## Article and resources provided by the African Society of Laboratory Medicine (ASLM)

## Quality control and external quality assessment strategies

To ensure that laboratories provide accurate and reliable results and reduce the risk of errors, implementing a quality management system (QMS) is important. However, in resource-constrained settings, most laboratories are not accredited and may only be partially implementing elements of a QMS. Introducing a new test, particularly under outbreak conditions, may therefore come with a high risk of errors. This step describes the key critical elements that laboratories should put in place to ensure quality results, particularly during COVID-19.

**What is QA?**

QA refers to the processes implemented by a laboratory to ensure that the final results are as accurate and reliable as possible. QA encompasses a range of activities that enable laboratories to achieve and maintain high levels of accuracy and proficiency despite changes in test methods and the volume of specimens tested. The main elements of QA include:

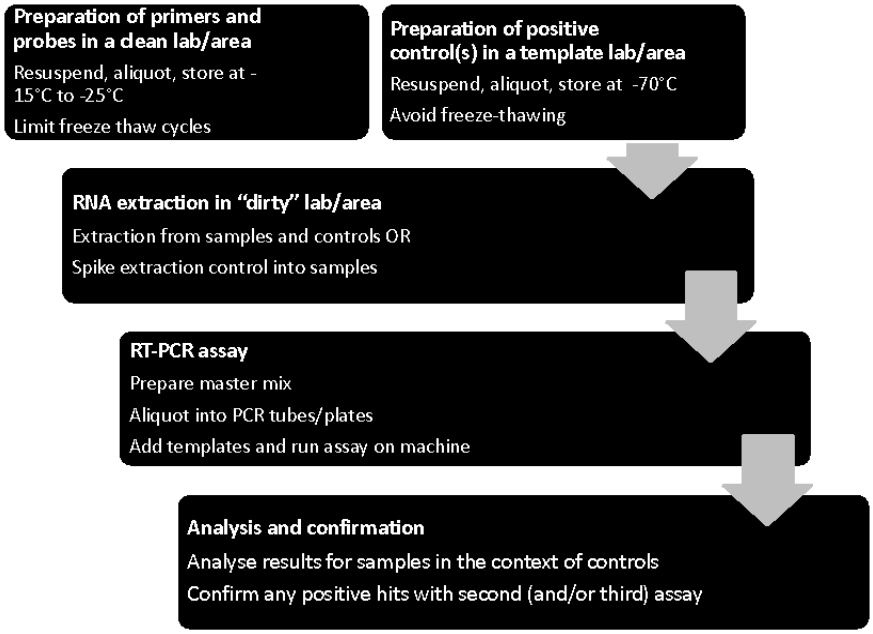
- **Standard operating procedures (SOPs),** which should be available, with staff trained on their use. SOPs should cover all procedures, from managing incoming specimens to authorizing and issuing test reports.
- **Documentation,** such as laboratory forms and registers, which should be standardized, and staff should be trained to fill out all documentation consistently and fully.
- **QC**, which refers to procedures used in each assay to ensure a test run is valid and results are reliable.
- **Quality indicator monitoring**, which refers to the collection and analysis of results or data that can serve as indicator for correct performance of the total testing process.
- **External quality assessment**, which analyses the accuracy of the entire testing process from receipt of sample to reporting of results.

## QC

QC aims to detect, evaluate and correct errors resulting from test system failure, environmental conditions or operator performance, before patient results are reported.

QC involves examination of control materials or known substances at the same time and in the same manner as patient specimens to monitor the accuracy and precision of the complete analytical process. QC must be included in each run and should cover each critical step of the PCR analysis as shown in **Figure 1.**

**Figure 1: COVID-19 PCR workflow. Key steps in sample/QC processing**




The QC includes the following components:

- **Extraction negative control:** indicates whether contamination was introduced from the extraction phase.
- **Extraction positive control:** provides an indication of the quality of the extracted template and whether the PCR was in anyway inhibited.
- **No template control:** indicates whether contamination was introduced from the PCR phase. Also indicates whether PCR reagents have been compromised and is used to determine threshold levels.
- **Positive template control(s):** uses synthetic SARS-CoV-2 RNA/DNA (either gene fragment or whole genome) to indicate the limit of detection and robustness of the assay.

A number of third-party commercial companies supply full process controls for SARS-CoV-2 testing. Some of the current providers include those listed below with prices ranging from 50−550 US dollars for 100 tests. Links to these organizations are provided in the See also section of this step:

- ZeptoMetrix
- SeraCare
- European Virus Archives Global
- Bio-Rad

Commercial QCs are preferred but in the absence of commercial controls, laboratories can use the following controls:

- **negative control:** water, universal transport media or viral transport media; and
- **positive control:** a patient sample with a known (and preferably low 25−30) Ct value (virus concentration) for human gene target e.g. RNAse P or non-human, non-SARS-CoV-2 extraction control e.g. equine arteritis virus.

