6020

Quality Assurance/Quality Control

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6020 A. Introduction

Quality control (QC) results render meaningful the results of analytical tests. Essential QC measures (described in Part 1000) include method calibration, reagent standardization, assessment of each analyst's capabilities, analysis of blind check samples, determination of the method's sensitivity (method detection level or quantification limit), and daily evaluation of bias, precision,

and the presence of laboratory contamination or other analytical interference. The details of these procedures, their performance frequency, and expected ranges of results should be formalized in a written Quality Assurance Manual and standard operating procedures (SOPs).

Some of the methods in Part 6000 include specific QC procedures, frequencies, and acceptance criteria. These are considered the minimum quality controls needed to perform the method successfully; additional QC procedures can and should be used. If the QC criteria listed in this section exceed those listed in the individual methods, the criteria in this section must also be included. Some regulatory programs may require further QC or have alternative acceptance limits.

Each method typically includes acceptance-criteria guidance for precision and bias of test results. If not, the laboratory should determine its own criteria via control-charting techniques. Evaluate bias using recoveries from laboratory-fortified blanks (LFBs). Evaluate precision by analyzing duplicate or spiked duplicate samples. Additional acceptance criteria guidance may be provided by program- or project-specific requirements.

To help verify the accuracy of calibration standards and overall method performance, participate in an annual or preferably semi-annual analytical program of single-blind QC check samples (QCS), ideally provided by an external entity. Such programs are sometimes called proficiency testing (PT)/performance evaluation (PE) studies. An unacceptable result on a PT sample is often a strong indication that a test protocol is not being followed accurately. Investigate circumstances fully to find the cause. In many jurisdictions, participation in PT studies is a required part of laboratory certification and accreditation.

6020 B. Quality Control Practices

1. Initial Quality Control

a. Initial demonstration of capability (IDC): Before new analysts run any samples, verify their capability with the method (see Section 1020 B.1 for specifics). Run at least four LFBs (6020 B.2*e*) and compare results to the limits listed in the method. All instrument performance checks and calibration requirements must be met before analysis. (Note: Analysis and evaluation of a method blank is required.) If no limit is specified, use the following procedure to establish initial limits:

Calculate the standard deviation of the 4 samples. The LFB's recovery limits are

$$\text{LFB's initial recovery limits} = \text{Mean} \pm (5.84 \times \text{Standard Deviation})$$

where:

5.84 = the two-sided Student *t* factor for 99% confidence limit for 3 degrees of freedom.[1]

The initial limits provided by this process should be considered temporary. Limits developed from more replicates (e.g., at least 20) will give a better determination of accuracy and precision. (For basic guidance on demonstrating capability, see Sections 1020 B.1 and 3.)

b. Method detection level (MDL): If data will be reported below the calibrated range, then before analyzing samples, determine the MDL for each analyte using Section 1020 B.4 or other applicable procedures.[2] MDL determination and verification are not required if 1) data are not reported below the instrument's calibrated range, and 2) the ability to provide quantitative data at the reporting limit is verified. Determine the MDL for each analyte in a method and matrix category. The laboratory should define all matrix categories in its QA plan. Perform a new MDL determination whenever changes in the method's instruments or operating conditions may affect sensitivity. Ideally, samples for MDL determinations should be analyzed over at least a 3-d period to generate a more realistic value. Include all sample preparation steps in the MDL determination.

Ideally, use pooled data from several analysts rather than data from one analyst to determine overall lab MDLs. (For specific information on MDLs and pooling, see Section 1020 B.4.)

Verify the MDL on each instrument used in the laboratory by analyzing a QC sample (subjected to all sample-preparation steps) spiked at a level 1 to 4 times the MDL. A successful verification is one that meets all the method's detection criteria. Repeat the verification at least annually.

c. Operational range: Before using a new method or instrument, determine its operational range (upper and lower limits), or at least verify that the intended range of use is within the operational range. For each analyte, use standard concentrations that provide increasing instrument response. The minimum reporting level (MRL) is set to a concentration at or above the lowest standard used in the analysis. Quantitation at the MRL must be verified initially and at least quarterly (preferably daily) by analyzing a QC sample (subjected to all sample-preparation steps) spiked at a level 1 to 2 times the MRL. A successful verification meets the method's or laboratory's accuracy requirements at the MRL. Laboratories must define acceptance criteria for the operational range, including the MRL, in their QA documentation.

2. Ongoing Quality Control

a. Calibration: Initially calibrate with at least 5 non-zero calibration standards of the analyte(s) of interest. If using second-order fits, include at least 6 non-zero standards.

