Supplementary Requirements for Accreditation of Biological Testing Laboratories
Foreword

This Philippine Accreditation Bureau (PAB) Supplementary Requirements for Accreditation of Biological Testing Laboratories was developed by the Laboratory Accreditation Technical Committee (LATC) – Biological Testing to supplement ISO/IEC 17025 requirements by providing specific technical criteria and guidelines for both assessors and for laboratories carrying out biological testing.
1. Introduction

All applicant Biological Testing Laboratories are required to meet this supplementary requirements in addition to the general requirements for the competence of testing and calibration laboratories PNS ISO/IEC 17025. The numbering system of this Supplementary Requirements follows the numbering of PNS ISO/IEC 17025.

This document describes additional, specific accreditation requirements for laboratories performing biological testing of food products (raw, in-process and processed products), ingredients in the production of food, veterinary products, human drugs, waters including effluents, cosmetics, biocidal agents, microbial cultures and environmental samples pertinent to the products/materials mentioned above.

Majority of the accredited biological testing laboratories are primarily involved in microbiological testing. Thus this document does have a bias toward these types of laboratories. However this document can also provide guidance to laboratories using techniques in areas related to toxicology, veterinary science, biochemistry, molecular biology and cell culture, although there may be additional requirements for such laboratories.

2. Authorship

This document was prepared by the PAB LATC for Biological Testing. It is based on deliberation by the group of stakeholders convened by the PAB.

3. Definitions

3.1 Correction: Action to eliminate a detected nonconformity

3.2 Corrective Action: Action to eliminate the root cause of a detected nonconformity or other undesirable situation, thus, preventing recurrence of nonconformity.

3.3 Preventive Action: Action to eliminate the cause of a potential nonconformity or other undesirable potential situation, thus preventing occurrence of potential nonconformity.

3.4 Certified Reference Culture (CRC Microbiological): a reference culture certified or other documentation which is issued by a certifying body; e.g., cultures used to verify test systems, validate methods, perform quality control of test media, etc. must be traceable to a type of culture collection. Synonymous with Standard Reference Materials (SRM).

3.5 Reference Stock culture: A microorganism preparation that is derived from a reference culture.

3.6 Control Samples: Sets of samples tested by laboratories to determine if their processes are in control. A test sample with known properties of microorganisms examined on a routine basis to evaluate laboratory performance.

3.7 Culture: An isolated microorganism grown on laboratory medium.
3.8 Food Testing Laboratory: Laboratory that performs microbiological tests on finished food products, ingredients, in-process samples and associated environmental samples.

3.9 Test Samples: Samples in the laboratory that are in the process of being tested (not to be confused with in-process product samples from a manufacturing standpoint.)

3.10 Inspection: Activities such as measuring, testing and examining one or more characteristics of a product or services and comparing these with specified requirements to determine conformity.

3.11 Proficiency Test (PT) Samples: Test materials (split samples) with microorganisms (antibiotics and toxins) that are tested periodically by a number of locations to determine that proficiency of recovery, using statistical analysis where appropriate.

3.12 Reference Culture (RC): A culture, with cultural characteristics sufficiently well established to be used to calibrate/verify test systems and test media and validate methods.

3.13 Replicate Testing: Samples of RCs or CRCs which are tested by the same analysts. In each case, the results are compared for precision.

3.14 Split Samples: Unknown test samples of adequate homogeneity, sub-sampled and sent to laboratories for proficiency testing.

4. MANAGEMENT REQUIREMENTS

4.1 Organization

For laboratory staff that may also have production or marketing-related responsibilities, clear policies must be available to define how impartiality is assured for their testing responsibilities.

4.2 Management System

4.2.1 Quality documentation must include reference, approved signatories, scope of accreditation and the policy on the use of the PAB endorsement.

4.2.2 Measurable overall objectives must be established showing key performance indicators, key result areas, or other criteria and reviewed during the management review.

4.5 Subcontracting of Tests

4.5.1 A competent subcontractor is defined as an appropriate PAB accredited laboratory or a laboratory accredited by one of APLAC Mutual Recognition Arrangement (MRA) signatories. All results reported by the subcontractor shall be covered by an appropriate endorsed report.

4.5.4 The accreditation status of subcontractors should be regularly reviewed to ensure that it is updated.
4.5.5 In case no PAB/APLAC-accredited subcontractor is available, the laboratory shall ensure that the chosen subcontractor complies with the ISO/IEC 17025 requirements, e.g. the subcontracting laboratory may conduct quality audit to check if the management and technical requirements of this international standard is being followed by the chosen subcontractor.

Note: PAB may be contacted or the subcontracting laboratory may check the PAB website: www.dti.gov.ph regarding any information on accreditation status and scope of accreditation of its accredited laboratories.

4.6 Purchasing Services and Supplies

Calibration Service Providers

Providers of critical services such as calibration service shall be placed with a competent supplier. A competent supplier is preferably any PAB accredited calibration laboratory, a calibration laboratory accredited by one of APLAC Mutual Recognition Arrangement (MRA) signatories or any recognized national metrology institute. In such cases wherein supplier is the sole service provider or there is no PAB/APLAC-accredited calibration service provider for particular equipment, the supplier shall demonstrate its traceability of measurement. Refer to LA/SR10- Supplementary Requirements for Traceability of Measurements.

4.11 Corrective Action

4.11.1 The laboratory shall perform a timely investigation of nonconforming works or departures from policies and procedures. A time frame shall be defined in the laboratory’s management system from detection of non-conformity to raising or generating a corrective action report (however named).

