with known concentrations of analyte and by establishing the minimum level at which the analyte can be quantified with acceptable accuracy and precision. The quantitation limit must not be greater than 50% of the specification, where technically feasible.

- Based on Signal-to-Noise Approach

This approach can only be applied to analytical procedures that exhibit baseline noise. Determination of the signal-to-noise ratio is performed by comparing measured signals from samples with known low concentrations of analyte with those of blank samples, and by establishing the minimum concentration at which the analyte can be reliably quantified. A typical signal-to-noise ratio is 10:1.

- Based on Capability of the Instrument

In some cases the instrument itself is the limiting factor for the analysis regardless of the sample. An example of this is an LOD test using an analytical balance. In this case a discussion of the quantitation limit may be constructed in the validation documentation based on the calibration tolerance of the equipment rather than analysis of actual samples. The actual limit of quantitation would still be presented in numerical terms relevant to the assay method based on the discussion.

Another example of this may be for KF titration assays where the ability of the instrument to deliver a minimum amount of titrant would be the limiting factor. It is recommended that experiments to determine this minimum amount of sample should be conducted for the specific instrument model if this approach is taken. The experiment(s) could then be referred to in any validation that utilizes the same model of equipment.

- Based on the Standard Deviation of the Response and the Slope

The quantitation limit (QL) may be expressed as: $QL = 10\ \sigma / S$ where, σ= the deviation of the response; S = the slope of the calibration curve. The slope S may be estimated from the calibration curve of the analyte. The estimate of σ is carried out in a variety of ways including:

  - Based on the Standard Deviation of the Blank:
    Analyzing an appropriate number of blank samples and calculating the standard deviation of these responses and perform measurement of the magnitude of analytical background response.

  - Based on the Calibration Curve:
    A specific calibration curve should be studied using samples containing an analyte in the range of the QL. The residual standard deviation of a regression line or the standard deviation of y-intercepts of regression lines may be used as the standard deviation.
    In all cases, the quantitation limit can be subsequently validated by the analysis of a suitable number of samples known to be near or prepared at the quantitation limit or reporting level.

Two possible approaches include:

A)
Three replicate preparations of a spiked sample are prepared at the quantitation level or reporting level and analyzed. Calculate the % recovery. Calculate the average of the replicates and % RSD.

