



# Certificate of Analysis

## Standard Reference Material<sup>®</sup> 723e

Tris(hydroxymethyl)aminomethane (HOCH<sub>2</sub>)<sub>3</sub>CNH<sub>2</sub>

### Acidimetric Standard

This Standard Reference Material (SRM) consists of highly purified tris(hydroxymethyl)aminomethane (HOCH<sub>2</sub>)<sub>3</sub>CNH<sub>2</sub> [2-amino-2-(hydroxymethyl)-1,3-propanediol; “Tris”; “THAM”], hereafter referred to as Tris. SRM 723e is intended primarily for use in acidimetric standardization. A unit of SRM 723e consists of 50 g in a clear glass bottle.

**Certified Values:** The certified values, reported in Table 1 as a mass fraction of total base expressed as Tris (mass fraction,  $w_{\text{Tris}}$ ) and amount-of-substance content of total base (amount content,  $v_{\text{base}}$ ), are based on coulometric assays of ground and dried material (see “Instructions for Handling, Storage, and Use”), including the effects of air buoyancy.

Table 1. Certified Values for SRM 723e

|   |   |
|---|---|
| Mass Fraction of Tris, $w_{\text{Tris}}$  | 99.9796 % ± 0.0088 %  |
| Amount Content of Base, $v_{\text{base}}$ | 8.253 56 mol · kg <sup>-1</sup> ± 0.000 65 mol · kg <sup>-1</sup> |

The uncertainties in Table 1 are expanded uncertainties,  $U$ , calculated as  $U = ku_c$ , where  $k$  is a coverage factor that governs the confidence level of  $U$  and  $u_c$  is the combined standard uncertainty calculated according to the ISO Guide [2]. The quantity  $u_c$  represents, at the level of one standard deviation, the potential combined effects of the uncertainty arising from instrumental sources, chemical interferences, and uncertainties in fundamental constants, combined with a component of uncertainty reflecting the heterogeneity of the material. The value of  $k$  is calculated from the effective degrees of freedom,  $v_{\text{eff}}$ . The value  $k = 1.98$ , corresponding to  $v_{\text{eff}} = 119$ , was used to obtain the cited value for  $U$  for  $w_{\text{Tris}}$ . The value  $k = 1.99$ , corresponding to  $v_{\text{eff}} = 73$ , was used to obtain the certified value of  $U$  for  $v_{\text{base}}$ . The values of  $k$  were chosen to obtain an approximate 95 % level of confidence.

The certified values were obtained using the 2010 value for the Faraday constant, 96 485.3365 C · mol<sup>-1</sup> [3]. Corrections for air buoyancy were made using 1.35 g · cm<sup>-3</sup> for the density of Tris [5]. The value of  $w_{\text{Tris}}$  was calculated using the molar mass of Tris, 121.135 18 g · mol<sup>-1</sup> (calculated from [4]). The certified values are based on the results of duplicate determinations from each of 10 randomly selected bottles from the entire lot of SRM 723e. Each determination was obtained by coulometric acidimetric back-titration [1] to the inflection point (pH ≈ 5.1 for a 300 mg sample) after addition of excess coulometrically-standardized HCl.

**Expiration of Certification:** The certification of **SRM 723e** is valid, within the measurement uncertainties specified, until **28 December 2019**, provided the SRM is handled and stored in accordance with instructions given in this certificate (see “Instructions for Handling, Storage, and Use”). The certification is nullified if the SRM is damaged, contaminated, or otherwise modified.

**Maintenance of SRM Certification:** NIST will monitor this SRM over the period of its certification. If substantive technical changes occur that affect the certification before the expiration of this certificate, NIST will notify the purchaser. Registration (see attached sheet) will facilitate notification.

Coordination of the technical measurements leading to the certification of SRM 723e was provided by K.W. Pratt of the NIST Analytical Chemistry Division.

Stephen A. Wise, Chief  
Analytical Chemistry Division

Gaithersburg, MD 20899  
Certificate Issue Date: 26 October 2011

Robert L. Watters, Jr., Chief  
Measurement Services Division

Coulometric analyses were performed by K.W. Pratt of the NIST Analytical Chemistry Division.

Statistical consultation was provided by H.-K. Liu of the NIST Statistical Engineering Division.

Support aspects involved in the issuance of this SRM were coordinated through the NIST Measurement Services Division.

**Reference Value:** The reference value and uncertainty (see Table 2) are presented for cases where the additional time and care required for grinding is not warranted and where a significantly larger uncertainty is acceptable for the given use of SRM 723e.

Table 2. Reference Value for SRM 723e

|  |                        |
|--|------------------------|
| Mass Fraction of Tris, $w_{\text{Tris}}$ | 99.925 % $\pm$ 0.054 % |
|--|------------------------|

Owing to the slow loss of occluded water from the material with time, a gradual increase in assay of the unground material is expected. This increase is included in the assigned uncertainty of the reference value.

