National Institute of Advanced Industrial Science and Technology

National Metrology Institute of Japan

Reference Material Certificate

NMIJ CRM 6207-b
No. +++

Dinophysistoxin-1 (DTX1) Standard Solution

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 intended for use in the preparation of standard solution for DTX1 determination in diarrhetic shellfish toxin testing.

Certified Value

The certified value of this CRM is given in the table below. The uncertainty of the certified value is the expanded uncertainty obtained by multiplying the combined standard uncertainty by a coverage factor (k) of 2, and it is the half-width of an interval estimated to have a level of confidence of approximately 95 %.

Substance	CAS No.	Certified value, Mass concentration (μg/mL)	Expanded uncertainty, Mass concentration (μg/mL)
DTX1	81720-10-7	0.958	0.053

Analysis

This CRM was prepared by the gravimetric blending of the solvent (0.5 % (volume fraction) ethanol in methanol) and DTX1 solution, in which mass fraction of DTX1 was determined by qNMR. The mass concentration was calculated from the mass fraction of DTX1, dilution rate at the gravimetric blending and the density of the diluted solution. The standard uncertainties due to qNMR measurement, dilution, homogeneity, stability and the density variation by temperature were combined to evaluate uncertainty of the certified value. The uncertainty of qNMR method evaluated from a collaborative study was included in the uncertainty of qNMR measurement.

Metrological Traceability

In qNMR 1,4-bis(trimethylsilyl)benzene-d_4 calibrated with 3,5-bis(trifluoromethyl)benzoic acid certified reference material (NMIJ CRM 4601-a) was used as an internal standard. The JCSS (Japan Calibration Service System) calibrated balance was used for gravimetric dilution of the DTX1 solution. The densimeter calibrated by water and methanol was used for density measurement. Thus, the certified value is traceable to the International System of Units (SI).

Indicative Values

The indicative values for mass fraction and amount–of-substance concentration of DTX1 at 25 °C are given in the table below. The uncertainty of the indicative 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 %.

Substance		Indicative value	Expanded uncertainty
DTX1	Mass fraction	1.217 μg/g	0.066 μg/g
	Amount-of-substance concentration	1.169 μmol/L	0.066 μmol/L

Mutual Recognition Arrangement under Metre Convention

The certified value of this CRM is recognized for international equivalence based on the Mutual Recognition Arrangement under

the Metre Convention (CIPM MRA). The calibration measurement capability (CMC) of NMIJ related to this CRM is registered in the Key Comparison Database (KCDB) (see https://www.bipm.org/kcdb/) of the International Bureau of Weights and Measures (BIPM).

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.

Description of the material

This CRM is a methanol solution containing ethanol and DTX1. This CRM is in the form of a colorless clear liquid. This CRM of approximately 1 mL in net volume is kept in an amber glass ampule.

Instructions for Storage

This CRM should be stored below –20 °C and shielded from light.

Instructions for Use

Use this CRM at ambient temperature (between 20 °C and 30 °C). Before opening, this CRM should be kept at ambient temperature (between 20 °C and 30 °C) until the temperature reached to an equilibrium. This CRM should be used promptly once the ampule is opened. Evaporation of methanol should be avoided. This CRM is for laboratory use only.

Precautions for Handling

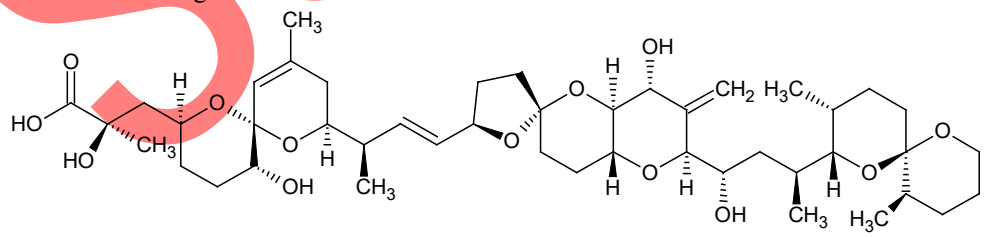
Keep away from heat and ignition source. Handle this CRM in a well-ventilated area. A mask, gloves, and other protective equipment should be worn during handling. Refer to the safety data sheet (SDS) on this CRM before use. Be careful with injury at opening a glass ampoule. Follow local rules or regulations at disposal of this CRM or diluted solutions.

Preparation

DTX1 was produced by a large culture of the toxic dinoflagellate *Prorocentrum lima* and isolated by liquid-liquid partitioning from the cell fraction and the medium separated by centrifuge, and several column chromatography steps at National Research Institute of Fisheries Science (NRIFS). The CRM solution was prepared by dissolving purified DTX1 in methanol containing ethanol and dispensed into amber glass ampoules at NMIJ.

Technical Information

The density of this CRM is 0.7871 g/cm^3 at 25 °C and it will change approximately 0.6 % by 5 °C of temperature shift. The molar mass of DTX1 is 819.03 g/mol. The structure of DTX1 is shown below.



NMIJ Analysts

The technical and production manager for this CRM is NUMATA M. and the analysts are YAMAZAKI T., KAWAGUCHI M., NAKAMURA S. and EYAMA S.

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.

Note

This CRM was developed in collaboration with National Research Institute of Fisheries Science (NRIFS), with support of a grant from Cross-ministerial Strategic Innovation Promotion Program (SIP), Technologies for creating next-generation agriculture, forestry and fisheries (funding agency: Bio-oriented Technology Research Advancement Institution) and Government of Japan and grants from the Project of the NARO Bio-oriented Technology Research Advancement Institution (the special scheme project on vitalizing management entities of agriculture, forestry and fisheries).

Production of this CRM was based on the notable research achievements made by Prof. YASUMOTO Takeshi on discovery of diarrhetic shellfish toxins and production of their standards.

April 1, 2020

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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
March 24, 2022: The description on "Mutual Recognition Arrangement under Metre Convention" was added.