National Research Council Canada | Conseil national de recherches Canada

Certificate of Analysis

Certified Reference Material

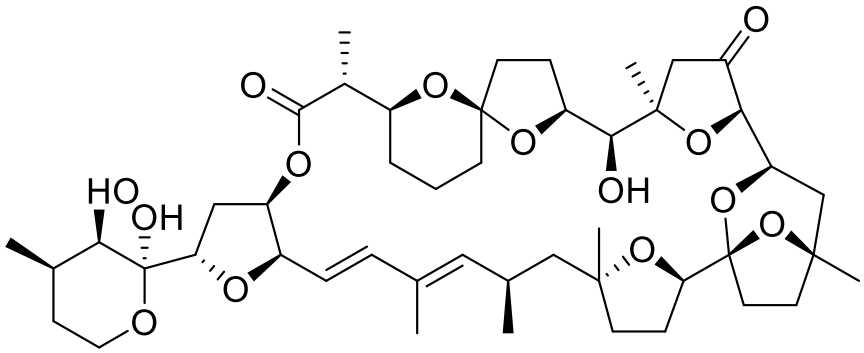
CRM-PTX2-c (Lot #20260206)

Calibration Solution Certified Reference Material for Pectenotoxin-2

Pectenotoxins (PTXs) are a group of regulated cyclic polyether toxins associated with diarrhetic poisoning that are produced by several dinoflagellates [1,2]. CRM-PTX2-c is a certified reference material (CRM) calibration solution of PTX2 in methanol.

Table 1: Certified values and expanded uncertainties (*k* = 2) for CRM-PTX2-c

Compound	Mass fraction µg/g	Mass concentration µg/mL (15-30 °C)	Concentration µmol/L (15-30 °C)
pectenotoxin-2	4.95 ± 0.27	3.90 ± 0.22	4.54 ± 0.25



pectenotoxin-2

CAS registry number: 97564-91-5
InChIKey: PTKFEDGHUVZLPL-LLUYWJARSA-N
Molecular formula: $C_{47}H_{70}O_{14}$
Molar mass: 859.05 g/mol
$[M+NH_4]^+$: *m/z* 876.5104

Period of validity: 1 year from date of sale
Storage conditions: −12 °C or below

DOI: 10.4224/crm.2026.ptx2-c.20260206

Intended use

CRM-PTX2-c is a certified calibration solution for analytical method development and accurate quantitation of PTX2. The concentration is suitable for preparing a dilution series for calibration of instruments such as liquid chromatography–mass spectrometry (LC–MS), and for spiking control samples for recovery experiments.

Preparation of material

Purified PTX2 was acquired from the Cawthron Institute, New Zealand, with final purity assessment performed at the National Research Council of Canada (NRC). The structure and purity of PTX2 were confirmed by ^{1}H NMR, LC–HRMS and LC-CAD [3]. A measured accurate *m*/*z* of 876.5102 (Δ = −0.17 ppm for $C_{47}H_{74}NO_{14}^{+}$) was obtained for the $[M+NH_4]^+$ ion of PTX2 using LC-HRMS (Figure 1A). The product ion spectrum (Figure 1B) matches previous PTX2 CRMs as well as the literature [4,5].

The stock solution was prepared by dissolving the purified PTX2 in CD_3OD for quantitation using ^{1}H NMR [6]. The CRM-PTX2-c solution was prepared by accurately diluting the stock solution in degassed high purity methanol. Aliquots were dispensed into clean argon-filled amber glass ampoules and immediately flame-sealed. Each ampoule contains approximately 0.5 mL.

Characterization of material

The certified values for CRM-PTX2-c (Table 1) are based on results obtained at the NRC by ^{1}H qNMR using benzoic acid for calibration, and LC–MS and LC–CAD using PTX2 for calibration. CRM-PTX2-c contains low levels of structurally related compounds (Figure 2) and several isomers, which combined are approximately 3.6 % of the total PTX2 concentration.

Metrological traceability

Results presented in this certificate are traceable to the International System of Units (SI) through gravimetrically prepared standards of a benzoic acid CRM (NIST PS1) and a PTX2 CRM (NRC CRM-PTX2-b, Lot #20120516).

Homogeneity

A representative number of CRM-PTX2-c ampoules were selected from across the fill series and analyzed by LC–HRMS. No heterogeneity was observed.

Stability

Studies on PTX2 in methanol have demonstrated good stability in sealed ampoules when stored at temperatures of −12 °C or below.

Uncertainty

The expanded uncertainty (*U*) for all values is equal to $U = ku_c$, where u_c is the combined standard uncertainty calculated according to the Joint Committee for Guides in Metrology (JCGM) [7] and *k* is the coverage factor. A coverage factor of $k = 2$ was applied which corresponds to a level of confidence of approximately 95 %.

All reasonable sources of uncertainty related to the certified values in Table 1 were considered. Included in the combined uncertainty estimate are uncertainties in the batch characterization, uncertainties related to possible between-unit variation, and uncertainties related to stability.

Storage

The material shall be stored at −12 °C or below. The ampoule should be stored unopened.