### INSTRUCTIONS FOR HANDLING, STORAGE, AND USE

**Use:** This SRM is certified for acidimetric assay **ONLY** and is not intended for use in pH standardizations.

**Storage:** This SRM should be stored in its original bottle at room temperature. It must be tightly re-capped after use and protected from moisture and light.

**Handling:** Crush a 1 g to 3 g portion of SRM 723e to a fine powder using an agate or other nonreactive mortar and pestle for 60 s to 90 s. Take special care to crush any larger crystals. Dry the crushed material at room temperature (22 °C to 23 °C) for 24 h in a vacuum desiccator over anhydrous magnesium perchlorate or equivalent. Drying of this material at elevated temperatures is not recommended, owing to the possibility of decomposition.

The reference value is obtained for unground SRM 723e. Eliminate any clumps of crystals present in the source material using gentle force from a spatula, without crushing the crystals themselves. Dry the resulting sample at room temperature (22 °C to 23 °C) for 24 h in a vacuum desiccator over anhydrous magnesium perchlorate or equivalent. Test portions of the resulting material should consist of a representative mixture of smaller and larger crystals. The lower assay for unground material results from occluded water in the crystals [6].

**Homogeneity:** Tests indicate that this SRM is homogeneous within the uncertainty limits for sample sizes greater than 300 mg. Samples less than 300 mg are not recommended in order to avoid possible inhomogeneity with smaller sample sizes.

Test portions of uncrushed material that contain a larger fraction of larger crystals have lower values of  $w_{\text{Tris}}$  and  $v_{\text{base}}$  than the reference value. However, the  $w_{\text{Tris}}$  value for uncrushed, dried material lies within the 95 % confidence interval of the reference value, provided the test portion is a nominally representative sample.

**Source of Material:** The Tris used for this SRM was obtained from a commercial source. The material was examined for compliance with the specification for reagent-grade Tris as specified by the American Chemical Society [7]. The material was found to meet or exceed these specifications in all respects.

## REFERENCES

- [1] Pratt, K.W.; *Automated, high-precision coulometric titrimetry. Part II. Strong and weak acids and bases*; Anal. Chim. Acta, Vol. 289, p. 135–142 (1994).
- [2] JCGM 100:2008; *Evaluation of Measurement Data – Guide to the Expression of Uncertainty in Measurement (ISO GUM 1995 with Minor Corrections)*; Joint Committee for Guides in Metrology (JCGM) (2008); available at [http://www.bipm.org/utls/common/documents/jcgm/JCGM\\_100\\_2008\\_E.pdf](http://www.bipm.org/utls/common/documents/jcgm/JCGM_100_2008_E.pdf) (accessed Sep 2011); see also Taylor, B.N.; Kuyatt, C.E.; *Guidelines for Evaluating and Expressing the Uncertainty of NIST Measurement Results*; NIST Technical Note 1297; U.S. Government Printing Office: Washington, DC (1994); available at <http://physics.nist.gov/Pubs/> (accessed Sep 2011).
- [3] Mohr, P.J.; Taylor, B.N.; and Newell, D.B.; *CODATA recommended values of the fundamental Physical constants: 2006*; Rev. Mod. Phys., Vol. 80 No. 2, pp. 633–730 (2008); 2010 CODATA recommended values available at [http://physics.nist.gov/cgi-bin/cuu/Value?fsearch\\_for=faraday](http://physics.nist.gov/cgi-bin/cuu/Value?fsearch_for=faraday) (accessed Sep 2011); overview of 2010 CODATA available at <http://physics.nist.gov/cuu/Constants/briefOverview2010.pdf> (accessed Sep 2011).
- [4] Coplen, T.B.; *Commission on Isotopic Abundances and Atomic Weights*; Pure & Appl. Chem., Vol. 83, No. 2, pp. 359–396 (2011).
- [5] SRM 723a; *2-Amino-2-(hydroxymethyl)-1,3-propanediol [tris(Hydroxymethyl) aminomethane] (HOCH<sub>2</sub>)<sub>3</sub>CNH<sub>2</sub>*; NIST; U.S. Department of Commerce: Gaithersburg, MD (20 April 1981).
- [6] Koch, W.F.; Biggs, D.L.; Diehl, H.; *Tris(Hydroxymethyl)aminomethane – a Primary Standard?*; Talanta, Vol. 22, pp. 637–640 (1975).
- [7] *Reagent Chemicals*; 8th ed.; American Chemical Society, Washington, DC (1993).

*Users of this SRM should ensure that the Certificate of Analysis in their possession is current. This can be accomplished by contacting the SRM Program: telephone (301) 975-2200; fax (301) 926-4751; e-mail [srminfo@nist.gov](mailto:srminfo@nist.gov); or via the Internet at <http://www.nist.gov/srm>.*