CERTIFICATE OF ANALYSIS

U-[13C34]-Fumonisins B1, B2 Mixture in Acetonitrile/Water (50/50) LCMS grade

This document is designed, and the certified values and uncertainty are determined in accordance with ISO Guide 31, ISO Guide 34, ISO Guide 35, AOAC, and Eurachem/ CITAC Guides.

Description of the Reference Material (RM)

	Product name:	U-[13C34]-Fumonisins B1, B2 Mixture				
	Product number:	FIA000150				
	CAS number:	U-[13C34]-Fumonisin B1	1217458-62-2			
		U-[13C34]-Fumonisin B2	1217481-36-1			
	Lot number:	FB1213C17012602				
	Expiry date:	25-Jan-2025				
	Certified value (s):	U-[13C34]-Fumonisin B1	**10,00**	**±**	**0,19**	**µg/mL**
		U-[13C34]-Fumonisin B2	**10,00**	**±**	**0,18**	**µg/mL**
	Isotope incorporation by mass spectrometry 13C/Molecule	U-[13C34]-Fumonisin B1	**99,3%**			
		U-[13C34]-Fumonisin B2	**99,3%**			
	Physical description:	Clear solution of toxins mixture in Acetonitrile/Water (50/50) LCMS grade				
	Packing	Amber glass vial filled with 5 mL of solution				
	Storage conditions	2-8°C				
	Matrix and starting material:	This material was prepared with/from:				
		Acetonitrile UPLC/MS		Batch:	0001204102BS	
		U-[13C34]-Fumonisin B1		Internal ID:	SS-FB113C	
		U-[13C34]-Fumonisin B2		Internal ID:	SS-FB213C	

Intended use of the RM:

For laboratory use for R&D purposes only. The main purpose of this material is for analytical instrument calibration (e. g. external calibration, standard addition). Not for drug, household or other uses.

Instruction for the correct use of the RM:

The vial should be stored in a dark place at 2-8°C. Before usage of the RM, allow the vial to warm to room temperature. The expiry date of this RM is based on the current knowledge and holds only for proper storage conditions in the originally closed vials / packages. Solutions prepared for calibration purpose should be protected from exposure to light. Discard solutions after use in accordance with appropriate safety regulations for chemical substances.

Hazardous situation:

H225 : Flammable liquid - Category 2 - Highly flammable liquid and vapour
H302 : Acute toxicity - Oral - Category 4 - Harmful if swallowed
H319 : Eye irritation - Category 2 - Causes serious eye irritation

In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible). Avoid exposure. Wear suitable protective clothing.

Safety measures:

Special care must be taken when manipulating this standard. Avoid contact with eyes, skin and clothing. Avoid prolonged or repeated exposure. Use only in a chemical fume hood. Safety shower and eye bath must be near. In case of spills, cover and absorb with an inert dry material such as dry-lime, sand or soda ash and place in an appropriate waste disposal container.
Keep container tightly closed. Do not store in direct sunlight. Keep away from heat, sparks, flame and incompatible material. Storage area should be cool, dry and away from incompatible materials.

Further information:

Further information is available in the MSDS provided along with this certificate. Final users should make their own investigations to determine the suitability of the information for their particular research purposes. In no event the supplier of this RM shall be held liable for any damage resulting from handling or from contact with the product.

Traceability

The certified values are based on the results of analytical techniques previously used for purity assessment of solid mycotoxins. High purity material represents a practical realization of concentration units, through conversion of mass to molar quantity.

Calculation of certified values and associated uncertainties

This calibrant is certified on solution preparation. Toxin is pipetted and diluted in Acetonitrile/Water (50/50) LCMS grade. Mass concentration calculation is based on certified concentration, purity and dilution step.
The pipet was calibrated with traceability to national and international standards (Dakks & ilac-MRA). All weights used for metrological control are connected to national and international standards. The weights are calibrated by an accredited laboratory.

$$C\ (\mu g/mL) = \frac{m \times P}{V}$$

Toxin	Source				Standard uncertainty
U-[13C34]-Fumonisin B1	Purity				98,57
	Liquid solution	concentration	405,98	µg/mL	1,69
	Volumetry procedure	volume	2,46	mL	0,01
	Dilution	volume	100,00	mL	0,06
$Combined_u = \sqrt{\left(\frac{u_p}{P}\right)^2 + \left(\frac{u_{Cm}}{V_{Cm}}\right)^2 + \left(\frac{u_{Vp}}{V_p}\right)^2 + \left(\frac{u_{V1}}{V_1}\right)^2}$					0,01
$Concentration_{Toxin} = \frac{Concentration\ mother}{V_{D1}}$				µg/mL	**10,00**
Total expanded uncertainty (using a coverage factor k=2)					**0,19**

