accreditation along with supporting documents.
3. Pre-assessment Audit : The NABL secretariat organizes the pre-assessment audit at the site to evaluate the preparedness of the laboratory for accreditation. A report of the audit is prepared and a copy is given to the applicant laboratory to help prepare for final assessment.
4. The applicant laboratory takes corrective action on the report and submits a Corrective Action Report to NABL.

5. The final assessment is then organized by NABL. The assessing team consists of a Lead Assessor and technical experts who assess the laboratory. The assessment includes evaluation of procedures, techniques, equipment, recording & reporting of data, staffing, training, SOP manuals, IQC measures and management of laboratory.
6. The final assessment report indicates non-conformance issues if any, and the laboratory is required to take corrective action within 3 months and submit a report.
7. After satisfactory corrective actions, accreditation is granted to the laboratory for a period of 2 years.
8. The whole process takes 6-8 months.
9. Laboratories can appeal to NABL against the assessment report if required.
10. NABL conducts annual surveillance of the accredited laboratory.

Advisory Committee Members

Chairperson

Lt. Gen (Retd.) D. Raghunath
Principal Executive
Sir Dorabji Tata Centre for Research in Tropical Diseases
Innovation Centre, Indian Institute of Science Campus,
Bangalore - 560012

Members (in alphabetical order)

Dr A.S. Kanagasabapathy
(Formerly Professor & Head of Clinical Biochemistry, CMC Hospital, Vellore)
Consultant Clinical Biochemist
Kamineni Hospitals,
Hyderabad - 500068

Dr B.S. Ramakrishna
Director
Dept. of Gastrointestinal Sciences
Christian Medical College
Vellore-63200

Dr G. Balakrish Nair
Scientist 'G' & Director
National Institute of Cholera & Enteric Diseases
P-33, CIT Road XM, Scheme Beliaghata
Kolkata-700 010

Dr R. Ravi
Scientist-F
Vector Control Research Center
Medical Complex
Puducherry – 605006

Dr R. Selvakumar
Professor & Head
Dept. of Clinical Biochemistry
Christian Medical College
Vellore - 632004

Mr Sambhu Chakraborty
Quality Management Consultant
Joyaboti
Flat No HF-2/16, 4/P-11
Ho Chi Minh Sarani, Shakuntala Park
Kolkata - 700 061

Dr Shalini Singh
Scientist 'D'
Division of RHN
Indian Council of Medical Research
New Delhi -110029

Dr S. K. Bhattacharya
Additional Director General
Indian Council of Medical Research
New Delhi -110029

Dr T. Ramamurthy
Scientist 'E'
National Institute of Cholera & Enteric Diseases
P-33, CIT Road XM, Scheme Beliaghata
Kolkata -700 010

Dr Vasantha Muthuswamy
Scientist 'G' & Head
Divisions of RHN & BMS
Indian Council of Medical Research
New Delhi -110029

Drafting Committee Members

Dr Shalini Singh
Scientist 'D'
Division of RHN
Indian Council of Medical Research
New Delhi -110029

Prof (Dr) A.S. Kanagasabapathy
(Formerly Head of Clinical Biochemistry, CMC Hospital, Vellore)
Consultant Clinical Biochemist
Kamineni Hospitals,
Hyderabad – 500068

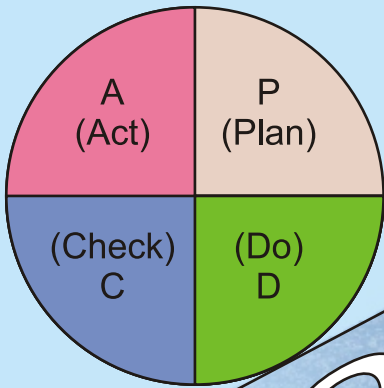
Mr Sambhu Chakraborty
Quality Management Consultant
Joyaboti
Flat No HF-2/16, 4/P-11
Ho Chi Minh Sarani, Shakuntala Park
Kolkata - 700 061

Reviewers

Dr Arti Kapil
Addl Professor
Department of Microbiology
All India Institute of Medical Sciences
New Delhi – 110029

Dr Renu Saxena
Professor & Head,
Department of Haematology
Institute Rotary Cancer Hospital
All India Institute of Medical Sciences
New Delhi – 110029

Prof (Dr) Subir Kumar Dutta
(Formerly Head of Dept of Pathology, University College of Medicine, Kolkata)
Consultant Pathologist
Scientific Clinical Research Lab (P) Ltd
2 Ram Chandra Das Row
Kolkata – 700013



QUALITY

DEMING'S CYCLE



INDIAN COUNCIL OF MEDICAL RESEARCH

V. Ramalingaswami Bhawan
Ansari Nagar, New Delhi-110 029
Phone : 91-11-26588980, 26589794, 25688895
E-mail : icmrqds@sansad.nic.in
Website : <http://www.icmr.nic.in>