4.11.2 The cause analysis must be extensive enough so as to identify the root cause(s) and not simply the symptom(s).

4.11.3 Correction, as defined in section 3, should be completed as soon as the non-conformity is detected (this is particularly important where the nonconformity could have significant impact such as affecting the quality of testing or customer satisfaction).

4.11.4 Timely corrective action(s) shall be appropriate such that the root cause of the nonconformity is addressed and eliminated.

4.12 Preventive Action

Preventive action is a proactive process to identify improvement opportunities, rather than a reaction to the identification of problems or complaints. Trend analysis of complaints and turn-around time of samples may assist this process. Consideration should also be given to providing staff with a formal mechanism for contributing suggestions for improvement.
4.13 Control of Records

4.13.1 General

4.13.1.1 All records must include the identity of the person making the record and the date of such creation, the person(s) checking data transcriptions and calculations and the date of such checking. Appropriate checking must be done, i.e. the person checking the records is not the same person who made/created them.

4.13.1.2 Unless otherwise prescribed by legislation or contractual obligations, retention times will not be less than five years or the maximum recalibration interval of the equipment, or five years, whichever is the longer period.

4.13.1.3 Test records that are created and/or retained electronically (e.g. Compact Disc, DVD, Hard Disk Drive, USB Flash Drive, etc.) shall be stored in a manner that protects them from hazards that degrade such media. Provision shall be made for the printing of such records when required.

4.13.2 Technical records

4.13.2.1 The records system must include a copy of each report and certificate that includes work covered by the scope of accreditation, or must allow one to be reproduced, including details such as the endorsement (if applicable) and identification of the person who authorized the report.

4.13.2.2 Alterations to data must also include the date the change was made. Corrections or amendments to test records are made in a manner that does not obliterate the original data and are signed or initialled and dated by the person responsible.

4.14 Internal Audits

The laboratory shall conduct an internal audit, at least over a twelve-month period that covers both the management and technical requirements of ISO/IEC 17025.

For the minimum requirements, refer to LA/GD 15 – Guidance Document for Internal Audits for Laboratories and Inspection Bodies (APLAC TC 002).

4.15 Management Review

The laboratory’s management shall review the effectiveness of the management system and testing activities at least once per year.

For the minimum requirements, refer to LA/GD 16 – Management Review for Laboratories and Inspection Bodies (APLAC TC 003).
5 Technical Requirements

5.2 Personnel

5.2.1 Staff competence and technical control

5.2.1.1 The minimum qualification for the technical staff in a biological testing laboratory shall be a graduate in Microbiology/Food Science/Pharmacology/Biotechnology/Biochemistry/Toxicology/Veterinary Science/Medical Technology. Alternative qualifications in biological sciences may meet requirements where staff has relevant experience relating to the laboratory’s scope of accreditation.

5.2.1.2 The laboratory shall be under the supervision of the officer who has education, experience and training in relevant discipline sufficient to meet the membership of an appropriate professional body, e.g. In some cases additional qualifications and/or experience may be required to meet condition set by a regulating or accrediting body.

5.2.1.3 There shall be a trained, competent supervisor having at least two years relevant laboratory experience for graduates of microbiology/biology major in microbiology and three years relevant laboratory experience for graduates of biomedical sciences, food science, or allied profession.

5.2.1.4 Any testing away from the base laboratory (such as in field laboratories, mobile testing laboratories or in the field) must be under adequate technical control. This would normally require either the location of an approved signatory at each facility or having an approved signatory visit each facility at least once each week and maintain a diary record of the dates and relevant activities of each visit.

5.2.1.5 In case testing requires visual observation (e.g. color retention), laboratory management must consider color vision as one of its qualification when determining the suitability of staff to perform such tests.

PAB Approved signatory shall meet the following requirements:

5.2.1.5.1 At least two years relevant laboratory experience for graduates of microbiology/biology major in microbiology and three years relevant laboratory experience for graduates of biomedical sciences, food science, or allied profession and have done at least 30 tests conducted/evaluated including Proficiency Testing (PT) participation.

5.2.1.5.2 The competence of approved signatory will be assessed and approved by the PAB once he/she meets the requirements. Consideration in minimum qualification and/or experience requirements for approved signatory on competence with objective evidences for proven competency may be considered by PAB upon recommendation by the assessment team.

5.2.1.5.3 The approved signatory for specific class of test structure under biological/microbiological testing must pass the corresponding Board examination for the practice of profession already regulated in the Philippines by 2018. Existing approved signatories by 2018 will be exempted from this requirement and will apply only to new and additional scope of tests that will be applied after the transition period. In the
absence of such regulations, the other minimum requirements already mentioned in 5.2.1 will apply.

5.2.2 The laboratory shall have a selection procedure and training system to ensure technical competence of all staff members.

5.2.2.1 Training shall include all methods or portions of methods and techniques that each person is responsible for performing. At a minimum, each analyst shall demonstrate competency for each parameter being applied for through observation by management and verification using replicate and/or check samples. For technicians performing only portions of a specific method, competency may be confirmed/verified by observation only.

5.2.2.2 On-going competence should be monitored objectively with provision for re-training where necessary. Where a method or technique is not in regular use, verification of personnel performance is necessary before the testing is undertaken. The interpretation of test results for identification and verification of microorganisms is strongly connected to the experience of the performing analyst and should be monitored for each analyst on a regular basis.

5.2.2.3 In addition to test methods, in some cases, it may be more appropriate to relate competence to a particular technique or instrument, for example use of approved biochemical, serological kits or microbial identification kits.

5.2.2.4 Training records shall include documentation of all relevant internal and external education and individual performance verifications.

5.2.3 Biological testing shall be performed by a competent analyst. Contractual and probationary staff shall be placed under training with adequate supervision.