Toxin	Source				Standard uncertainty
U-[13C34]-Fumonisin B2	Purity				98,57
	Liquid solution	concentration	186,46	µg/mL	0,46
	Volumetry procedure	volume	5,36	mL	0,01
	Dilution	volume	100,00	mL	0,06
$Combined_u = \sqrt{\left(\frac{u_p}{P}\right)^2 + \left(\frac{u_{Cm}}{V_{Cm}}\right)^2 + \left(\frac{u_{Vp}}{V_p}\right)^2 + \left(\frac{u_{V1}}{V_1}\right)^2}$					0,01
$Concentration_{Toxin} = \frac{Concentration\ mother}{V_{D1}}$				µg/mL	**10,00**
Total expanded uncertainty (using a coverage factor k=2)					**0,18**

Notes: The purity of the mycotoxin used for this RM was determined by liquid chromatography.
Following the Guide to the Expression of Uncertainty in measurement (GUM) the expanded uncertainty of toxin level is obtained by multiplication with a coverage factor K for which 2 is usually chosen to obtain a confidence level of 95 %.

Carbon 13 calculation

Isotopic incorporation			
Compound	**Isotopic distribution**	**Compound**	**Isotopic distribution**
$^{13}C_{33}$ Fumonisin B1	20,0%	$^{13}C_{33}$ Fumonisin B2	22,0%
$^{13}C_{34}$ Fumonisin B1	80,0%	$^{13}C_{34}$ Fumonisin B2	78,0%
Calculated isotopic incorporation (13C/molecule)	99,3%	***Calculated isotopic incorporation (^{13}C/molecule)***	99,3%

The calculation are based on LC-MS/MS data

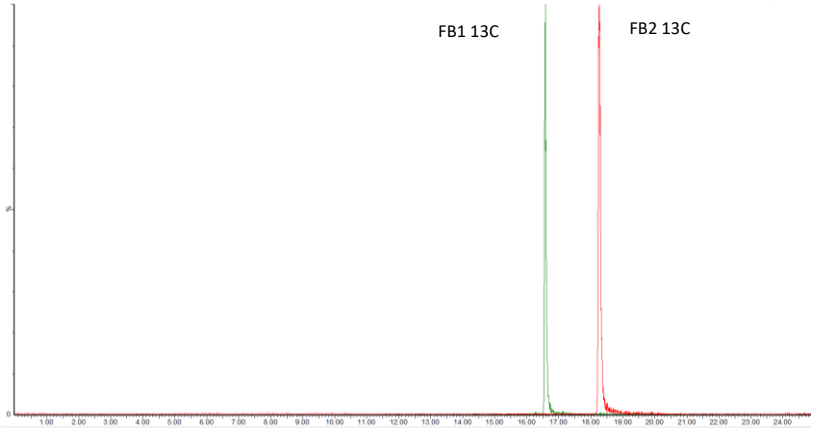
Quality control

Confirmation of the certified concentration by LC-MS/MS

The certified concentrations of the prepared solution was confirmed by LC-MS/MS against a reference batch.

Chromatogram
Chromatographic conditions:

Column:	Acquity UPLC HSS T3 100 x 2,1 mm1,8 µm			
Mobile phase:	MeOH / H2O + 0,1% formic acid + 5mM ammonium acetate /			
Flow mL/min:	0,30			
Temperature °C:	30,00			
Detector	MS/MS			
U-[13C34]-Fumonisin B1	10,03	±	0,11	µg/mL
U-[13C34]-Fumonisin B2	9,87	±	0,60	µg/mL
Mean of 2 replicates measurement against reference batch, confidence interval with P = 95%				



Chromatogram of Toxins

References:

a-ISO GUIDE 31:2015, Reference Materials - Contents of certificates, labels and accompanying documentation.
b-ISO GUIDE 34:2009, General requirements for the competence of reference material producers
c-ISO GUIDE 35:2006, Reference materials - General and Statistical Principles.
d-ISO/IEC Guide 98-3:2008 Uncertainty of measurement-Part 3 : Guide to the expression of uncertainty in measurment (GUM:1995)
e-Eurachem/CITAC guide (2019), Traceability in Chemical Measurement.
f-Eurachem/CITAC guide (2012), Quantifying Uncertainty in Analytical Measurement.
g-AOAC Official Method 970.44-1971 - Preparation of Standards for Mycotoxins.

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Quality Control

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