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EDUCATIONAL GUIDE

Developing Patient Centric Quality Control Material

QUALITY CONTROL

Introduction

The clinical laboratory is an enigma to the uninitiated. While most are aware that many medical decisions are made based on results of laboratory tests[1], many are unaware of the intricate processes involved. Laboratory quality control (QC) is just one of the essential elements of laboratory medicine.

Laboratory QC material can be categorised as internal quality control (IQC) or external quality assessment (EQA). The former is designed to challenge precision and reproducibility of the test system through frequent testing and monitoring. The latter is used to assess the accuracy of the method by testing blind samples and reporting the results to an independent provider who compares the results with that of peers using the same methods and instrumentation to provide a performance score. Both are crucial parts of good laboratory practice and must be reliable. QC material that is of poor quality creates more problems than it solves; it is difficult to be sure if a novel bias or error is truly that, or simply a result of substandard test components.

All aspects of the clinical laboratory are subject to rigorous regulations. For QC, ISO15189:20222 accreditation is the gold-standard, with many other global accreditations bodies basing their recommendations on this guidance. The recent update to these regulations was designed to place more focus on mitigating the risk to the patient and now includes point-of-care testing (POCT), previously covered by ISO22870. The 2022 version of ISO15189 is a more robust and detailed standard which highlights the importance of many aspects of IQC and EQA and shows how crucial these factors are to providing accurate and reliable results to aid clinicans in their medical decisions and diagnosis. Under ISO15189:2022, all processes and procedures involved must be assessed in relation to the risk of impact on the patient[2]. Risk assessment is key.

Due to the complexity of some QC procedures and the high levels of risk associated with many incorrect laboratory results, it is imperative that the QC material used is reliable and of the utmost quality. Ensuring a high-quality QC product involves considering various factors such as material matrix, commutability, stability, lot-to-lot consistency, and the relevance of levels. Herein, we discuss some of the challenges faced in the development of patient centric QC material and how premium QC products and services can help, not only meet ISO15189:2022 accreditation requirements, but provide laboratories with unreserved confidence in the results they produce and provide to clinicians.

Designed to Fail

QC materials should be designed to provide a challenge to the test system and assess the suitability of these factors rather than compliment them; they are designed to fail. That is, to highlight problems with the test system that would not be apparent without the QC procedure.

There can be many sources for internal QC materials. Most instrument and assay manufacturers provide internal QC material which complement the relevant instrument, assay, calibrator, or lot. These are known as first-party controls. First-party QC materials are often designed to align with the test system, potentially masking inherent biases and hiding flaws. No test system is error free and a QC that is optimised to produce minimal failures is, in fact, not a control at all.

Good QC practices are designed to identify biases, not just faults. Therefore, it is vital that the QC material used is reliable and provides a relevant challenge, as to not undermine these strategically formulated QC procedures. Third-party QC materials are those designed and manufactured independently of a specific platform, test, or method and are truly impartial controls.

ISO15189:2022 states, ***"The use of third-party IQC material should be considered, either as an alternative to, or in addition to, control material supplied by the reagent or instrument manufacturer."***[2] The use of the word should here means that if laboratories decide not to use third-party controls, they must provide sufficient justification as to why they made this decision if they want to achieve their accreditation. This inclusion in the latest edition of the standard highlights the importance of effectively scrutinising the test procedure to reduce the risk to patients of erroneous results – an unidentified bias could, for example, generate a false negative result which if passed to a clinician could cause a missed diagnosis.

When considering patient centric quality control material, third-party IQC emerges as the optimal choice for mimicking real patient samples. A case report, published by Lima-Oliveria, et al. (2015) describes a situation in which an assay was recalled as the manufacturer discovered the kit was producing a positive bias of up to 45%. The first party control supplied as part of the kit was unable to detect this shift and therefore, as many as 3500 patient results were unreliable and needed to be recalled[3]. A true third-party control should have detected this shift before patient results were provided. The assay in question was one for the measurement of parathyroid hormone[3], which can have critical implications on patient therapeutic approaches.

The development of patient-centric QC material necessitates careful consideration from the manufacturer, ensuring that the IQC is versatile enough to operate across various instruments, accommodate diverse reagent methods, and yield results that instil confidence among laboratorians. This alignment across different instruments mirrors the consistency observed in authentic patient samples, reinforcing the reliability and relevance of the QC material in diverse laboratory settings.