5.3 Accommodation and Environmental Conditions

5.3.1 The internal layout for the microbiological laboratory must provide for sample receipt, washing-up and sterilization, media preparation and general testing areas. There should be separate areas of general testing and confirmation to reduce the likelihood of in process contamination. The design of each section should minimize hazards to personnel. Consideration should also be given to office, storage and staff facilities.

5.3.1.1 The storage facilities must be sufficient to allow for the retention of all samples for designated intervals and provide conditions that maintain sample integrity. Refrigerators or freezers must have adequate capacity when samples require refrigeration before or after testing.

5.3.1.2 For microbiological testing, reference cultures and certified reference culture shall be kept separated from samples at check-in and during storage. Sample check-in and storage shall be segregated (ideally, in a separate area from the testing laboratory) and shall include proper sanitation to exclude the possibility of cross contamination.

5.3.1.3 Bench tops (work surfaces) and floors shall be made of impervious, smooth, easily cleaned materials. It is desirable to have at least four linear feet of bench or surface workspace for each analyst while working. Walls and ceilings should be made of materials that are smooth and easily cleaned.
5.3.1.4 Animal housing requirements and handling of laboratory animals, laboratory hygiene and housekeeping shall be consistent with essential elements described in PALAS Code of Practice for the Care and Use of Laboratory Animals in the Philippines and the Animal Welfare Act of the Philippines.

5.3.2 The laboratory test area should be air-conditioned to control humidity and temperature. The laboratory shall be ventilated to reduce the level of contamination. Where air conditioners are used filters should be appropriate, inspected maintained and replaced according to the type of work being carried out. The required relative humidity in the test area is not more than 50% RH for the usual test and the temperature in the test area should be 20-25°C.

5.3.2.1 Excessive numbers of environmental bacteria, yeast and molds shall be controlled by air system with filters. Verification and monitoring of control shall be performed using air sampling devices, air settling plates, surface swabs or other appropriate means. These checks are critical to aerobic plate count and yeast and mold tests. The laboratory should establish acceptable limits of environmental microbial counts based on approved standards (e.g. SMEWW, BAM, Compendium of Methods for the Microbiological Examination of Foods, etc.)

5.3.2.2 Pathogen testing shall be strictly controlled so as to prevent cross contamination. Critical work surfaces shall be monitored for pathogens pertinent to the laboratory’s scope of testing (Salmonella, Listeria monocytogenes and E.coli O157:H7) – after sanitation but before testing operations begin.

5.3.3 The laboratory shall be arranged to minimize cross contamination and shall be segregated from other activities in the laboratory with limited access. Suggested means of accomplishing this are:

- Adequate hand washing facilities should be available
- Carry out procedures using appropriate precautions to ensure test and sample integrity (e.g. use of sealed containers) (Eurachem);
- Segregate activities in time and space. for:
  - Ante room prior to sterile room for changing laboratory gowns / footwear / head cover
  - Sample receipt and sample storage
  - Sample preparation
  - Handling and storage of reference cultures/Reference materials
  - Media preparation and Sterilization
  - Sterility testing
  - Decontamination
  - Animal House
  - Incompatible activities
- Use biological containment hoods;
- Restrict highly contaminated samples to separate areas;
- Restrict operations to selected areas when high levels of pathogens may be encountered (e.g., pre-enrichment, selective enrichment transfer).

5.3.3.1 For microbiological testing, laboratories located in facilities where products or ingredients are manufactured shall not test for infectious pathogens (such as Listeria monocytogenes,
Salmonella, Escherichia coli O157:H7, Shigella, Campylobacter, Vibrio cholera), unless the laboratory is physically separated with limited access, equipped with bio-safety cabinets and supervised by a qualified microbiologist.

5.3.3.2 Laboratories should take all precautions to avoid cross contamination.

5.3.3.3 For procedures that involve the handling of pathogens and reference stock cultures, they shall be operated within a safety cabinet of a class commensurate with the risk level of the microorganism handled. Most of the microbes encountered in a non-clinical testing laboratory belong to Risk Group 2 microorganisms e.g. Salmonellae, Staphylococcus aureus.

5.3.3.4 When working with samples containing microorganisms transmissible by the respiratory route e.g. Legionellae or when the work produces a significant risk from aerosol production, a biological safety cabinet of Class II shall be used.

5.3.3.5 Laboratories should have safety program appropriate for its requirements and take all necessary precautions to avoid laboratory accidents.

Additional Requirements for GMO Testing Laboratories

General

There shall be effective separation of the PCR testing area from neighboring laboratory areas to minimize the spread of contamination from nucleic acids and or nucleases (both DNase and RNase). A separate room shall be used for PCR testing in a laboratory. Procedure and precaution taken in avoiding the cross contamination shall be documented.

Reagents, consumables and equipment shall be located at appropriate designated areas to serve their specific purposes. Separate chambers or enclosures shall be provided for the following:

a. GMO negative control
b. GMO Positive control/plasmid/vector
c. Sample extracts and
d. Kits, master matrix, taq polymerase, primers, probes, reagents

Hygiene

Clothing appropriate to the type of testing performed (including, if necessary, protection for hair, beard, hands, shoes, etc.) should be worn in the laboratory and removed before leaving the area. This is particularly important in the molecular biology laboratory, where for example, movement from an area of high DNA load to one of low DNA load may unwittingly introduce cross contamination. (EA / Eurachem EA-04/10)

5.3.5 The laboratory shall have pest control programme /schedule.
5.4 Test methods and Method Validation

5.3.5 General

5.3.5.1 Standard Methods

Test methods and/or procedures held by the laboratory shall include:
- Scope
- Description of the sample to be tested
- Quantities to be tested
- Material, equipment and tolerances required
- Description of procedure
- Physical and environmental conditions required
- Sample identification
- Method of recording observations and results
- Safety measures
- Data required for reporting
- Sensitivity of method when applicable
- Method of data analysis and reporting

5.3.5.2 The laboratory shall use test methods that meet the needs of the customer. Where possible, these methods shall comply with the essential/critical elements of international, national and/or regional standards. Where no method is specified, the laboratory shall use an appropriate method that is traceable to recognized, validated method.