Instructions for handling and use

Prior to opening, each ampoule should be allowed to equilibrate to room temperature and the contents thoroughly mixed. The ampoule should be opened at the pre-scored mark immediately prior to use. The CRM solution should be transferred using calibrated equipment for accuracy. The certified values are only guaranteed if the ampoule is sampled immediately after opening in order to limit solvent evaporation. It is important to note that the volume of the solution is not certified; only the mass fraction and concentration are certified. Therefore, the entire contents of the ampoule should not be diluted to volume.

Repeated sub-sampling and storage of the CRM solution after initial opening may impact certified values. Users shall take responsibility for demonstrating that their sub-sampling and storage procedures do not impact certified values.

Health and safety information

Only qualified personnel should handle the material and appropriate disposal methods should be used. A Safety Data Sheet (SDS) is available at 10.4224/crm.2026.ptx2-c.20260206. For laboratory use only; not for human consumption, therapeutic, drug, household, or any other uses.

Period of validity

The certified values are valid for 1 year from the date of sale, provided the storage and instructions for handling and use specified in this certificate are followed.

Quality Management System

The NRC is Canada's national metrology institute (NMI) and is a signatory of the International Committee for Weights and Measures Mutual Recognition Arrangement (CIPM MRA). The CIPM MRA was developed in a response to a growing need for an open, transparent, and comprehensive scheme to give users reliable quantitative information on the comparability of national metrology services and to provide the technical basis for wider agreements negotiated for international trade, commerce, and regulatory affairs. Our Quality Management System for measurement services and certified reference materials conforms to the requirements of ISO/IEC 17025 and ISO 17034.

References

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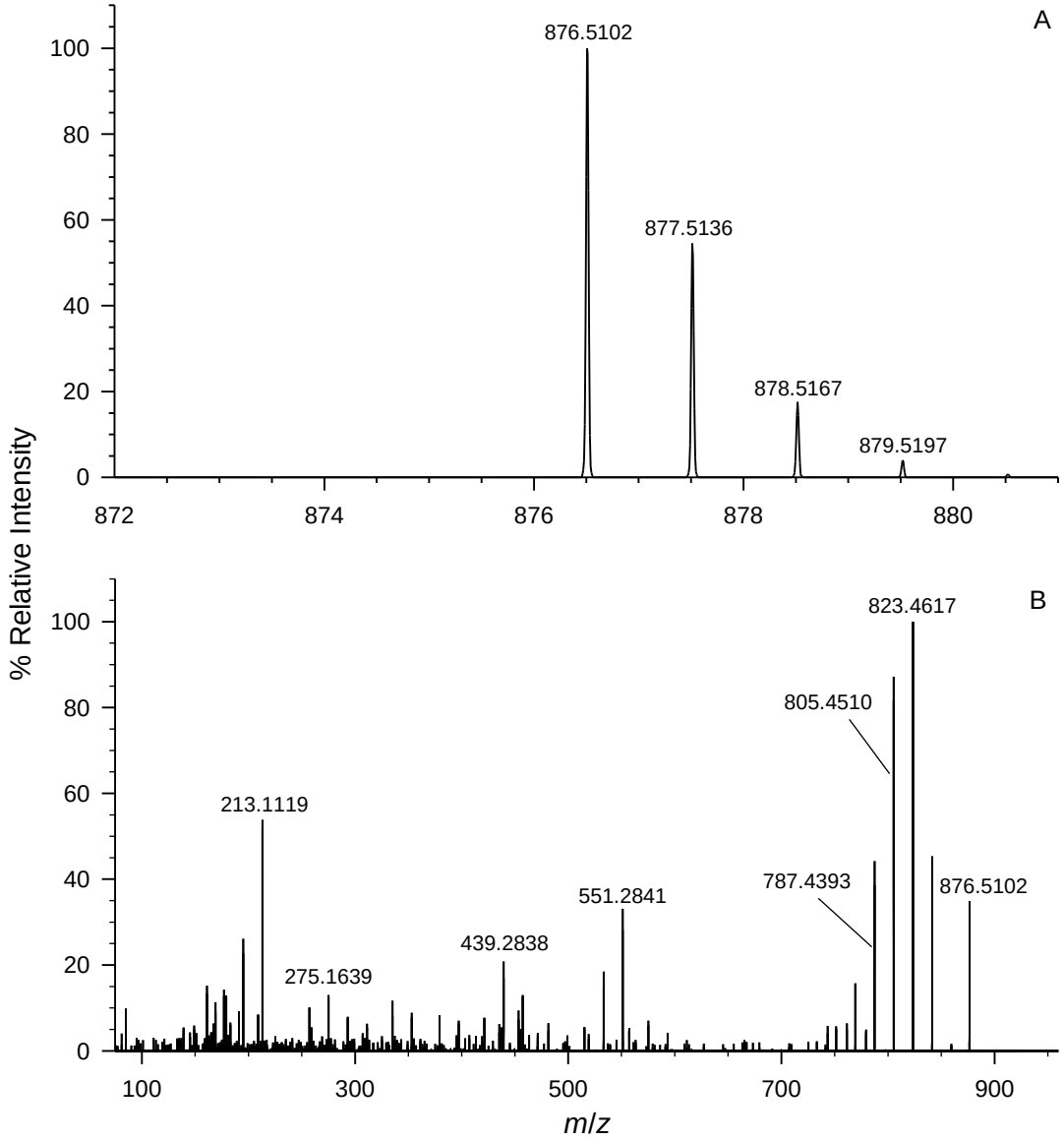