Composition and Compatibility

One of the fundamental principles of the scientific method is the importance of introducing only one variable when investigating a hypothesis. Consequently, it is imperative to use QC materials that closely resemble the patient samples on which the test is designed to report. ISO151589:2022 states the following in relation to QC materials: ***"the matrix is as close as possible to that of patient samples"*** and ***"the IQC material reacts to the examination method in a manner as close as possible to patient samples."***[2]

A primary consideration in developing patient centric QC is the matrix composition. QCs designed as 100% human, or as close to as possible, are ideal for providing a challenge analogous to a patient sample and in many cases this is achievable. 100% human controls are those that do not contain animal constituents or additives which may behave differently to a patient sample, thereby providing an almost identical challenge. Again, in QC material development, the objective is to closely replicate, whenever feasible, the characteristics of an authentic patient sample.

This is important for protein-based assays, where the methods rely on highly specific antibody binding. While certain QC materials are manufactured to be as close to 100% human serum as possible, they may contain stabilisers, buffers, or antimicrobials to ensure other crucial features of the QC, such as stability, are maintained. These additives are thoroughly investigated to ensure they don't cause an adverse reaction in test systems which would affect the result, allowing them to be classified as 100% human controls. A balance must be achieved to ensure that the laboratory can use the QC material effectively.

Human-based QC materials are the next consideration in the development of QC. In some cases, maintaining stability or achieving appropriate concentrations can be troublesome in 100% human material. In laboratory medicine, the concentration of some analytes will rise or decline transiently while others degrade rapidly. When developing patient centric QC, adjusting to these challenges means adjustment of the material is needed. In such cases, human serum, plasma, or urine is diluted using a suitable buffer which allows features, such as stability or clinically relevant levels, to be upheld. In rarer cases, an aqueous matrix may be required to preserve stability, particularly at low concentrations, for example, ultra-low concentrations of PSA degrade quickly in human serum.

A common alternative to human QC material involves the use of animal serums, like bovine or equine[4]. However, this approach deviates considerably from the essence of patient centric QC since the reactions of QC based on animal serums differ significantly from those in human samples. While animal-based serums offer a cost-effective solution utilised by many QC manufacturers[4], the compromise is significant, as it fails to present a clinically relevant challenge.

Polymerase Chain Reaction (PCR) is becoming increasingly common in the clinical laboratory, and with the inclusion of POCT into ISO15189:2022, these diagnostic tools require the same level of quality management as other forms of laboratory medicine[2]. Regarding commutability, for example, human blood contains natural PCR inhibitors such as haem and heparin, which are removed through successful extraction and purification. An unsuitable matrix will not provide a challenge to this part of the molecular workflow.

There are 2 main types of molecular control: recombinant and whole pathogen. Recombinant QC materials are synthetic controls manufactured through genetic recombination techniques. Briefly, the target gene sequence of interest is excised from the pathogen and inserted into a plasmid of a donor cell. This gene sequence is then expressed by the donor and can be detected by the test. This manufacturing method allows large volumes of controls to be produced at low expense but will give rise to issues. Firstly, as the donor cells are not human, these controls do not provide a challenge representative of a patient sample. Single-target PCR assays are designed to look for a single gene sequence relating to the pathogen of interest. However, the best assays, multiplex assays, can detect many gene sequences providing a more thorough examination of the pathogen.

Conversely, whole pathogen, or whole genome QCs, consist of state-of-the-art, characterised QC material that contain fully intact organisms of interest providing an examination of all gene targets relevant to the pathogen. They not only provide a clinically appropriate challenge to the test method, but examine the whole molecular workflow, from extraction to result, a feat recombinant QC material cannot achieve. When developing a truly patient centric QC, the aim is that the QC material reacts with the entire test system in the same manner as the patient sample.

Clinically Relevant Challenges

Many modern clinical assays boast extensive analytical measuring ranges. This provides the advantage of being able to detect a wide range of concentrations confidently and accurately. The purpose of quality control procedures is to confirm the suitability and clinical functionality of a test method or system. To achieve this, it is paramount that the test is challenged at the concentrations used by clinicans to make judgments on diagnosis, in other words, at clinical decision limits. Quality control procedures are designed to confirm that assays perform at the high levels required at these concentrations. It follows that the QC materials used must contain concentrations relevant to these clinical decision levels.

ISO15189:2022 states, ***"the IQC material provides a clinically relevant challenge to the examination method, has concentration levels at or near clinical decision limits and when possible, covers the measurement range of the examination method."***[2]

This requirement is a distinguishing factor between QC providers. Particularly in internal controls such as multianalyte and serology controls, consistently achieving and maintaining clinically relevant concentrations from lot to lot and ensuring stability over time requires expertise. The development of the QC material to meet these stringent criteria involves substantial costs, necessitating careful consideration in feasibility studies, validation lots, and trials. Consequently, distinct variations emerge among QC manufacturers in their ability to adhere to this patient centric QC design.