5.3.5.3 Many trade associations publish their own methods and may be useful resources. New methods, especially involving emerging pathogens, may be obtained from scientific journals.

5.4.2 Selection of Methods

5.4.2.1 Where a test can be performed by more than one method there must be documented criteria for method selection. Where relevant, the degree of correlation between the methods must be established and documented.

5.4.2.2 Standard methods do not require validation other than establishing the applicability of the method to the products under test and establishing staff competence in the method. However where standard methods are prescribed and followed, the laboratory is expected to maintain current versions of the standard methods (reference texts) and up-date laboratory bench methods in accordance with these. Although full validation is not required, a laboratory must verify that it can properly operate the method, and can demonstrate (where specified) the limits of detection, selectivity, repeatability and reproducibility.

5.4.2.3 Laboratories shall pay attention to the limitations, concentrations range and sample matrix specified in the test standards.

5.4.3 Laboratory-developed methods

Accreditation for draft Standards is not available. Laboratories may, however, be accredited for such methods if they are documented and validated as laboratory-developed methods.
5.4.4 Non-standard methods

5.4.4.1 This category includes modified standard methods, rapid method techniques (instrumental or biochemical) and in-house methods.

5.4.4.2 Any variation of a standard test method that could affect the outcome of the test result (e.g. change to time or temperature of incubation or the use of alternative growth media) must be validated.

Some rapid test systems may not require further validation if:
- The method is an AOAC, APHA, or FDA BAM approved method
- Validation data has been published and is applicable to the laboratories scope of work
- Extensive validation has been done by the manufacturer (e.g. based on collaborative testing) and is available and applicable.

5.4.4.3 The use of commercial test systems (kits) will require further validation if the laboratory is unable to source the validation data. When the manufacturer of the test kits supplies validation data, the laboratory will only perform secondary validation (verification).

5.4.4.4 Laboratories shall retain validation data on commercial test systems (kits) used in the laboratory. These validation data may be obtained through collaborative testing, from the manufacturers and subjected to third party evaluation (e.g. AOAC. Refer www.aoac.org for information on methods validation). If the validation data is not available or not applicable, the laboratory shall be responsible for completing the primary validation of the method.

5.4.4.5 It has been found in some cases (e.g. veterinary microbiological testing) that a specific test kit performs differently under local environmental conditions, to that of the original environmental conditions it was subjected to primary validation. In such cases the laboratory should conduct the validation to prove that the kit performs under local environmental conditions.

5.4.4.6 The laboratory must keep records of documented performance of the rapid test method and its applicability to the laboratory's scope of testing.

5.4.5 Validation of methods

5.4.5.1 The validation of microbiological test methods should reflect actual test conditions. Validation can be achieved by using a combination of naturally contaminated products and low level spiked products.

The laboratory shall validate standards methods applied to matrices not specified in the standard procedures. All in-house methods and rapid methods that have no published validation data must be validated.

5.4.5.2 Qualitative microbiological test methods, confirmation and identification procedures should be validated while validation of quantitative microbiological test methods if necessary, should be quantitatively determined in assays. The results should be evaluated with appropriate statistical methods.
5.4.5.3 If a modified version of a method is required to meet the same specification as the original method, then comparisons should be carried out and where necessary, experimental design and analysis of results must be statistically valid.

5.4.5.4 All validation data must be recorded and analyzed to establish sensitivity, specificity, repeatability and reproducibility of the method. Records must be kept at least as long as the method is in use, and as long as necessary to ensure adequate traceability of raw data and results. Validation data including applicability to the range of the products must be kept and be available for review at the assessment.

**Test Methods and Method Validation of GMO Testing**

**Test Methods:**

Laboratories should be clear about which matrices can and cannot be tested. There are some processed food matrices (e.g. soy sauce) where the integrity of the DNA needs to be assessed to decide whether the test has any validity.

When a GM screening test is used as a preliminary detection tool, the use of such test needs to be validated to demonstrate that it would detect a defined range of foreign DNA. If a GM screening test is negative and no further testing conducted, the result should be reported as no foreign DNA sequence detected with respect to the specific test conducted, with a specification of which traits have been excluded.

If a GM screening test is positive, then the laboratory should proceed to determine the specific trait present and can also specify the range of traits tested.

**Validation of Methods:**

The laboratory should be clear about which matrices are suitable for quantification. Basing quantification on a line from reference materials prepared from one matrix may not be appropriate for the same trait in a different (e.g. processed) matrix. As the availability of GM reference materials for quantification will always lag behind the traits that are on the market, the laboratory may mix its own quantification standards from 100% GM material, provided that the purity of the materials (GM and non GM) shall be established and proper validation undertaken.

Commercial test systems (kits) may not require further verification if validation data based on collaborative testing are available. Otherwise, laboratory shall be responsible for validation of the method. The laboratory shall demonstrate their capability to achieve the limit of detection quoted by the manufacturer or the laboratory has to establish its own limit of detection to minimise false positive and false negative results.

DNA assessment for analysis of items containing several ingredients or having been processed (e.g. food), laboratories shall verify that the extraction and clean up procedures used are capable of extracting good quality amplifiable DNA and the resultant extracts are free from inhibiting substances. Extraction method shall be validated for their ability to remove inhibiting substances.

