Pre- and post-analytical phases

Andreas Bietenbeck

P. B. Luppa, R. Junker (Eds.), *Point-of-Care Testing*
https://doi.org/10.1007/978-3-662-54497-6_4

4.1 Introduction

The objective of all quality assurance measures in laboratory medicine is to produce a proper diagnosis based on conclusive findings. Thereupon rests the foundation of optimal treatment for the patient. Conversely, this means for both laboratories and POCT users a paradigm shift from quality control of analyses to comprehensive quality management. This objective can only be achieved if the overall diagnostic process is calculated into the equation; this process extends from the correct patient-focused ordering of tests for the disease-relevant analytes to the correct interpretation of the analysis results aimed at rendering the proper diagnosis and monitoring the therapy based on that diagnosis.

This process can be divided into a pre-analytical analytical and a post-analytical phase. The pre-analytical phase includes everything from choosing an appropriate test and correctly identifying the patient to taking a sample, transporting it if necessary and preparing for analysis it in the laboratory. Validation and the appropriate reporting of findings follow post analysis. Recently, considerations about non-analytical factors (extra-analytical quality) have become increasingly important [3, 12, 17] and been incorporated into the German Medical Association's Guideline on Quality Assurance in Medical Laboratory Tests (RiliBÄK) [1].

Although fewer errors happen in the pre-analytical phase of POCT than in a central laboratory, this phase is still meaningful. Approximately one third of all errors in POCT occur prior to the actual analyses [14]. POCT pre- and post-analytical phases depend on the specific analytes to be assayed as well as on the devices used. Several overarching principles shall be addressed below.

4.2 Pre-analytical phase

The most important pre-analytical subroutines

1. Choosing a suitable test
2. Preparing the patient (e.g. diet, drugs, position of the body, time of the sample collection)
3. Collecting the sample
4. Transporting and storing the sample
5. Inspecting the sample (hemolyzed, icteric, lipemic appearance)
6. Processing the sample (e.g. centrifugation).

Bullet points 4 to 6 play a less important role within the POCT concept. As POC tests are carried out promptly, possible false readings caused by sample instability are not an issue either. The correct way of sample collection (point 3), however, is particularly important in the pre-analytical process. Many POCT systems use venous, arterial or capillary blood samples. Swabs are increasingly used to detect infectious diseases. POC tests usually require considerably less sample material. Rigorously implemented hygiene measures are particularly important for mobile devices to avoid the spread of pathogens [21].

4.2.1 Choosing a suitable test

Many POCT systems deliver results quicker than conventional central laboratories as no sample transport is required. However it is often not possible to achieve the same accuracy with highly integrated POCT devices as with larger laboratory devices. Therefore, investigations with a POCT device should not be regarded as equivalent to those carried out in a central laboratory even if they measure the same analyte [18]. POCT is, however, well suited for disease monitoring and controlling therapy.

4.2.2 Capillary blood sampling

Capillary blood is a mixture of blood from arterioles, venules and capillaries, sometimes diluted by interstitial or intracellular fluid (hemolysis). The relative composition depends on the blood circulation of the puncture site; heating leads to arterialization of the blood sample.

Differences exist between venous and capillary blood, which can influence hematological tests or oral glucose tolerance tests. In adults, capillary blood is usually taken from the fingertip or ear lobe, while the heel is used in neonates. For therapy monitoring, capillary blood can also be taken from other skin areas (▶ Chapter 12). Good blood circulation around the puncture site is important. To obtain arterialized capillary blood for blood gas analysis, the puncture site needs to be treated with warm water compresses (42° C) or by applying a special cream (e.g. Finalgon) to hyperemize the local tissue.

There is a linear correlation between blood volume and puncture depth. Therefore, puncture aids that can be adjusted to the required sample volumes are recommended. For self-monitoring, an extensive range of puncture aids for capillary blood sampling are available where the puncture depth can be adjusted. In addition, there are multiple lancets to go with some puncture aids, which differ in their bevels and suppression of painful vibrations.

In Germany, the technical regulations for biological materials (TRBA 250) need to be considered when using puncture aids in hospitals, physicians' practices, emergency vehicles etc. In blood sampling, "the use of safe instruments where body fluids are present in amounts that can transmit infections" is mandatory. Among other tasks, the following device safety specifications must be met:

- The safety mechanism must be part of the device and needs to be equipped with an audible or tactile signal. It may be used only once.
- The safety mechanism must be triggered single handedly, immediately after use. Self-activating systems are recommended as they are usually easier to handle.
- Safe work equipment must be compatible with the accessories and other systems used.