Issues can emerge due to various factors. For example, proteins used in some controls can compete for binding with others, causing havoc with the antibody interactions used for quantitative analysis. This means QC material manufacturers must play Goldilocks, ensuring that they get the concentrations of analytes just right, not only to maintain exceptional performance, but to provide this performance at the physiological relevant levels required for clinical decision making. In qualitative detection, for example serology controls, providing QC material around the cut-off values is essential to providing a clinically relevant challenge.

A different obstacle is posed for molecular QC material. Unlike clinical chemistry, molecular techniques like PCR are more novel and established clinical references aren't as readily available, if they are defined at all. This means QC manufacturers have less definitive values to target when specifying the concentration of their QC material. To produce QC material at the most appropriate levels, 2 approaches can be taken. First, large scale distribution studies can be used to calculate the mean and upper and lower ranges of the most commonly achieved results for a positive or negative diagnosis – a technique used during COVID-19. Alternatively, extensive literature review must be undertaken to evaluate the available literature from around the world to gather data from different clinical observations to determine the appropriate concentrations in which to target for QC material values.

Cost of Poor Quality

Quality is a laboratory's reputation; time consuming to achieve, difficult to maintain and quick to ruin. When time is spent constructing procedures to maintain high levels of quality, the impact of the wrong choice in quality control material can be detrimental. QC materials play a pivotal role in instilling confidence in the accuracy of results from patient samples, and any error in this aspect can significantly affect clinical outcomes and patient care. To illustrate, the effect of repeat testing can be significant. Delays in result reporting have been associated with 61% longer emergency department residency and 43% delays in receiving treatment[5]. When considering the diagnosis of life-threatening conditions such as stroke and heart disease, every minute wasted increases the risk of mortality[6].

Using QC material that does not effectively challenge the test system can lead to false confidence in the test. Not all QC material is created equal; a distinction exists between materials designed to complement the testing process, potentially concealing errors or biases present in reagents, calibrators, or instruments, and those designed to truly challenge the test system. The consequences of this can be mild - a single stray result may go unnoticed. On the other hand, the repercussions can be much more severe. If even a small bias or trend is hidden, these can, over time, cause results to drift continually further away from the true value. This could potentially cause many false negatives, which if used to make clinical decisions may result in missed diagnosis, and worse outcomes for the patient. If the challenge is near the clinical decision limits, even a small bias may cause significant effects from erroneous results.

In one case, an error in a test system result in falsely low results of sodium in a sample taken from a boy with insulin-dependent diabetes (age 6). After treatment for hyponatraemia and upon discovering the test system was faulty, subsequent tests determined the boy's sodium concentration to be 222mmol/L, following which the patient died due to intracranial haemorrhage[7]. With the increased emphasis on the mitigation of risk to the patient in ISO15189:2022, it is essential that high quality QC material is used to reduce the potential harm caused by erroneous laboratory results.

On the other hand, Suboptimal QC material can result in increased financial burden. Low stability may mean laboratories need to replenish their stock more frequently or the QC material will degrade before it has been used, both of which increase raw material costs for the laboratory. In cases where stability does not meet the manufacturer's claims, this can result it false errors which must be investigated, potentially leading to increased downtime and costs associated with retesting, such as reagent and material costs.

Conclusions

The development of patient-centric QC material stands as a critical determinant of the accuracy and dependability of laboratory results. Navigating the complexities of laboratory processes, particularly within the realm of quality control, plays a pivotal role in ensuring trustworthy outcomes that underpin medical decisions. The significance of IQC and EQA cannot be overstated; they are integral components of good laboratory practice.

Adherence to rigorous regulations, notably the ISO15189:2022 accreditation, is paramount for upholding the gold standard in laboratory quality control. The recent update to these regulations strategically amplifies the focus on mitigating risks to patients, extending its purview to encompass point-of-care testing and providing a more comprehensive standard for evaluating IQC and EQA.

The cost of subpar materials extends beyond a mere hit to a laboratory's reputation, potentially leading to erroneous results with far-reaching consequences for patient outcomes. The use of QC materials that obscure errors or biases poses a significant risk, underscoring the importance of opting for high-quality materials that genuinely challenge the test system.

In essence, investing in premium QC products and services not only facilitates compliance with accreditation requirements but also instils confidence in the reliability of results provided to clinicians. This aligns seamlessly with the overarching objective of delivering accurate and dependable outcomes in the dynamic landscape of medical diagnostics.

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