For extraction method that has not been shown to remove consistently the inhibitors, an inhibitor control shall be used.
Test Methods for Toxicological Testing

Toxicological laboratories should preferably use standard study protocols and standard operating procedures/test methods referred in the OECD Test guidelines, ICH guidelines, etc. Modifications to such standard guidelines should be described and justified with proper validation. Details of test method validation should be retained with the raw data wherever applicable.

Laboratories should maintain details of experimental design including justification for selection of test system and its characteristics (species, strain, substrain, source, sex, age, weight, etc), justification for the method, frequency and dose of exposure, chronology of events, methods and materials, type and frequency of analysis/measurements and statistical evaluation etc.

5.4.6 Estimation of uncertainty of measurement

5.4.6.1 Biological testing laboratory shall apply a procedure to estimate its uncertainty of measurement however in the field of Biological Testing the estimation of uncertainty is inherently difficult or impossible to determine by the traditional metrological and statistically based procedures. It is recognized that the current state of knowledge regarding uncertainty of measurement across the full range of biological discipline is variable. PAB will only require estimation of uncertainty for biological methods where statistical analysis of uncertainty forms part of and is required by the method, however, laboratories are encouraged to have an understanding of the variability of all their results wherever this is possible.

Note: Laboratory may refer to the following useful references for establishing its procedure for estimating its uncertainty of measurement:
- URACHEM/CITAC document “Quantifying Uncertainty in Analytical Measurement” published by LGC, UK,
- ISO 19036,”Microbiology of Food and Animal Feeding Stuffs - Guidelines for the Estimation of Measurement Uncertainty for Quantitative Determinations. Published in Switzerland
- Other references appropriate for biological testing.

5.4.6.2 Laboratories need to make a formal estimate of measurement uncertainty for all tests in the scope of accreditation that provide numerical results. Where the test results are not based on the numerical data, e.g. detected/not detected, pass/fail, negative / positive or based on visual/tactile or other qualitative examinations uncertainty estimation is not required. Nevertheless the individual sources of variability, e.g. consistency of reagent performance and analyst interpretation should be identified and demonstrated to be under control. Similarly, no additional estimates of MU are required for well-recognized rapid methods that produce qualitative results.

a. Where the laboratory needs to estimate the measurement uncertainty, the laboratory must have a procedure for identifying the sources of uncertainty associated with testing
methods. This procedure must identify the mechanism used for documenting and identifying the major components that contributes to the uncertainty, and where applicable, present the calculations used for quantifying the measurement uncertainty for the test method. If customer do not currently require reporting or use of MU, the laboratory procedure must define the method for determining the uncertainty used by the method. The uncertainty estimation methods given by reputable professional and standard writing bodies can be accepted within the testing discipline and may be used. ISO/IEC 17025 does not specify any particular approach.

b. Once a documented procedure is established, the laboratory needs to develop and commence implementation of a programme for applying this procedure to all relevant tests within the scope.

c. The variability can be estimated for internal purpose through analysis of performance of the methods and operators in QC activities, proficiency, monitoring and training. Laboratories are required to establish and apply acceptance/rejection criteria wherever applicable.

5.5 Equipment

For calibration interval of equipment in microbiology laboratories refer to LA/SR07-Supplementary Requirements for Accreditation in the Field of Calibration, Appendix A-Equipment Calibration Intervals.

5.4.2 The biological testing laboratory shall be furnished with all items of sampling, measurement and test equipment required for the correct performance of the tests, including sampling, preparation of test items, processing and analysis of test data. Equipment commonly used for microbiological test includes anaerobic jars and cabinets, autoclaves, automatic pipettors, balances, centrifuge, clean work stations, colony counter, dispensers, freezers, hygrometers, incubators, microbiological safety cabinets, microscopes, pH meters, refrigerators, thermometers, waterbaths and equipment used in rapid methods, depending on the test/analysis performed.

5.4.2.1 All accredited biological laboratories are required to maintain a documented programme for the maintenance, calibration and performance verification of its equipment necessary to carry out the tests included in the scope of accreditation.

5.4.2.2 Calibration/verification and maintenance program for key microbiological instruments shall be established by the laboratory. Maintenance of essential equipments used in the laboratory shall be carried out at specified intervals as determined by factors such as the frequency of use. Detailed records shall be kept. If a test method or operating environment requires a more stringent calibration/verification interval than that set by the laboratory, more frequent calibration will apply.

5.5.2 Commonly used equipment for biological tests that requires calibration and/or performance verification include balances, thermometers, pH meter, timer, ovens, incubators, autoclaves, water bath, Laminar Flow chamber, Biosafety cabinets (for P2 level), thermal cycler and volumetric glassware. Calibration and Performance verification/Maintenance of equipment for biological tests shall be established as follows:
a) Anaerobic jars and cabinets must be monitored by means of an indicator, growth of known anaerobes.

b) Biological safety cabinets/ Biohazard cabinet shall be used for personnel protection when testing for hazardous microorganisms. It shall be maintained periodically depending on the class of the cabinet and shall undergo annual performance verification based on the type/source of BSC by a qualified/accredited calibration laboratory. Parameters such as final filter and exhaust filter integrity, air velocity and uniformity, air barrier containment, induced air leakage and UV radiation shall be monitored.

c) Centrifuge must be checked yearly by a tachometer.

d) Clean workstation such as Laminar flow cabinet must undergo annual performance verification and maintenance. Filters shall be checked and cleaned or replaced as needed. Airflow rate or particle count shall be monitored regularly or at least annually to comply with relevant standard. Cleanliness of hood surfaces shall be routinely monitored using appropriate method. Aerial microbial contamination shall also be checked regularly.

