

Bundesinstitut für Risikobewertung

IRMS – Reference Material and Quality Control

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Classification of Reference Material

- 1) **Primary** (or calibration) materials
- 2) Secondary (or reference) materials
- 3) Inter-laboratory comparison materials
- 4) In-house (or laboratory) standards

Element Ratio	Isotope delta scale
Hydrogen	VSMOW
² H/ ¹ H	(<i>S</i> tandard <i>M</i> ean <i>O</i> cean <i>W</i> ater)
Carbon	VPDB
¹³ C/ ¹² C	(<i>P</i> ee <i>D</i> ee <i>B</i> elemnite)
Nitrogen	Atmospheric Nitrogen
¹⁵ N/ ¹⁴ N	(Air N ₂)
Oxygen ¹⁸ O/ ¹⁶ O	VSMOW



Primary (Calibration) Material I

- Define the δ -scales versus which natural variations in isotopic compositions are expressed
- The IAEA has defined these scales by reference to natural or virtual materials identified by the "V" prefix
- Kept and distributed

by the IAEA (International Atomic Energy Agency), NIST (National Institute of Standards and Technology) and USGS (United States Geological Survey)

- Primary standards are only available in small quantities
- Some original primary standards have become exhausted and have been replaced by similar materials
- Isotopic laboratories calibrate their own working standards using reference material









Primary (Calibration) Material VSMOW δ -scale

✓ VSMOW2:

- VSMOW: exhausted; was prepared by blending distilled ocean water (latitude 0°/ longitude 180°) with small amount of other waters existed
- VSMOW 2: prepared in 1999 by blending three selected fresh water samples, obtained from Lakes in Italy, Israel and Egypt to produce an isotopic composition close to the definition of SMOW (Craig, 1961)
- δ^2 H value was adjusted isotopically by a modified platinum catalytic exchange reaction

✓ SLAP2:

- Lower end of the scale: Standard Light Antarctic Precipitation (SLAP), depleted in heavy isotopes with respect to VSMOW2
- was prepared in 2006 from four selected Antarctic snow/ice samples obtained from the U.S. National Science Foundation's South Pole station (coordinated by T.B. Coplen), from an ice core of the Vostok drilling site (J.M. Petit), from the Dome F drilling site (O. Abe) and from the Dome C drilling site (O. Abe).



Primary (Calibration) Material VSMOW δ-scale II

Table: δ^2 H and δ^{18} O values for the relative difference in hydrogen and oxygen stable amount ratio for the international measurement standards (source: IAEA Reference Sheet RS_VSMOW2_SLAP2_rev1/2017-07-11)

IAEA	NIST	Material	Reference value	Combined	Reference	Combined	
name	code		3	standard	value	standard	
			10 [°] x δ ² H _{VSMOW-SLAP}	uncertainty $10^3 x$ $\delta^2 H_{VSMOW-SLAP}$	10 ³ x δ ¹⁸ Ο _{VSMOW-SLAP}	uncertainty $10^3 x$ $\delta^{18}O_{VSMOW-SLAP}$	
VSMOW2 SLAP2	RM 8535a RM 8537a	Water Water	0 -427.5	0.3 0.3	0 -55.50	0.02 0.02	







Primary (Calibration) Material VPDB δ-scale I

VPDB δ -scale

- PDB: calcium carbonate from a Cretaceous belemnite from the PeeDee formation in South Carolina (USA), exhausted
- Considerably enriched in ¹³C with respect to organic carbon compounds



source: www.wikipedia.org

- VPDB: after exhausting of PDB values were assigned to TS-limestone (NBS 19) and a hypothetical VPDB was created
- Now the calcite (CaCO₃) IAEA 603 is the primary reference material for the PDB-Scale with a δ^{13} C of +2.46 ‰





Secondary (Reference) Material

- Natural or synthetic compounds which have been carefully calibrated versus
 primary calibration material
- δ -values agreed and adopted internationally but have associated uncertainties