Figure 1: Full-scan (A) and product-ion (MS/MS) (B) spectra from LC-HRMS analysis of PTX2 ($[M+NH_4]^+$ at *m/z* 876.5) in CRM-PTX2-c acquired on a Thermo QExactive-HF. Data was acquired with a resolution setting of 60 000 in positive mode with 3000 V spray voltage, 350 °C capillary temperature and a 300 °C heater temperature. MS/MS data acquired in parallel reaction monitoring scan mode using normalized stepped collision energies of 15, 20 and 25 V.

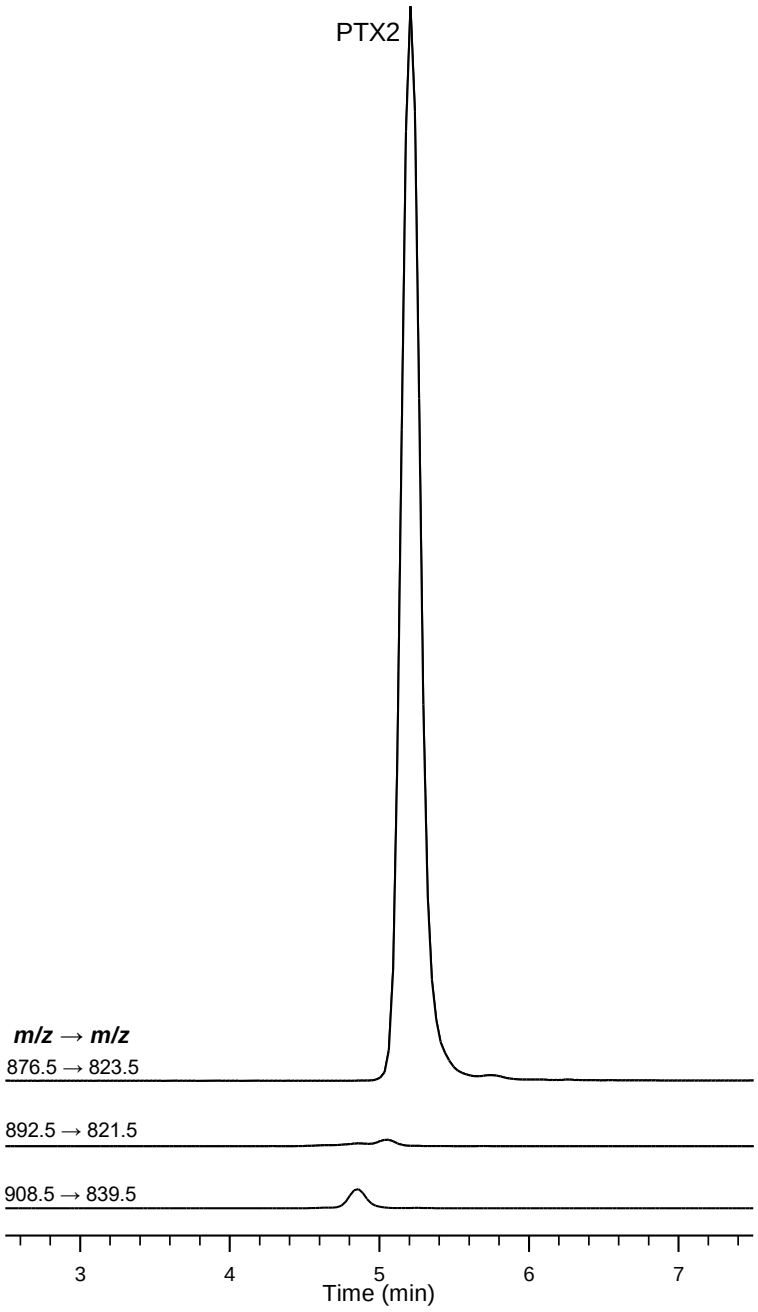

Figure 2: LC–MS/MS analysis of CRM-PTX2-c: Phenomenex Synergi Max-RP 2.5 µm column (2.0 × 50 mm); 2 mM ammonium acetate (pH 6.8) in both deionized water (A) and 95 % acetonitrile (B); gradient: 25 to 100 % B over 5 min, 300 µL/min at 20 °C; injection volume: 5 µL. MS data acquired with positive electrospray ionization (4.5 kV) using selected reaction monitoring scan mode with a declustering potential of 80 eV and optimized collision energies for each transition (35-60 eV).

Authorship

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Approved by: Pearse Mc Carron.

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This certificate is only valid if the corresponding material was obtained directly from the NRC or an authorized reseller. Users should ensure that the certificate they have is current. For updates, please refer to 10.4224/crm.2026.ptx2-c.20260206.

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