When using puncture aids in pediatrics/neonatology, the puncture depth on the child's heel is critical because of the danger of injuring the calcaneus, particularly in pre-term infants. For this reason, lancets with a shorter puncture depth (max. 2.0 mm) are preferred.

After skin disinfection, a single-use lancet is used to puncture. The blood sample is collected by gently pressing onto the tissue (not squeezing) to avoid or minimize hemolysis – one of the most important pre-analytical confounding factors. The first drop of blood is discarded as it is often contaminated with tissue components. In blood glucose monitoring, the first drop can be discarded; although this is not the case in INR self-monitoring where the first drop needs to be analyzed. The next few drops are collected in capillary tubes (e.g. end-to-end capillary tubes) or special capillary blood containers. A wide range is available from different manufacturers. Containers with additives should be inverted 5 times (not shaken), after filling with blood.

The correct technique for capillary blood sampling from the heel of an infant is shown in ◘ Fig. 4.1.

4.2.3 Venous blood sampling

Before phlebotomy, the patient should have been in a seated or prone position for 15 minutes, if possible. After disinfection of the puncture site, venous stasis is achieved with a tourniquet or blood pressure cuff (stasis between systolic and diastolic blood pressure) applied for no longer than a minute. The patient should not be asked to make a fist! The stasis is terminated immediately after successful venipuncture. There are different recommendations regarding the order in which samples are taken [4, 8], not all of which specifically address POC tests.

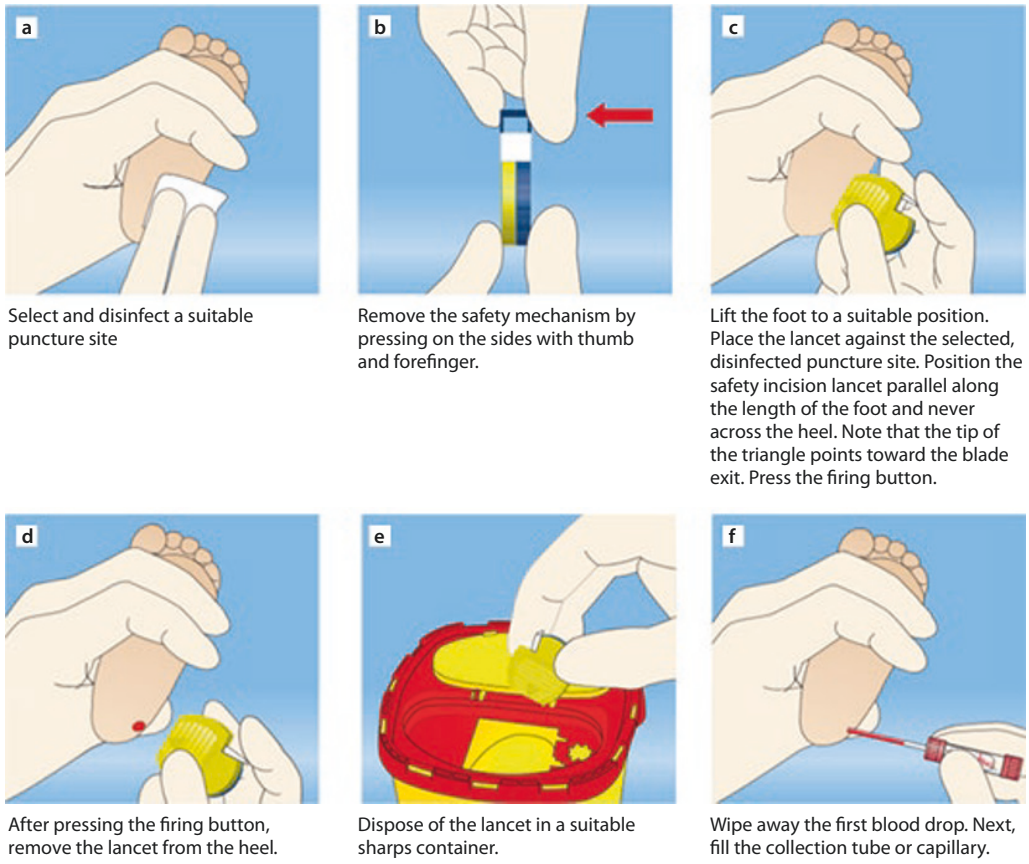

Fig. 4.1 Correct use of an incision safety lancet for newborns (Courtesy of Sarstedt, Nümbrecht, Germany).

In German speaking countries, for safety reasons, only closed blood collection systems are used. These are based on two different principles: Vacuum and stamp blood collection. The systems are difficult to combine as they are designed differently and have various attachments; with different sizes and accessories.