Select calibration standards that bracket the sample's expected concentration and are within the method's operational range. The number of calibration points depends on the width of the operational range and the shape of the calibration curve. One calibration standard must be at or below the method's reporting limit.

As a general rule, the range of standard concentrations should not be greater than 3 orders of magnitude, and may be much less. For example, concentration variables of 1, 5, 10, 50, and 100 can be used if the operational range is 2 orders of magnitude.

Apply response-factor, linear, or quadratic curve-fitting statistics, as appropriate, to analyze the concentration–instrument response relationship. If the relative standard deviation of the response factors is ≤15%, then the average response factor may be used. Otherwise, use a regression equation. The appropriate linear or nonlinear correlation coefficient for standard concentration-to-instrument response should be greater than or equal to 0.995 for

linear calibrations and 0.990 for quadratic calibrations. Weighting factors (e.g., $1/x$ or $1/x^2$) may be used to give more weight to the lower concentration points of the calibration.

Back calculate each calibration point's concentration. The back-calculated and true concentrations should agree within ±30% for points above the MRL and ±50% at or below the MRL, unless different criteria are specified in an individual method.

Use initial calibration to quantify analyte concentrations in samples. Use calibration verification only to check the initial calibration, not to quantify samples. Repeat initial calibration at least annually or when calibration verification criteria cannot be met. (For basic calibration guidance, see Section 1020 B.11.)

b. Calibration verification: Verify calibration by periodically analyzing a continuing calibration standard during a run. If not specified otherwise in the individual method, analyze after each 20 samples and at the end of the run. Analyses using internal standards may omit the verification at the end of the run. The calibration verification standard's analyte concentration may be varied over the calibration range to determine detector response. Some methods may also require the analysis of an instrument blank after the continuing calibration standard.

For the calibration verification to be valid, check standard results must not exceed the limits specified in the method or in Table 6020:1 (if not specified in the method).

If a calibration verification fails, immediately cease analyzing samples and take corrective action. Often, the problem can be fixed by performing injector maintenance or trimming a few cm from the front of the column. Then, reanalyze the calibration verification. If the calibration verification passes, continue the analysis. Otherwise, repeat initial calibration and reanalyze samples that have been run since the last acceptable calibration verification.

If the LFB is not prepared from a second source to confirm method accuracy, the laboratory must also verify the accuracy of its standard preparation by analyzing a mid-level second-source calibration standard whenever a new initial calibration curve is prepared. Results must agree within 25%, unless otherwise specified in a method. (A second source is either from another vendor or a completely different lot from the same vendor. If neither option is feasible, then the second-source calibration standard must be prepared from primary stock materials by a different analyst.)

c. Quality control sample (QCS): Analyze an externally generated, blind QCS (unknown concentration) at least annually (preferably semi-annually or quarterly). Obtain this sample from a source external to the laboratory, and compare results to that laboratory's acceptance results. If testing results do not pass acceptance criteria, investigate the reasons, take corrective action, and analyze a new QCS. Repeat this process until results meet acceptance criteria. Record all attempts to meet criteria. Multiple failures indicate problems with method operation. External proficiency test (PT) samples meet this criterion.

d. Method blank (MB): Include at least 1 MB daily or with each batch of 20 or fewer samples, whichever is more frequent. Prepare and analyze the MB in exactly the same manner as field samples, including all preparation and cleanup steps, and all preservatives used in samples. Any constituents recovered must generally be less than or equal to one-half the reporting level (unless the method specifies otherwise). If any MB measurements are at or above one-half the reporting level (if reporting to MRL) or greater than the MDL (if reporting to the MDL), take immediate corrective action (as outlined in Section 1020 B.5). This may include reanalyzing the sample batch or qualifying the reported data. Sample results that are below the MRL are considered valid even if the MB has a detection above the MRL, but should be qualified for information purposes. For common lab contaminants, such as methylene chloride, a lab may need to use a higher MRL to meet the MB criteria.

e. Laboratory-fortified blank (LFB): The LFB and LFM may be made from the same source standard as the initial calibration or from a second source. If the LFB and LFM are from the same source as the ICAL, the ICAL must be verified using a second source standard (see 6020 B.2*b*).

Using stock solutions, prepare fortified concentrations so they are within the calibration curve. Prepare at least 1 LFB each day samples are prepared or with each preparation batch of 20 or fewer samples, whichever is more frequent. Prepare and analyze the LFB in exactly the same manner as the field samples, including all preparation and cleanup steps and all preservatives.