e) Regular cleaning of microscope is required and maintenance must be carried out by competent personnel.

f) pH meters should be checked using at least two (2) standard buffer solution prior to use (slope should be within the acceptable criteria). Preferably, there must be a separate pH meter exclusive for biological testing.

g) Autoclave shall not be used to sterilise clean equipment and to decontaminate used equipment during the same sterilisation cycle. Ideally the laboratories should have separate autoclave for these two purposes. Autoclave shall have gauges for both temperature and pressure. Biological with or without chemical indicators shall be used to check the effectiveness of sterilization. The lab shall determine the frequency of use of these indicators (As per SMEWW and Compendium, Biological indicators should be used at least monthly; Compendium also mentions use of temperature sensitive tape with each use). Records of autoclave operations including temperature and time shall be maintained. Acceptance and rejection criteria for operation conditions shall be set and implemented. Temperature controller shall be calibrated at least annually by an accredited calibration laboratory (Pressure gauge is calibrated separately from the thermometer). The temperature calibration results will reveal the pressure gauge deficiencies. Some autoclaves have a built-in timer for automatic operation and this shall be included in the annual calibration.

h) Temperature controllers, temperature recording device and thermocouples need to be calibrated every two years or more frequently if the device shows erratic readings.

Temperature-controlled equipments such as waterbaths, incubators, ovens and refrigerators must be monitored when in use to ensure compliance with the required temperature stated in the test methods. Calibration, where necessary, shall be done at least annually against the temperature specifications of the tests.
Where the accuracy of the temperature measurement has a direct effect on the result of the analysis, the temperature measuring/monitoring devices such as thermometers must be of sufficient accuracy to ensure that the equipment complies with the temperature tolerances specified in the test method. The graduation of the device shall be appropriate for the required accuracy. Traceability of measurement of the temperature measurement device also has to be established and overall uncertainty of measurement shall be estimated and appropriate for the measurement.

i) Weights and balances shall be calibrated traceably at regular intervals according to their intended use.

j) Initial verification and regular checks of volumetric equipment such as automatic dispensers, dispenser/diluters, mechanical hand pipettes and disposable pipettes shall be carried out to ensure that the equipment is performing within the required specification. Verification is not necessary for glassware if it has been certified to a specific tolerance. Calibration every two years shall be required for glasswares that are not within the specific tolerance. Equipment should be checked for the accuracy of the delivered volume against the set volume (for several different settings in the case of variable volume instruments) and the precision of the repeat deliveries should be measured.

k) For ‘single-use’ disposable volumetric equipment, laboratories should obtain supplies from companies with a recognized and relevant management system. After initial validation of the suitability of the equipment, it is recommended that random checks on accuracy are carried out. If the supplier does not have a recognized management system, laboratories should check each batch of equipment for suitability.

l) Microbiological water generation system – microbiological and chemical parameters must be monitored at regular intervals to ensure the continued production of microbiologically suitable water for laboratory use

m) The performance of the PCR equipment such as thermal cycler and the built in spectroscopic components of PCR equipment shall be verified regularly.

5.6 Measurement Traceability

5.6.1 General

For PAB recognized calibration service providers and additional information on measurement traceability, refer to LA/SR10 Supplementary Requirements for Traceability of Measurements.

5.6.2 Specific requirements

Reference standards and equipment shall be calibrated over the range and to the appropriate level of accuracy specified in relevant test methods.

5.4.2 5.6.3 Reference standards and reference materials
5.6.3.2 Laboratories shall demonstrate traceability by use of certified reference culture(s) obtained from a recognized national institute.

All cultures held by a laboratory must be uniquely identified. The system of identification must be traceable to a recognized culture collection or samples from which the cultures were sourced.

Certified reference culture(s), if available should be used to provide essential traceability in measurements and shall be used for the following:

- To demonstrate the accuracy of results,
- To calibrate equipment
- To monitor laboratory performance,
- To validate methods

Note: When a reference culture(s) is biochemical or immunological in nature, the mechanisms to ensure traceability of such reference culture(s) are not well developed, thus biological testing laboratories are expected to source their reference culture(s) (particularly when biochemical or immunological in nature) from the following possible sources (generally in decreasing order of preference) where availability permits:

a) Reference culture(s) from recognized culture collection or from accredited (to ISO Guide 34) reference culture(s) provider;

b) Reputable suppliers with ATCC/NCTC-derived organisms preferably with certification.

c) In-house produced reference culture(s) that are properly characterized and identified.

d) Isolates from PT samples as they are identified during the release of PT results.

5.6.3.3 Procedures for verification of stocks should be documented. Records related to preparation, verification and monitoring of reference stocks shall be maintained. Reference cultures may be sub-cultured once to provide reference stocks. Reference stocks shall be used to prepare working stocks of routine work (maximum of 5 passages from the reference culture).

a. Laboratories must hold and maintain a collection of cultures of organisms required to perform verification checks on methods and to conduct performance checks on batches of prepared media.

b. Documented procedures must be in place, covering the acquisition, preservation, maintenance and confirmation testing of the cultures in the collection. Hierarchical control of master and working cultures must be established and documented.

c. The following records must be kept:
   - identity, source and history of a culture (e.g. certificate of authenticity)
   - date of acquisition
   - conditions of resuscitation, preservation and storage (i.e. media, time, temperature of master and stock)
- number of units of master and stock cultures prepared/used
- number of passage
- results of purity, microscopic examination and biochemical tests performed
- dates of sub-culturing of stock to prepare working cultures
- results of purity checks on each new stock culture slope used to prepare a working culture
- conditions used to maintain working cultures (i.e. media, temperature, time)

5.6.3.4 Laboratories shall have a policy and procedures for handling, storage, preservation, maintenance and use of reference cultures and stocks. Reference culture(s) shall be packaged, stored and handled to minimise exposure to moisture, air, heat and light and eventually prevent deterioration. The laboratory shall also establish procedure for disposal of old or outdated reference culture(s). Records shall be maintained of receipt, use and disposal of reference culture(s).