Description	Nature	δ ¹³ C ‰	SD
IAEA-CH-3	Cellulose	-24.724	0.041
IAEA-CH-6	Sucrose	-10.449	0.033
IAEA-CH-7	Polyethylene	-32.151	0.050
IAEA-600	Caffeine	-27.771	0.043
USGS-24	Graphite	-16.049	0.035
USGS-40	L-Glutamic acid	-26.389	0.042
USGS-41	L-Glutamic acid	+37.626	0.049
NBS-22	Oil	-30.031	0.043
LSVEC	Lithium carbonate	-46.6	0.2
BCR 656	Ethanol	-26.91	0.07

https://nucleus.iaea.org/rpst/ReferenceProducts/ReferenceMaterials/Stable_Isotopes/2H13C15Nand18O/index.htm



Inter-laboratory Comparison Material

- 1) **Primary** (or calibration) materials
- 2) Secondary (or reference) materials
- 3) Inter-laboratory comparison materials
- 4) In-house (or laboratory) standards

- Materials circulated in inter-laboratory comparison exercises
- δ-values are assigned as the consensus mean of results from participating laboratories, following appropriate statistical treatments
- Participation in **Proficiency Testing Schemes** (PTS)



Inhouse/ Laboratory Standards

Characteristics of Standard Material

- Stable, isotopically homogeneous
- Easy to weigh out (\rightarrow tin capsules) or to inject
- Similar isotopic ratio to the samples
- Similar composition and characteristics to the samples
- Easy to store and to handle, not hazardous
- Non-hygroscopic (for hydrogen and oxygen)
- Low priced

Examples

- ${}^{13}C/{}^{12}C$ in wine \rightarrow wine
- ${}^{13}C/{}^{12}C$ in solids \rightarrow fructose
- ${}^{13}C/{}^{12}C$ and ${}^{15}N/{}^{14}N$ in caseine \rightarrow caseine
- $^{18}\text{O}/^{16}\text{O}$ in wine water \rightarrow wine, light and heavy water



Preparation of "light" and "heavy" water as inhouse standard I

• Fractionation of deionised water

→ Light Water: $\delta^{18}O = -14,6 \%$ vs. VSMOW → Heavy Water: $\delta^{18}O = +15,6 \%$ vs. VSMOW

- Equipment/ tools:
 - ✓ Round bottom flasks 1 I
 - ✓ Distiller
 - ✓ Heating mantles
 - ✓ Boiling chips





Preparation of "light" and heavy water - procedure



Melanie Gimpel, Stable Isotopes Analysis of Wine – Referencing and Quality Control

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Quality Control Charts I

• Example EA liquid ¹³C/¹²C, Wine

ExcelKontrol 2.3 - Mittelwert-Regelkarte

© Dr.-Ing. Michael Koch

	Labor	abor FG 83						Bearbeiter	Panitz			
	Parameter 13C/12C-Verhältnis von Ethanol							Karten-Nr.				
	Norm / SOP PV_48, OIV-MA-AS312-06							Standard	MOW 2017			
	Matrix		Destillat vom Wein					Vorgabewert	-28,34 (Einmessung am IRN			
	Einheit		%0						nach 10 Messungen prüfen!			
	Vorperiode		Kontrollperiod					de				
	Datum Wert		Datum	Wert			Kommentar/AK-Situation/Maßnahme					
1	03.05.2017	-28,4	18.04.2018	-28,51	Prüfen		Messung mit BCR als REF					
2	20.09.2017	-28,44	20.04.2018	-28,52	Prüfen							
3	20.09.2017	-28,43	24.04.2018	-28,37	Prüfen							
4	21.09.2017	-28,46	07.05.2018	-28,4	Prüfen							
5	22.09.2017	-28,39	08.05.2018	-28,45	Prüfen							
6	25.09.2017	-28,46	09.05.2018	-28,4	Prüfen							
7	26.09.2017	-28,43	Ì	^	Prüfen							
8	11.04.2018	-28,51			Prüfen							
9	12.04.2018	-28,51			Prüfen							
10	13.04.2018	-28,5			Prüfen							
11	16.04.2018	-28,46			Prüfen							
12	17.04.2018	-28,48			Prüfen							
13					Prüfen							
14					Prüfen							
15					Prüfen							
16					Prüfen							