Calculate percent recovery and determine the control limits (Section 1020 B) for these measurements. Some methods may have specific limits to use in lieu of plotting control charts. In those cases, control charts may still be useful in identifying potential problems but are not required. Ensure that the LFB meets the method's performance criteria when such criteria are specified. If the LFB does not meet the acceptance criteria, the method is out of control; take corrective action. Re-prepare and reanalyze as samples with an acceptable LFB. If impossible, qualify the reported data.

f. Laboratory-fortified matrix (LFM)/Laboratory-fortified matrix duplicate (LFMD): Prepare at least one LFM/LFMD each day samples are prepared or with each preparation batch of 20 or fewer samples. (For basic guidance on LFMs and LFMDs, see Sections 1020 B.7 and 8.) Some regulatory programs require more frequent use of LFMs. When analytes of interest are expected to be present, the laboratory may substitute a duplicate analysis for the LFMD. If the client does not provide enough sample volume for the LFM and LFMD analyses, the laboratory may perform duplicate LFB analyses to generate precision data for the analysis.

To prepare an LFM, add a known concentration of analytes to a randomly selected routine sample without increasing its volume by more than 1%. Otherwise, account for the dilution mathematically. Ideally, the new concentration should be at or below the midpoint of the calibration curve. Spike all analytes of interest to the client. Process the LFM and LFMD as separate samples through entire sample preparation and analysis. If necessary, dilute the spiked sample at analysis to bring the measurement within the calibration curve.

Calculate percent recovery and relative percent difference, plot control charts (unless the method specifies acceptance criteria), and determine control limits for spikes (Section 1020 B). Ensure that the method's performance criteria are satisfied. If the LFB meets acceptance criteria, failures usually indicate problems created by the sample matrix. If the native analyte concentration is more than four times (4×) greater than the spike concentration, spike recoveries may be unreliable. Precision data may still be usable based on the total analyte concentration (native + spike).

g. Duplicates: Using duplicates is appropriate when there is a high likelihood that the compounds of interest will be present in the sample, particularly at high concentrations that make spiking difficult. If not, use LFMDs instead. Methods in this section routinely use LFMDs. Process duplicate samples independently through the

Table 6020:1. Minimum Quality Control for Methods in Part 6000

Topic	Section	MB	LFB	LFM, LFMD	Surrogate	ISTD	Notes
Constituent concentration by gas extraction	6040 B	•	•	•	•	•	1
	6040 C	•	•	•	•	•	2
	6040 D	•	•	•	•	•	2
	6040 E	•	•	•	•	•	
Volatile organic compounds	6200 B	•	•	•	•	•	2
	6200 C	•	•	•	•	•	3
Methane	6211 B	•	•	–	–	–	
	6211 C	•	•	–	–	–	
EDB and DBCP	6231 B	•	•	•	–	–	4
	6231 C	•	•	•	–	–	
	6231 D	•	•	•	–	–	4
THMs and chlorinated organic solvents	6232 B	•	•	•	–	(•)	5
	6232 C	•	•	•	–	–	
	6232 D	•	•	•	–	–	
DBPs: HAAs and trichlorophenol	6251 B	•	•	•	•	•	
DBP: Aldehydes	6252 B	•	•	•	•	•	6
Extractable base/neutrals and acids	6410 B	•	•	•	•	•	2, 3
Pharmaceuticals and personal care products	6810 B	•	•	•	•	•	5
Phenols	6420 B	•	•	•	–	•	2, 3
	6420 C	•	•	•	•	•	
PCBs	6431 B	•	•	•	•	–	
	6431 C	•	•	•	•	•	
Polynuclear aromatic hydrocarbons	6440 B	•	•	•	–	–	
	6440 C	•	•	•	•	•	
Nitrosamines	6450 B	•	•	•	•	•	5
	6450 C	•	•	•	•	•	5
Carbamate pesticides	6610 B	•	•	•	–	(•)	
Organochlorine pesticides	6630 B	•	•	•	•	–	
	6630 C	•	•	•	•	–	4
	6630 D	•	•	•	•	•	
Acidic herbicide compounds	6640 B	•	•	•	•	•	
Glyphosate herbicide	6651 B	•	•	•	–	–	
Tributyltin	6710 B	•	•	•	•	•	
	6710 C	•	•	•	•	•	4

ISTD = internal standard; MB = method blank; LFB = laboratory-fortified blank; LFM = Laboratory-fortified matrix; LFMD = laboratory-fortified matrix duplicate.
• indicates a test is mandatory and (•) indicates it is optional.
1. LFM plus duplicate is acceptable.
2. Gas chromatograph/mass spectrometer (GC/MS) tuning required.
3. Chromatography checks required.
4. Second-column confirmation or GC/MS confirmation required.
5. Additional QC guidelines in method.
6. Confirm optional.

entire sample preparation and analysis. Include at least 1 duplicate for each matrix type each day samples are prepared or with each preparation batch of 20 or fewer samples. Calculate control limits for duplicates when method-specific limits are not provided. (For basic guidance on duplicates, see Section 1020 B.8.) Some regulatory programs require more frequent use of duplicates.

h. Surrogate standards: Where indicated in methods, surrogate standard recoveries are used to monitor for matrix effects in field samples and analytical problems in all samples. Before preparation, spike all QC and field samples using a concentrated solution of surrogate standards. Ideally, target a concentration at or below the midpoint of the calibration range.