5.7 Sampling

Laboratories responsible for sampling are encouraged to gain accreditation for sampling. The following conditions must be met to gain accreditation for sampling:

5.7.1 Sampling should only be performed by trained personnel. It should be carried out aseptically using sterile equipment. Sampling must be performed in accordance with recognized national and international procedures that are documented in sufficient detail to provide unambiguous instructions to all operators. Documentation of sampling must make a clear distinction between sample collection and the sampling plan.

5.7.2 The sampling procedures for sample collection and sampling plan must be cited on the test report whenever the laboratory wishes to extend the tests results from a sample to an entire batch.

5.8 Handling of Test Items

5.8.1 Maintenance of sample integrity.

5.8.1.1 Sample containers must be leak-proof and impervious to contamination during transport. Temperature during transportation and storage must be controlled and monitored as indicated in test procedures. Samples should be stored until the test results are obtained, or longer if required.

5.8.1.2 There shall be a written procedure and defined period for the retention and disposal of the samples in the laboratory. Laboratory sample portions that are highly contaminated should be decontaminated prior to being discarded.

5.8.2 Sample identification

Identification labels must be secured and legible. Samples must be labelled on the body of the container. Labelling only on caps and lids is not acceptable because of the risk of wrongly replacing lids.
5.9 **Assuring the Quality of Tests Results**

Laboratories shall establish and implement quality control plans to ensure and demonstrate that the measurement process is in-control and test results generated are accurate and reliable. The plans shall include types of quality control checks, their frequency and acceptance criteria, and actions to be taken when results will be outside the defined acceptance criteria.

5.9.1 The program for monitoring the reliability of results shall be based on the implementation and documentation of a comprehensive internal quality control program. For microbiological laboratories this will include:

- Maintenance, propagation and use of positive and negative controls
- Media quality control
- Instrument calibration, maintenance and use
- Staff training and competency performance evaluation
- Checking of calculations and results
- Accommodation and environment

5.9.1.1 Criteria for rejecting suspect results must be based on non-compliance with the predetermined criteria as defined in the internal quality control program (e.g. incubator temperature out of the range for duration or part of the test; negative control samples analyzed concurrently with test samples found to contain the determinant or target organism; insufficient dilution of perishable samples etc.).

5.9.1.2 An internal verification program must be run periodically to check the effectiveness of the quality control system. The extent and frequency of such program will depend on the number of testing staff and the range of tests for which the laboratory is accredited. The critical interval between performances of non-routine tests should be established and documented by the laboratory to ensure continuing competence.

5.9.1.3 Techniques for verification programs for training of staff should include any of the ff.:

- Regular evaluation of individual operators in quantitative skills (i.e. sample preparation, dilutions, plating and enumeration) and in qualitative skills (i.e. Competency to interpret selective/differential media and biochemical tests results; selection and enumeration of typical/atypical colonies for confirmation testing);
- Regular testing of split samples by more than one analyst;
- Checking against cross-contamination in daily operations by inclusion of negative (sterile) control samples; testing for recovery of specific microorganisms using samples spiked with low levels of reference cultures or natural isolates.

5.9.1.4 Quality control data (both internal and external) shall be fully documented in such a way that it is readily accessible for trouble shooting and following up on possible errors and trends.

5.9.1.5 Records of test verification and proficiency programs must be kept with other quality control data and be available for examination during assessment.
**Culture Media**

For in-house media preparation and quality control, the laboratory shall maintain an effective media preparation and quality control program. Details of the procedures for preparation and quality control must be documented as part of the laboratory quality system. Records must be kept of the preparation details for all types of media. This must include:

a) type of media  
b) unique identity (batch code)  
c) date of preparation and identity of an operator  
d) volume of media/solutions made  
e) ingredients, manufacturer, manufacturer’s batch number and quality of each component  
f) initial pH (pre-sterilization),  
g) final pH (post-sterilization),  
h) method of sterilization, including time, temperature and pressure as appropriate  
i) volume dispensed (if medium is used as a diluent or the volume is critical for other reasons).  
j) Volume check post-sterilization (if medium is used as a diluent or the volume is critical for other reasons).

All media produced must be checked for performance and records maintained. Information must include physical appearance, sterility results (including sample size), performance checks using positive and negative control organisms (e.g. biochemical reactions, morphology and recovery rates for semi-quantitative or quantitative methods).

Records of performance testing must be traceable to batch preparation records. The data generated must be used to assess the performance of each new batch against acceptance/rejection criteria.

**Reference Cultures**

Reference cultures of microorganisms available not directly from, but claimed to be traceable to a national collection may be used for quality control checks, but the requirements on number of passages and the relevant verification procedures required shall also be observed.

**Reagents and standard solutions**

Laboratories should ensure that the quality and grade of reagents including detergent used is appropriate for the test concerned.

Details of the preparation of all types of reagents must be recorded. The records should include the date of preparation, the identity of the person who prepared and an estimate expiry date.

Quality of reagent water used for critical processes should be specified and checked regularly for compliance against the requirements.
External Quality Assessment through Proficiency Testing Programme

Proficiency testing programme shall be scheduled and implemented on a regular basis relevant to their scope of accreditation. Preference should be given to proficiency testing schemes, which use appropriate matrices. In specific instances, participation may be mandatory.