Quality Control Charts II

• Example EA liquid ¹³C/¹²C, Wine



• Definition of thresholds: from the official method or from previous period



Quality Control Charts III – Control / Warning situations

- 1 value beyond the control limit or
- 3 values beyond the warning limit

Mittelwertregelkarte - 13C/12C-Verhältnis von Ethanol - PV_48, OIV-MA-AS312-06 - Destillat vom Wein -27,9 8.04.2018 20.04.2018 24.04.2018 07.05.2018 08.05.2018 09.05.2018 20.06.2018 -28 -28,1 -28,2 -28,3 -28,4 -28,5 -28,6 -28,7 -28,8



Quality Control Charts III – Control / Warning situations

• 7 successive values increasing or decreasing





Quality Control Charts III – Control / Warning situations

7 successive values at the same side of the center line •



Mittelwertregelkarte - 13C/12C-Verhältnis von Ethanol - PV 48, OlV-MA-AS312-06 - Destillat vom Wein

obere WG

untere WG

untere KG

MW+1s

---- MW-1s

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Tr BfR

ober KG

Mittelwert

Proficiency Test (PTS) I

Food Analysis using Isotopic Techniques (FIT) – PTS

- Implemented by Eurofins Scientific in 1994
- Since 1999 FIT-PTS was expanded to laboratories around the world (> 50) performing stable isotope measurements on food
- 3 rounds per year (7 samples each round)
- Techniques: IRMS and SNIF-NMR



https://www.eurofins.com/food-and-feed-testing/food-testing-services/authenticity/fit-pts/

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Proficiency Test (PTS) II - Samples

Round	Date (sample beginning of	Deadline (reporting of end of	sample code	Matrix	Analyses					
1	March 17	May 17	17/1/A	Dry wine	SNIF-NMR, C13 & O18-Py (ethanol) O18 (water)					
			17/1/B	Apple Juice	SNIF-NMR, C13 & O18-Py (ethanol), C13 (juice, sugars) O18 (water)					
			17/1/C	Honey	C13 (honey & proteins), N15 (proteins), H2 (proteins), S34 (proteins)					
			17/1/D	Olive oil	C13 O18 & H2 (raw product)					
			17/1/E	Beer	SNIF-NMR, C13 & O18-Py (ethanol) O18 (water), C13 CO2					
			17/1/F	Set of 2 water samples	O18 and H2 (water)					
			17/1/G	Agave Syrup	2H-SNIF-NMR, 13C-SNIF-NMR, C13-IRMS (ethanol), C13-IRMS (raw product)					
2	July 17	September 17	17/2/A	Sweet wine	SNIF-NMR, C13 & O18-Py (ethanol), C13 (glycerol) O18 (water) // Residuals sugards-> SNIFNMR +C13 Ethanol					
			17/2/B	Vanilla extract + ready to inject standard	GC-C-IRMS (vanillin, pHB, van. Acid, pHB acid)					
			17/2/C	Tomato passata	C13, N15, H2, S34 (freeze dried sample/pulp), O18 (water)					
			17/2/D	Wine vinegar + Balsamic vinegar	SNIF-NMR & C13 (acetic acid) O18 (water)					
			17/2/E	Pure ethanol	SNIF-NMR, C13 & O18-Py (ethanol)					
			17/2/F	Orange Juice	SNIF-NMR, C13 & O18-Py (ethanol), , C13 (juice,sugars,pulp) O18 (water)					
			17/2/G	Dairy product	C13, N15, H2, O18, S34 (casein)					
3	November 17	January 18	17/3/A	Fruit Spirit	SNIF-NMR (raw and corrected from impurities), C13 & O18-Py (ethanol), O18 (water)					
			17/3/B	Pineapple juice	SNIF-NMR, C13 & O18-Py (ethanol), C13 (juice,sugars/pulp) O18 (water)					
			17/3/C	Honey	C13 (honey & proteins), N15 (proteins), H2 (proteins), S34 (proteins)					
	4		17/3/D	Pure organic product	C13, H2, N15, O18, S34 (raw product)					
			17/3/E	Pure acetic acid	SNIF-NMR, C13 (raw product)					
			17/3/F	Sparkling wine	SNIF-NMR, C13 & O18-Py (ethanol) O18 (water), C13 CO2					
			17/3/G	Bovine casein	C13, N15, H2, O18, S34 (raw product)					