Calculate percent recovery and determine control limits (Section 1020 B) for these measurements. Some methods may have specific limits to use in lieu of calculating control limits. If so, control charts may still be useful in identifying potential problems but are not required. Ensure that surrogate recoveries meet the method's performance criteria (when such criteria are specified) or the laboratory-generated limits. Failures may indicate analytical problems or problems tied to the sample matrix. Establish actions to be taken if surrogates do not satisfy acceptance criteria.

i. Internal standards: Internal standards are used in some methods to normalize instrument responses and provide retention time references. Where used, track internal standard response(s) and retention time(s) and compare to the criteria stated in the method. Establish actions to be taken if internal standards do not satisfy acceptance criteria.

j. Retention times: The laboratory must have procedures to develop retention time windows and monitor retention times. Although advances in chromatographic instrumentation controls mean that minor shifts in retention times may not be noted in some analyses, the laboratory must make at least initial determinations of retention-time windows on each type of analytical system for each analyte. Follow the criteria in the method. If there are none, then the laboratory must follow its own procedure for determining retention-time windows and analyte identification criteria.

k. Second column confirmation: If a method requires that analyte identification be confirmed via a dissimilar second column, ensure that the phases are dissimilar enough to invert the elution order of some compounds in the analysis or—if the method only involves a few target analytes—significantly change the pattern of elution. One column may be used solely to quantitate analytes and the other just to confirm analyte identification. If so, the confirmation column need not meet all of the method's calibration and QC criteria; however, demonstrate daily that the confirmation column is sensitive enough to identify all compounds at the level being reported. This may be accomplished by analyzing the lowest calibration standard showing adequate signal for all analytes on both columns. Some methods or programs may require quantitative analysis on both columns. If so, the laboratory must meet all QC criteria on both columns.

l. Additional instrument checks: Certain methods may require additional QC checks on analytical performance (e.g., endrin/DDT breakdown checks in the analysis of chlorinated pesticides or mass spectrometer tuning). If noted in a method, they are required and must be performed as indicated. However, instrument parameters relating to chromatography (e.g., temperature or gradient ramps and profiles and even column choices) may be optimized as long as all QC and compound identification criteria can be met. All calibration standards, QC samples, and field samples must be analyzed using identical conditions.

m.Demonstration of ongoing proficiency: Each laboratory analyst must demonstrate ongoing proficiency with the method according to criteria established in the laboratory's Quality Assurance Manual and SOPs. The demonstration may be accomplished be repeating the IDC or by an evaluation of ongoing QC data. Analysts who have not performed the analysis in more than a year should repeat the IDC to verify their proficiency.

3. Calculations

a. LFM recovery:

$$LFM\,\%\,Recovery = \left[\frac{LFM\,conc \times (spike\,vol + sample\,vol) - (sample\,conc \times sample\,vol)}{spike\,solution\,conc \times spike\,vol}\right] \times 100$$

b. LFB and surrogate recovery:

$$\frac{C_b}{I} \times 100 = \%\,\text{Recovery LFB}$$

where:

C_b = LFB or surrogate concentration determined experimentally, and
I = initial concentration of analytes (or surrogate) added to LFB or sample.

c. Relative percent difference:

$$\left[\frac{|\,LFM - LFMD\,|}{\left(\frac{LFM + LFMD}{2}\right)}\right] \times 100 = RPD$$

or

$$\left[\frac{|\,D_1 - D_2\,|}{\left(\frac{D_1 + D_2}{2}\right)}\right] \times 100 = RPD$$

where:

LFM = concentration determined for LFM,
$LFMD$ = concentration determined for LFMD,
D_1 = concentration determined for first duplicate, and
D_2 = concentration determined for second duplicate.

References

1. Meier PC, Zund EE. Statistical methods in analytical chemistry, 2nd ed. New York (NY): Wiley Interscience; 2000.
2. U.S. Environmental Protection Agency. Definition and procedure for the determination of the method detection limit, revision 1.11. 40 CFR Part 136, Appendix B. (1995).

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