match" quality or fit that is within the defined limits that the laboratory has set and validated. Ion ratios determined from total spectra analysis are an acceptable identification method, and should fulfill the same criteria as given above for ion ratio identification.

Laboratories using mass spectrometric methods for quantitative purposes based on total ion current measurements (without ion ratios) should have ancillary information and assay characteristics that validate this process, eg, known compound of interest, retention times, potential interferences by endogenous compounds or other drugs/metabolites, etc.

Evidence of Compliance:
✓ QC and test records

REFERENCES
1) Clinical and Laboratory Standards Institute (CLSI). *Liquid Chromatography-Mass Spectrometry Methods.* 2nd ed. CLSI document C62. Clinical and Laboratory Standards Institute, Wayne, PA; 2022.

CBG.17400 Identification Criteria - Tandem Mass Spectrometry **Phase II**

The identification criteria for tandem mass spectrometry (MS/MS) are validated and recorded.

NOTE: In tandem mass spectrometry using multiple reaction monitoring (MRM) there is at least one transition monitored for the internal standard and another for the analyte.

Evidence of Compliance:
✓ QC and test records

***REVISED** 08/24/2023*
CBG.17500 Matrix Effect Assessment of Mass Spectrometry Assays - Validation **Phase II**

There is a record of assessment of matrix effects in development and validation of mass spectrometry assays.

NOTE: Matrix effects can affect analyte ionization and performance in both directions: suppression or, less frequently seen, enhancement. Evaluation of matrix effects must be performed during assay development and validation.

Examples of evaluation protocols may include:
1. *Post Column Infusion - Constant infusion of analyte followed by injection of blank matrix specimen extracts to measure ionization response*
2. *Mobile Phase/Post Extractions Spiking - Compare response of analyte spiked into mobile phase to that of analyte spiked into blank matrix specimen extracts*
3. *Internal standard monitoring - Evaluate trends in internal standard abundance and signal to noise ratios during an analytical run that includes blank and spiked matrix specimen extracts.*

The minimum number of different matrix sources may vary based on the matrix, analytical targets, and assay design. Associated data should be used to evaluate the impact of matrix effect on results and define appropriate acceptance criteria for each reportable analyte during routine testing of patient samples.

REFERENCES
1) Clinical and Laboratory Standards Institute. *Mass Spectrometry in the Clinical Laboratory: General Principles and Guidance; Approved Guideline.* CLSI Document C50-A. Clinical and Laboratory Standards Institute, Wayne, PA; 2007.
2) Clinical and Laboratory Standards Institute (CLSI). *Liquid Chromatography-Mass Spectrometry Methods.* 2nd ed. CLSI document C62. Clinical and Laboratory Standards Institute, Wayne, PA; 2022.
3) Annesley TM. Ion Suppression in Mass Spectrometry. *Clin. Chem.* 49, pp. 1041-1044 (2003).

***REVISED** 08/24/2023*
CBG.17600 Matrix Effect Assessment of Mass Spectrometry Assays - Routine Monitoring **Phase II**

The laboratory evaluates mass spectrometry assays for possible ion suppression or enhancement in patient samples during routine testing.

NOTE: Ion suppression (or less frequently, ion enhancement) is a recognized analytical anomaly in mass spectrometry assays. Such suppression can lead to false negative results or poor quantitative analyses (especially near assay limit of quantitation). While difficult to predict and observe from specimen to specimen, certain precautions should be used to try to detect ion suppression or enhancement.

Routine monitoring of the signal intensity of internal standard(s) is an effective way to recognize signal suppression/enhancement in a single patient sample, due to unexpected interfering components of the matrix. Internal standards to be used are those that cover the areas of the elution profile where matrix effects are most pronounced, and that the suitability of these internal standards has been determined (ie, with acceptance limits) during assay development and validation. Internal standard abundance acceptance criteria may be based on signal to noise ratio or may be compared to internal standard abundance in QC samples. As an example, for isotopically-labeled internal standards, if there is poor recovery of the internal standard, a signal to noise ratio greater than 3:1 should still suffice for acceptance of the specimen in question. If recovery of the isotopically-labeled internal standard is considered poor, then an alternate analysis should be considered (eg, the method of standard addition). For analogue-type internal standards, internal standard recovery may be used as a guide for identification of ion suppression/enhancement, although another option, such as the method of standard addition, would be a reasonable alternative. It should be noted that even isotopically-labeled internal standards do not always readily identify ion suppression or enhancement.

Evidence of Compliance:

✓ Records of monitoring of internal standards **OR** records of alternative methods used

REFERENCES

1) Clinical and Laboratory Standards Institute (CLSI). *Liquid Chromatography-Mass Spectrometry Methods.* 2nd ed. CLSI document C62. Clinical and Laboratory Standards Institute, Wayne, PA; 2022.

COLORIMETERS, SPECTROPHOTOMETERS, AND FLUORIMETERS

Inspector Instructions:

READ	• Sampling of colorimeter/spectrophotometer policies and procedures
ASK	• How does your laboratory verify calibration curves?

CBG.17700 Absorbance/Linearity **Phase II**

Absorbance and/or fluorescence linearity is checked and recorded at least annually or as often as specified by the manufacturer, with filters or standard solutions.

Evidence of Compliance:

✓ Records of absorbance and linearity checks at required frequency

CBG.17800 Spectrophotometer Checks **Phase II**