Designation: E579 – 04

Standard Test Method for Limit of Detection of Fluorescence of Quinine Sulfate in Solution[1]

This standard is issued under the fixed designation E579; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This test method employs the signal-to-noise ratio to determine the sensitivity of a fluorescence measuring system in testing for the limit of detection (LOD) of quinine sulfate dihydrate in solution. The results obtained with quinine sulfate dihydrate in solution are suitable for specifying instrument performance on samples having excitation and fluorescence bands wider than 10 nm at or near room temperature.

1.1.1 This test method is not intended to be used as (*1*) a rigorous test of performance of instrumentation, or (*2*), to intercompare the quantitative performance of instruments of different design. Intercomparison of the LOD between instruments is commonly expressed as the ratio of the water Raman peak intensity to the root-mean-square (rms) noise as measured on a fluorometer using an excitation wavelength of 350 nm This test method uses the excitation and emission peak wavelengths for quinine sulfate dihydrate in solution, which are approximately 350 nm and 450 nm, respectively..

1.2 This test method has been applied to fluorescence-measuring systems utilizing non-laser, low-energy excitation sources. There is no assurance that extremely intense illumination will not cause photodecomposition[2] of the compound suggested in this test method. For this reason, it is recommended that this test method not be indiscriminately employed with high intensity light sources. This test method is not intended to determine minimum detectable amounts of other materials. If this test method is extended to employ other chemical substances, the user should be aware of the possibility that these other substances may undergo decomposition or adsorption onto containers.

1.3 A typical LOD for conventional fluorometers using this test method is 1 ng of quinine sulfate per mL.

1.4 The suggested shelf life of a 1 mg/mL stock solution of quinine sulfate dihydrate is three months, when stored in the dark in a stoppered glass bottle.

1.5 *This standard does not purport to address all of the safety problems, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 *ASTM Standards:*[3]

E578 Test Method for Linearity of Fluorescence Measuring Systems

3. Summary of Test Method

3.1 To measure the concentration corresponding to the LOD, the fluorescence intensity scale and gain on the detector are adjusted such that noise observed with pure solvent in the sample cell is large enough to measure. The test solution is then diluted until readings on both the test solution and pure solvent can be read at the same intensity, scale, and instrument settings. The concentration corresponding to the limit of detection is that at which the noise intensity, multiplied by three, is equal to the signal intensity.

3.2 This test for limit of detection requires an instrument to meet the following conditions: stable, free of extraneous noise, electrical pickup, and internal stray light. The sample space must be covered to exclude room light. The instrument should be operated according to the manufacturer's recommendations, or, if they are modified, the modifications must be applied consistently to the test for limit of detection and to the analysis for which the test is a requirement, so that levels of performance are comparable for both. All modifications must be included in the report outlined in section 8.

NOTE 1—To obtain the lowest reading (the best instrumental response) for the limit of detection of fluorescent material, a number of precautions must be taken. The quality, condition, and position of the sample cell are most important. The cell must be made of fused silica that does not fluoresce at the excitation wavelength and be free of scratches and marks that scatter light into the fluorescence detection system. Only spectral grade chemicals and solvents (including water) that do not fluoresce

[1] This test method is under the jurisdiction of ASTM Committee E13 on Molecular Spectroscopy and Chromatography and is the direct responsibility of Subcommittee E13.01 on Ultra-Violet, Visible, and Luminescence Spectroscopy.

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[2] Lukasiewicz, R. J., and Fitzgerald, J. M., *Analytical Chemistry*, ANCHA, Vol 45, 1973, p. 511.

[3] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.