4.2.4 Arterial blood sampling

Arterial blood sampling is indicated, particularly for blood gas analysis (▶ Chapter 14) [2]. The samples are usually taken by puncturing the femoral, brachial or radial artery or via an arterial line. After the blood collection, firm pressure should be applied to the puncture site. Capillary blood can also be used for blood gas analysis when taken from hyperemized skin areas on the earlobe or fingertip.

Special closed glass or plastic needle-tube systems (see below) allow automatic filling due to the arterial pressure, while minimizing the risk of air bubbles in the sample. Anaerobic blood samples should be ensured; any air bubbles need to be removed immediately. Otherwise, the blood-dissolved gases equilibrate with the respective gas molecules inside the air bubbles. The CO_2 concentration is more affected than O_2 as CO_2 has a higher partial pressure in the blood than in the air. In contrast, the measurement of arterial O_2 partial pressure in the hyperoxic range (>200 mm Hg) is flawed because the diffusion gradient is greater towards room air [7]. After drawing the sample, it should be mixed by careful rolling between

both hands and promptly analyzed within 5–10 minutes. If this is not possible, it is recommended to store the sample in ice water for up to 30 minutes [5]. However, it is very important to avoid hemolysis. Mix it again before testing! If sample transport by tube mail is possible, it is important to check the specifications of the system. Pneumatic tube systems, which generate low shearing forces thanks to their controlled acceleration and careful cornering and are approved for blood sample transport, are completely suitable for blood gas samples [7].

4.2.5 Blood sampling systems and anticoagulants for blood gas analysis

Ideally, whole blood samples should be taken with glass syringes, as dissolved gases cannot diffuse through glass. When using plastic syringes, it should be remembered that false results of pO_2 and pCO_2 can occur due to diffusion. Therefore, the sample should be analyzed promptly, as described above. Heparin-coated plastic capillaries are suitable for taking capillary blood samples. Every manufacturer of blood gas devices offers their own blood sampling systems. Examples are Siemens Medical Solutions RAPIDLyte pro, a self-filling system with capacity selection, and Radiometer's safePICO with vented safeTIPCAP and an integrated mixing ball, ensuring fast and thorough mixing of the sample before analysis. Roche offers, for example, the BS2 Blood Sampler and the MICROSAMPLER for its blood gas systems. Other systems are also available on the European market.

Ca^{2+}-balanced heparin is the most frequently used and most suitable anticoagulant, as it does not change the acid-base balance. If lyophilized heparin syringes are not used, it is usually satisfactory to rinse a 2-mL syringe with heparin solution, making sure that only a volume of 0.1 mL of heparin remains in the syringe conus. This way, an excess of heparin solution can be avoided, which would otherwise lower the pH of the sample (heparin is an acid mucopolysaccharide) and would lead to dilution errors. A heparin solution using 500 or 1000 IU/mL to rinse the syringe is recommended. A final concentration of 20–30 IU/mL will remain in the sample.

Blood gas analysis is often combined with a measurement for electrolytes and therefore only lithium heparin is suitable, not sodium or potassium heparin. As all heparins can bind positively charged ions such as Ca^{2+}, syringe systems with electrolyte compensating heparins were developed for the use of combined blood gas and electrolyte analysis. These electrolyte-balanced heparin solutions eliminate the interference from ion binding (e.g. Pico sampler by Drott Medizintechnik).

4.2.6 Blood samples from central lines

The sampling of blood via intravascular needles, cannulas or central lines is very common, particularly in intensive care units. Before the sample is taken, the central line should be flushed with heparin and a minimum of twice the line volume, approximately 2–5 mL blood, should be discarded to avoid sample contamination with infusion solutions or medications [10]. The time between the last infusion and blood sampling should be at least 15 minutes. Particular caution needs to be taken when POCT systems are employed, as they are more sensitive to interfering substances than large central laboratory devices.

4.2.7 Taking swabs

Various sample swabs are used particularly for POCT diagnostics of pathogens. Common sites are nasal, pharyngeal, inguinal, rectal or wounds, depending on the suspected pathogen. Multiple sites may need to be swabbed to identify a specific pathogen [19]. Different swabs are used for different sites [15]. Generally, the swab should be moistened when taking a sample from a dry surface. Avoid contamination.

All contaminated materials need to be disposed of safely. The swab often remains in the single-use cassette of the POCT device. While antibiotic use prior to sample taking must be avoided with conventional microbiological methods based on pathogen culturing, this precaution is not relevant for molecular POCT methods as these tests adopt a nucleic acid amplification technique.