A failure of any one of these controls (for instance, the positive control turns out to be negative) invalidates the test result and the assay must be repeated either from stored or newly collected sample after investigating and fixing the cause of the failure (e.g. contamination or degradation of the sample, or expired reagents). If patient results were already issued, they should be recalled immediately (providing an explanation of the reason for recall) and the patient should be urgently retested.

**New lot QC, or lot-to-lot verification** describes the process in which newly received lots/batches of test kits or test components are tested using a panel of samples to confirm that their performance is acceptable relative to the existing lot in use.

## Key performance indicators (KPIs)

KPIs are used to monitor the routine performance of the whole testing process and should be analyzed and reported on a regular basis (at least monthly). KPIs should include the following:

- number of specimens tested, by specimen type
- number (%) of positive, negative and invalid test results
- specimen rejection rate
- number (%) of failed internal quality control results
- EQA/PT performance (pass/fail or % score)
- turnaround time (TAT), between specimen collection and result reporting (total TAT), and within-laboratory TAT (% results reported within target TAT, average and range of TAT).

## What options are available for EQA for COVID-19 testing?

EQA allows a laboratory's testing performance to be compared to the performance of a peer group of laboratories, national reference or WHO reference laboratories. The three different methods for EQA programs are described below.

1. **Proficiency Testing (PT):** an external provider sends a blinded, well-characterized panel at intervals (usually quarterly) to a set of laboratories for analysis. The blinded panel is treated like a patient sample during testing, and the results are analyzed, compared and feedback reports generated. Laboratories should choose providers experienced in delivering EQA in their region.

Some examples of COVID-19 PT are shown below. Links to these organizations are provided in the See also section of this step:

- QCMD
- INSTAND
- WHO External Quality Assessment Project for the detection of influenza viruses by PCR
- ECDC/ERLI-Net

Laboratories can enroll for free as part of the influenza laboratory network or at a cost not exceeding 420 US dollars, but this may vary by country.

2. **Rechecking or retesting:** samples tested by one laboratory are retested by another laboratory (inter-laboratory comparison). WHO recommends that the specimens of the first five positive cases and the first 10 negative cases that meet the COVID-19 case definition for testing should be shipped for confirmation to the national reference or international referral laboratory for COVID-19.[1,2] After that, the laboratory can test for SARS-CoV-2 independently but should still collaborate with national reference laboratories or WHO referral laboratories for troubleshooting. Rechecking can be employed in the absence of a PT program.

3. **On-site evaluation:** usually done in addition to PT or rechecking and may be done when it is difficult to conduct traditional PT or rechecking/retesting. An evaluator (e.g. staff from national reference laboratory) will visit the laboratory to check if the laboratory is meeting quality requirements, retest and verify few test results and provide advice to correct any faulty procedures. On-site visits are also important to motivate staff and provide refresher training if needed. Due to the current situation, air transportation is limited, and it may not be feasible to get PT or conduct on-site evaluations. Therefore, countries are strongly advised to use the rechecking/retesting method as an option for an EQA program (sending samples to the national reference or WHO reference laboratories)[3] and to consider remote mentoring/supervision of laboratories by the national reference laboratory using web conferencing systems such as Zoom.

## What challenges are associated with implementing QA?

| Challenges | Mitigation measure |
|---|---|
| Unavailability of controls | Positive control: use of a confirmed positive patient sample. Negative control: use water/universal transport media/viral transport media. |
| Most methods are under development hence no validation data | Use methods with emergency use listing by WHO. Check https://www.who.int/diagnostics_laboratory/EUL/en/ and third party evaluated methods and perform method verification to the extent possible. |
| Unavailability of EQA schemes | Develop inter-laboratory comparison and send positive samples to national reference and/or WHO reference laboratories. |

## See Also

- ZeptoMetrix: https://www.zeptometrix.com/informationcenter/resources/zeptometrix-coronavirus-products.
- SeraCare: https://www.seracare.com/.
- European Virus Archives Global: https://www.european-virus-archive.com/detection-kit/2019-ncov-e-gene-stabilized-rna-positive-control-shipping-room-temperature.
- Bio-Rad: https://www.bio-rad.com/featured/en/coronavirus-covid-19-assay-development-vaccine-research.html.
- QCMD: https://www.randox.com/coronavirus-qcmd/.
- INSTAND: https://www.instand-ev.de/en/eqas/eqa-program.html.
- WHO External Quality Assessment Project for the detection of influenza viruses by PCR: https://www.who.int/influenza/gisrs_laboratory/external_quality_assessment_project/en/.
- ECDC/ERLI-Net: https://www.ecdc.europa.eu/en/about-us/networks/disease-and-laboratory-networks/erlinet-influenza-lab-quality-control.
- Coronavirus disease (COVID-19) Pandemic – Emergency Use Listing Procedure (EUL) open for in vitro diagnostics: https://www.who.int/diagnostics_laboratory/EUL/en/