



CERTIFIED REFERENCE MATERIAL CERTIFICATE OF ANALYSIS

NMIA MX020: Boldenone and Boldenone Metabolite 1 certified for Carbon Isotope Delta Value

Certified values

Steroid	CAS No.	$\delta^{13}\text{C}_{\text{VPDB-LSVEC}} / \text{‰}$	Coverage Factor (k)	V_{eff}
Boldenone	846-48-0	-30.38 ± 0.29	2.0	53
Boldenone Metabolite 1	10529-96-1	-29.71 ± 0.28	2.0	39

The measurands are the carbon isotope ratio delta values of the stated steroids relative to that embodied in the primary isotopic reference material VPDB. The uncertainties are expanded to provide a level of confidence of 95%.

Expiry: 30 August 2033

Batch No.: 2019.08

Description: 2 mL sealed ampoule containing a dry steroid mixture. The ampoule contains approximately 200 µg of each steroid.

Intended use: Validation and calibration of Gas Chromatography Combustion Isotope Ratio Mass Spectrometry (GC-C-IRMS) for steroid carbon isotope ratio measurements in anti-doping analysis.¹ In-vitro use only.

Instructions for use: Each ampoule must be reconstituted in a minimum of 1 mL of an appropriate solvent such as 2-propanol and allowed to equilibrate for 3 hours before use.

Storage: Store in a refrigerator at 4°C out of direct light.

Metrological traceability: The certified carbon isotope delta value is traceable to the stable carbon isotope ratios of the primary isotopic reference material NBS19 via the secondary isotopic reference materials USGS40 ($\delta^{13}\text{C}_{\text{VPDB}} -26.39 \pm 0.04\text{‰}$) and IAEA-CH-7 ($\delta^{13}\text{C}_{\text{VPDB}} -32.15 \pm 0.05\text{‰}$).²

Stability: The stability of this material in sealed ampoule has been verified under the recommended storage conditions using GC-C-IRMS and will continue to be monitored. The material was also known to be stable in an accelerated stability trial of four weeks at 40°C. Reconstituted material in 2-propanol remained stable for repeated sampling for up to six months under storage conditions when sampled with clean pipette tips. If different reconstitution solvent is employed, the stability of reconstituted material will have to be verified by end user.

Homogeneity: Fifteen ampoules were selected using a stratified sampling plan and each ampoule was analysed twice by GC-C-IRMS. No significant inhomogeneity was detected.

Production: The stock solution for the mixture was prepared by dissolving nominally 0.25 g of pure steroid in 250 mL of a mixture of 2-propanol and methanol prior to dispensing 0.2 mL aliquots into ampoules. The ampoules were vacuum dried for two days then flame sealed under argon and stored at 4°C.

Analytical method: EA-IRMS was employed to assign $\delta^{13}\text{C}_{\text{VPDB}}$ values to the pure steroid materials used for preparation of MX020. Calibration was performed using a two-point normalization approach³ with USGS-40 and IAEA-CH-7 providing metrological traceability to the VPDB scale. The uncertainty contribution from scale calibration and ^{17}O correction by instrument software was estimated in each batch of analysis by using two CRMs (NBS 22 and IAEA-600) as quality control samples.

Measurement uncertainty: Standard uncertainties were estimated and combined as described in the JCGM Guide to the Expression of Uncertainty in Measurement.⁴ The combined standard uncertainties were expanded with coverage factors calculated from degrees of freedom obtained from Welch-Satterthwaite equation. Based on the purity analysis of the starting materials the potential for bias was estimated and included in the expanded measurement uncertainty. The major contributions to the combined measurement uncertainty were potential biases, between-unit homogeneity, measurement reproducibility, potential instability due to transport conditions and uncertainty in the $\delta^{13}\text{C}_{\text{VPDB-LSVEC}}$ values of the calibration materials.



Raluca Iavetz
Manager Chemical Reference Values
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References:

1. WADA. *Technical Document - TD2022IRMS: Detection of Synthetic Forms of Prohibited Substances by GC/C/IRMS, Version 1.0*, 1 January 2022.
2. IUPAC *Technical Report: Assessment of international reference materials for isotope-ratio analysis*, Pure Appl Chem, 2014, 86, 425-467
3. Debajyoti P, Skrzypek G and Istvan F, Normalisation of measured stable isotopic compositions to isotope reference scales – a review, Rapid Comm in Mass Spectrometry, 2007; 21:3006-3014.
4. JCGM, *Evaluation of measurement data — Guide to the expression of uncertainty in measurement*. JCGM100:2008

CIPM MRA Statement: This certificate is consistent with the capabilities that are included in Appendix C of the CIPM MRA drawn up by the CIPM. Under the CIPM 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. The "CIPM MRA Logo" and this statement attest only to the measurement(s) applied for determining the certified values on the certificate (for details see <http://www.bipm.org>).

This Certificate for NMIA MX020 supersedes any issued prior to 14 September 2026.

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