4.2.8 Urine sampling

Urine is usually collected by non-invasive methods, which makes it particularly practical for POC testing by the patients themselves. The time of collection is, however, important as many analytes in the urine show a significant circadian rhythm. The first morning urine is particularly useful for nitrate and protein detection. Urine fractions are not uniform. The first urine fraction is often contaminated with pathogens from the urethra. Mid-stream urine is the preferred sample for many tests, as most analytes are diluted in the last urine fraction.

4.2.9 Inspection of the sample

Routine inspections in the central laboratory identify hemolytic, lipemic and icteric blood samples, which is not possible with POCT, as whole blood samples are mainly used. Hemolytic blood samples are of particular concern and cause by far the most frequent pre-analytical errors in many assay methods. The only solution – albeit unsatisfactory – is the avoidance of hemolysis by employing the proper sampling technique. A similarly difficult problem to solve involves micro-clots present in samples for hemostaseological or blood gas analyses and often not detected by inspection. Many POCT systems carry out internal checks to identify and reject unsuitable samples. To date, blood gas analyzers do not yet feature integrated hemolysis detectors as explained in ▶ Chapter 14. Intra-vascular hemolysis cannot be detected when heparinized blood samples are used.

4.2.10 Reliable identification of patient and sample

Many POCT devices take the sample directly. Therefore, sample containers usually do not need to be labeled, but the patient must be identified properly and the test documented correctly. Where POCT devices are used professionally, a barcode e.g. from the patient's wrist band is scanned directly into the device. This kind of technical option is not available in simpler POC tests, such as lateral flow assays.

Important pre-analytical errors and problems

- Inadequate patient preparation (e.g. diet prior to function tests)
- Inadequate information about a patient's condition (e.g. medication history, body temperature and body position in blood gas analysis)
- Incorrect patient identification
- Incorrect sampling time (for example in oral glucose tolerance test and other function tests, lack of consideration of circadian rhythm)
- Incorrect sampling technique (for example by capillary blood: incorrect site or insufficiently hyperemized skin, hemolysis due to tissue squeezing; contamination when collecting from a central line)
- Transmission of infections from insufficient hygiene measures

4.3 Post-analytical phase

The post-analytical phase of the diagnostic process begins once the parameters have been measured. This phase comprises the following key steps:

- Technical and possibly medical validation of test results
- Reporting the results to the attending clinician
- Entry of the results in the (electronic) medical record

- Documentation of the person who carried out the test
- Recording of the results in an electronic information system (if available)
- Securing documentation over the period of time for medical, legal and organizational reasons.

Post-analytical errors are often less obvious than pre-analytical ones, but equally important for the quality of the results. The following post-analytical errors and problems are not necessarily POCT-specific, but definitely POCT-relevant. Because fewer checks are available with POCT, there is a risk that POCT errors will have a bigger impact on a patient until the findings affect further action by the physician [12]. Post-analytical errors often depend on organizational situations, such as how far devices, wards, outpatients, laboratories and hospital administration are interlinked (▶ Chapter 26). As in the pre-analytical phase, a well-designed device can prevent errors. Clearly visible warning signs displayed on the device can ensure that extremely divergent values are recognized as such.

Important post-analytical errors and problems

- Insufficient validation of results carried out under pressure for short turnaround times
- Incorrect classification of results
- Incorrect or incomplete verbal reporting of results, such as missing or wrong metrological units
- Delayed reporting of alarm limit excursions
- Confusing result reporting, missing notification of results outside the normal reference range
- Errors in data storage in the laboratory or hospital information system
- Incorrect or incomplete documentation, such as verbal result reporting without entry in the medical record. No documentation of person who performed the test

4.4 Avoiding pre- and post-analytical problems

Pre- and post-analytical – as well as analytical – errors are not completely avoidable [11, 13], but can often be significantly reduced by organizational measures. A number of recommendations have been issued [6, 9, 11, 12, 16, 17, 20]; detailed information can also be found in the RiliBÄK and DIN EN ISO 15189 (▶ Chapter 36 and 38). The likelihood of pre- and post-analytical errors can be significantly reduced by employing a well-designed POCT device. For example, if a device only allows a measurement when a patient's barcode has been scanned, this considerably reduces the risk of inadequate patient identification. Automated data transmission into a laboratory information system can eliminate transmission errors, too.

Sources of errors, which cannot be resolved by design changes, need to be minimized carefully when carrying out the test. All pre- and post-analytical steps ought to be described correctly and detailed procedural instructions established (standard operating procedures, SOP), which can be summarized in the POCT quality management manual.