Laboratories should use external quality assessment not only to assess laboratory bias but also to check the validity of the whole management system.

Laboratories are expected to select the proficiency testing activities according to the following criteria (in a generally decreasing order of preference):

(a) Mandated programmes. In some areas of biological testing, participation in a particular programme may be mandatory.
(b) International inter-laboratory comparison/PT programmes.
(c) National inter-laboratory comparison programmes.
(d) Proficiency testing programmes operated in accordance with PNS ISO/IEC 17043.
(e) Formal inter-laboratory comparison programmes involving several independent laboratories.
(f) Where none of the above is neither available nor applicable, an inter- or intra-laboratory comparison could be considered a valid proficiency testing activity.

The results from proficiency testing activities and their analysis will be reviewed in each PAB assessment.

For information relating to Proficiency Testing, please refer to LA/SR09: Supplementary Requirements on Participation to Proficiency Testing Programs.

5.10 Reporting of Results

5.10.2 Test reports

5.10.2.1 For microbiological testing of infectious agents where diagnostic (quantitative) tests can be differentiated serological types or biotypes, there shall be policies and procedures for interpreting, evaluating and reporting unequivocal results, since environmental factors/data may invalidate or cast doubt on the integrity of the results. (This is especially pertinent to Salmonella analyses where serological tests are well developed). The customer shall be notified of any factors that have affected or may potentially affect the integrity of reported results and shall be informed of any data interpretations or evaluations that are made.

5.10.2.2 In microbiological testing, results are reported following the prescribed reporting procedure based on the method used.

5.10.2.3 Where an estimate of the uncertainty of the test result is expressed on the test report on demand, any limitations (particularly if the estimate does not include the component contributed by the distribution of microorganisms within the sample) have to be made clear to the customer.
PAB endorsement
Endorsed test documents must include the information (a) – (k) detailed in this clause of ISO/IEC 17025. Additional details relating to the appropriate forms of endorsement and the reproduction of endorsed test reports are provided in LA/SR11: Requirements for the Use of PAB Laboratory and Inspection Body Accreditation Symbol.

Approved Signatories
The test document must be signed by a PAB approved signatory. Please refer to LA/GD 07: Guidelines for Laboratory Guidelines for Laboratory Personnel and Approved Signatories and LA/SR11: Requirements for the Use of PAB Laboratory Accreditation and Inspection Body Accreditation Symbols.

Unendorsed reports and Partial reports
An accredited laboratory may issue unendorsed documents reporting results within and outside its scope of accreditation or may issue partial test reports prior to final endorsed test reports. The final test report shall contain a reference to the partial test report. For information related to this, refer to LA/SR11: Requirements for the Use of PAB Laboratory Accreditation and Inspection Body Accreditation Symbols.

Electronic Signatures
The electronic report should show the identity of all the signatories in the original test report. This may involve an electronic signature. The security of these signatures should be such as to prevent inadvertent use or misuse.

5.10.3 Test Reports
In addition to the requirements detailed under 5.10.2, endorsed test documents must also include information as described in 5.10.3.1 and 5.10.3.2 where relevant.

5.10.6 Testing results obtained from subcontractors
Please refer to LA/SR11: Requirements for the Use of PAB Laboratory Accreditation and Inspection Body Accreditation Symbols for detailed information on results obtained from subcontractors.

5.10.7 Electronic transmission and remote issue of results

5.10.7.1 Test reports may be electronically issued (including from a site other than the accredited laboratory) provided that the reports have been appropriately authorized for release. The adequacy of such arrangements will be reviewed at assessment.

5.10.7.2 The laboratory must be able to demonstrate appropriate controls over the electronic generation, access, storage and back-up of results and reports and program controls such as password protection. If the report is to be accessed from a web site by the customer there must be an appropriate control in place to ensure the report can only be downloaded in a protected format.

5.10.7.3 Printing issues may also need to be considered. Any information normally included in a hardcopy report must be included on the electronically transmitted version and appear in any hard copy printed by the recipient. Flexible pagination to accommodate formatting changes when printed by the recipient may also be required.
6 References

General:

6.1 PNS ISO/IEC 17025:2005
6.2 Accreditation Procedures , LA/GD01/Issue 1/January 2015
6.3 Requirements for the Use of PAB Laboratory Accreditation and Inspection Body Accreditation Symbols LA/SR11/Issue 1/January 2015
6.4 Supplementary Requirements on Traceability of Measurements, LA/SR10/Issue 1/January 2015
6.5 Supplementary Requirements on Participation to Proficiency Testing Programs, LA/SR09/Issue 1/January 2015
6.6 Guidelines for Laboratory Personnel and Approved Signatories, LA/GD07/Issue 1/January 2015
6.7 Guide to the Expression of Uncertainty in Measurement (GUM), issued by BIPM, IEC, IFCC, ISO, IUPAC, IUPAP and OIML.

Biological Testing:

6.8 OECD Guidelines for Testing of Chemicals
6.10 PALAS Code of Practice for the Care and Use of Laboratory Animals in the Philippines. 2002. (PALAS)
6.12 Philippine Guidelines and Requirements for the Establishment of A Veterinary Biological Product Research and Development Laboratory. 2001. Philippine Council for Agriculture, Forestry and Natural Resources Research and Development - DOST
6.13 Guidelines for the Assessment of Microbiological Quality of Processed Foods. Bureau of Food and Drugs – Department of Health, Philippines
6.14 Philippine National Standards for Microbiological Methods
6.17 International Organization for Standardization (ISO) Microbiological Methods
6.20 USPHS. CBC Laboratory Manual, Quality Control in Microbiology. 1987. Atlanta, GA.
Bacteria:


Yeast:

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