

National Institute of Advanced Industrial Science and Technology

National Metrology Institute of Japan



Reference Material Certificate

NMIJ CRM 6020-a
No. +++

L-Threonine

This certified reference material (CRM) is produced in accordance with the NMIJ's management system and is in compliance with ISO 17034 and ISO/IEC 17025. This CRM is primarily intended for use in calibrating the analytical instruments and preparing the standard solutions used in amino acid analysis. It is also intended for controlling the precision of analysis and for confirming the validity of analytical methods and instruments.

Certified Values

The certified value for the purity (in mass fraction) of L-threonine is given in the table below. The uncertainty of the certified value is the half-width of the expanded uncertainty interval calculated using a coverage factor (k) of 2, which gives a level of confidence of approximately 95 %.

| | CAS No. | Certified value, Mass fraction (kg/kg) | Expanded uncertainty, Mass fraction (kg/kg) |
|---|---------|---|--|
| L-Threonine (<i>(2S, 3R)</i> -2-amino-3-hydroxybutanoic acid) | 72-19-5 | 0.999 | 0.002 |

The purity (in mass fraction) of threonine without enantiomeric separation is given in the table below, where the mass fraction of D-threonine is negligible. The uncertainty of the certified value is the half-width of the expanded uncertainty interval calculated using a coverage factor (k) of 2, which gives a level of confidence of approximately 95 %.

| | Certified value, Mass fraction (kg/kg) | Expanded uncertainty, Mass fraction (kg/kg) |
|--|---|--|
| Threonine (without enantiomeric separation) | 0.999 | 0.002 |

Analysis

The certified value is based on the results of acidimetric titration, nitrogen determination by the Kjeldahl method, and impurity determination by high-performance liquid chromatography (HPLC). Impurities of amino acids were identified and quantified by HPLC with fluorescence detection after derivatization using *o*-phthalaldehyde (OPA) and liquid chromatography/mass spectrometry (LC/MS). D-threonine was quantified by LC/MS using a chiral resolution column. The certified value is the weighted mean of purities determined by acidimetric titration and nitrogen determination. The uncertainty was the combination of standard uncertainties due to purity determinations, differences between the methods, stability, and homogeneity.

Metrological Traceability

The certified value was determined by titrimetry as the primary method of measurement with NMIJ CRM 3001-a (potassium hydrogen phthalate) and NMIJ CRM 3005-a (sodium carbonate) as primary standards and by impurity determination using HPLC calibrated with purity-evaluated amino acids. The certified value is traceable to the International System of Units (SI).

Mutual Recognition Arrangement under Meter Convention

The certified value and expanded uncertainty of this CRM without enantiomeric separation (mass fraction) is consistent with the calibration and measurement capabilities (CMCs) that are included in Appendix C of the Mutual Recognition Arrangement (MRA) drawn up by the International Committee for Weights and Measures (CIPM). Under the MRA, all participating institutes recognize the validity of each other's calibration and measurement certificates for the quantities, ranges and measurement uncertainties specified in Appendix C (as for Appendix C of MRA, see <https://www.bipm.org/kcdb/>).

Expiration of Certification

This certificate is valid for one year from the date of shipment, provided that the material remains unopened and is stored in accordance with the instructions given in this certificate.

Sample Form

This CRM is in the form of a white powder. This CRM of ca. 500 mg in net volume is kept in a brown glass vial and the vial is sealed in an aluminum-laminated plastic bag.

Homogeneity

The homogeneity of the CRM was evaluated by acidimetric titration, analyzing 10 vials randomly selected from 200 vials. The uncertainty of homogeneity is reflected in the uncertainty of the certified value.

Instructions for Storage

This CRM should be stored at a temperature between 15 °C and 25 °C in a clean desiccator and shielded from light.

Instruction for Use

From the homogeneity, a minimum sample mass of 30 mg should be used. This CRM is for laboratory use only and not for in vivo use. The CRM should be used promptly once the vial is opened.

Precautions for Handling

Refer to the safety data sheet (SDS) on this CRM before use.

Preparation

Preparation of the material was performed by Wako Pure Chemical Industries, Ltd. Highly purified L-threonine provided by AJINOMOTO Co., Inc. was bottled into vials under argon atmosphere. Each vial is sealed in an aluminum-laminated bag.

Technical Information

Impurity of amino acid over 0.1 g/kg was not detected at the time of certification.

NMIJ Analysts

The technical manager for this CRM is TAKATSU A. and the production manager is YAMAZAKI T. The analysts are YAMAZAKI T., KATO M., EYAMA S. and YOSHIOKA M.

Information

If substantive technical changes occur that affect the certification before the expiration of this certificate, NMIJ will notify the registered customer. Customer registration on the NMIJ Website (given below) will facilitate notification. Technical reports regarding this CRM can be obtained from the contact details given below.

Reproduction of Certificate

In reproducing this certificate, it should be clearly indicated that the document is a copy.

April 1, 2020

ISHIMURA Kazuhiko
President

National Institute of Advanced Industrial Science and Technology

If you have any questions about this CRM, please contact:
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Revision history

April 1, 2015: "Metrology Management Center" was renamed to "Center for Quality Management of Metrology."

January 28, 2019: The description on "Mutual Recognition Arrangement under Meter Convention" was added.

September 17, 2020 The description in "Expiration of Certification" was changed to "one year from the date of shipment."