The DGKL working group "Reference Values" has published an exemplary SOP for the pre-analytical phase, which is an important aid for the standardization of pre-analytical conditions [8].

Furthermore, a competent governance committee (e.g. the POCT committee; ▶ Chapter 30) should develop strategies for error prevention as well as track and reduce incidents, including possible changes in the working processes. Intensive staff training on a regular basis and good communications is of vital importance as well.

References

1. Bundesärztekammer (German Medical Association), Instand e.V. (2015) Guidelines of the German Medical Association on quality assurance in medical laboratory testing. GMS Z Forder Qualitatssich Med Lab 6:Doc03
2. Burnett RW, et al. (1995) International Federation of Clinical Chemistry (IFCC). Scientific Division. Committee on pH, Blood Gases and Electrolytes. Approved IFCC recommendations on whole blood sampling, transport and storage for simultaneous determination of pH, blood gases and electrolytes. Eur J Clin Chem Clin Biochem 33(4): 247–253
3. Carraro P, Plebani M (2007) Errors in a stat laboratory: types and frequencies 10 years later. Clin Chem 53(7): 1338–1342
4. CLSI document H3-A6. Edition, A.S.S., Wayne, PA: Clinical and Laboratory Standards Institute, 2007
5. Davis MD, et al. (2013) AARC Clinical Practice Guideline: Blood Gas Analysis and Hemoximetry: 2013. Respiratory Care 58(10): 1694–1703
6. Ehrmeyer SS, Laessig RH (2007) Point-of-care testing, medical error, and patient safety: a 2007 assessment. Clin Chem Lab Med 45(6): 766–773
7. Gruszecki AC, et al. (2003) Utilization, reliability, and clinical impact of point-of-care testing during critical care transport: six years of experience. Clin Chem 49(6 Pt 1): 1017–1019
8. Gurr E, et al. (2011) Musterstandardarbeitsanweisung Präanalytik/Exemplary standard operation procedure pre-examination/Arbeitsgruppe Richtwerte der Deutschen Vereinten Gesellschaft für Klinische Chemie und Laboratoriumsmedizin. LaboratoriumsMedizin 35:55–60
9. Laposata M, Dighe A (2007) "Pre-pre" and "post-post" analytical error: high-incidence patient safety hazards involving the clinical laboratory. Clin Chem Lab Med 45(6): 712–719
10. Lippi G, et al. (2006) Phlebotomy issues and quality improvement in results of laboratory testing. Clin Lab 52:217–230
11. Lippi G, et al. (2006) Preanalytical variability: the dark side of the moon in laboratory testing. Clin Chem Lab Med 44: 358–365
12. Meier, F.A. and B.A. Jones (2005) Point-of-care testing error: sources and amplifiers, taxonomy, prevention strategies, and detection monitors. Arch Pathol Lab Med 129(10): 1262–1267
13. Nichols JH (2005) Medical Errors: Can We Achieve An Error-Free System? Point of Care 4(4): 139–141
14. O'Kane MJ, et al. (2011) Quality Error Rates in Point-of-Care Testing. Clinical Chemistry 57(9): 1267–1271
15. Panpradist N, et al. (2014) Swab Sample Transfer for Point-Of-Care Diagnostics: Characterization of Swab Types and Manual Agitation Methods. PLoS ONE 9(9): e105786
16. Plebani M, Carraro P (1997) Mistakes in a stat laboratory: types and frequency. Clin Chem 43(8 Pt 1): 1348–1351
17. Plebani M (2007) Errors in laboratory medicine and patient safety: the road ahead. Clin Chem Lab Med 45(6): 700–707
18. Schifman RB, Howanitz PJ, Souers RJ (2016) Point-of-Care Glucose Critical Values: A Q-Probes Study Involving 50 Health Care Facilities and 2349 Critical Results. Archives of Pathology & Laboratory Medicine 140(2): 119–124
19. Senn L, et al. (2012) Which anatomical sites should be sampled for screening of methicillin-resistant Staphylococcus aureus carriage by culture or by rapid PCR test? Clinical Microbiology and Infection 18(2): E31-E33
20. Stahl M, Lund ED, Brandslund I (1998) Reasons for a laboratory's inability to report results for requested analytical tests. Clin Chem 44(10): 2195–2197
21. Thompson ND, Schaefer MK (2011) "Never events": hepatitis B outbreaks and patient notifications resulting from unsafe practices during assisted monitoring of blood glucose, 2009–2010. J Diabetes Sci Technol 5(6):1396–1402