Proficiency Test (PTS) III - Results

Results of the FIT Proficiency Testing Scheme

2018 Round 1			Product:	Dry Wine					Sa	mple code	: 18/1/A
Lab n°	tQ before distillation	Distillation yield	Ethanol C13	Ethanol O18-Py	Wine O18	Ethanol (D/H)I	(D/H)I SD	Ethanol (D/H)II	(D/H)II SD	Ethanol R	TMU (D/H)
A1	11.95	96.60	-27.23		1.92					2.55	
A2					2.34						
A3	12.00	95.00	-27.26		2.47	100.60	0.35	128.20	0.50	2.56	153.7
A4					2.38					2.53	
A5	11.94	97.20	-27.33		1.95	100.53	0.36	128.47	0.53	2.58	153.7
A6	11.96	98.00	-27.13	2.35		100.78	0.74	128.81	0.72		141.9
A7	11.85	98.13	-27.30		1.97	100.70		129.24			153.7
A8			-27.24		2.18					2.57	
A9			-27.31		1.73					2.57	
A10	12.49	92.90	-27.41		2.27	100.67	0.25	129.22	0.26	2.56	153.7
A11	11.91	99.99	-27.32	-27.14	1.78	100.60	0.21	129.10	0.32	2.56	153.7
A12	11.86	98.00	-27.34		2.16	100.64	0.42	129.01	0.55	2.56	153.7
A13	11.90	98.00	-27.40		1.48	100.15	0.44	128.34	0.40		147.7
A14	11.96	92.00			2.38	100.31	0.36	128.47	0.37	2.57	127.6
A15			-27.53		2.00	100.21	0.20			2.55	
A16	11.94	96.00	-27.30		2.30	100.60	0.52	129.10	0.42	2.57	153.7
A17	12.00	97.80	-27.30		2.10	100.22	0.35	127.60	0.49	2.56	153.7
A18	11.89	99.00	-27.31		2.05	100.33	0.48	128.74	0.64	2.54	153.7
A19	11.97	97.84	-27.21		2.19	100.86	0.27	129.29	0.34	2.56	153.7
A20		94.39	-27.22		2.23	101.24	0.68	128.68	0.74		
A21	11.85	96.30	-27.25		2.33	102.00	0.40	130.70	0.50	2.57	141.9
A22			-27.16								
A23	11.91	96.93	-27.38	27.42	2.42	100.60		129.20		2.55	153.7
A24	11.88	96.10	-27.14								
A25	11.85	98.14	-27.46		2.55	100.32	0.49	128.07	0.60	2.6	155.7
A26	11.87	96.00	-27.16		1.88						
A27	11.74	98.70				100.24	0.72	128.58	0.60		153.7
Size :	20		23		23	19		18			
Mean :	11.94		-27.29		2.13	100.61		128.82			
Robust Mean :	11.92		-27.29		2.14	100.54		128.78			
Standard Deviation :	0.14		0.10		0.26	0.43		0.66			
Robust SD:	0.07		0.11		0.27	0.30		0.52			
Target SD :	0.10		0.15	0.40	0.18	0.37		0.68			
Min :	11.74		-27.53		1.48	100.15		127.60			
Max :	12.49		-27.13		2.55	102.00		130.70			



Proficiency Test (PTS) IV

- Statistical Analysis: evaluation of data is performed according to the ISO/IUPAC/AOAC International Harmonised Protocol for Proficiency Testing of analytical laboratories
- Results are expressed as z-scores:

 $Z = (\underline{x} - \underline{X}) / \underline{\sigma}$

 \underline{x} : reported result from the participating laboratory

X: the assigned or "true value for the analyte being determined, here: taken as the robust mean of all reported results

<u>*o*</u>: target value for the standard deviation, in this exercise the target SD values are derived from recently reported collaborative trial results (AOAC, CEN, ...)or from the robust standard deviation of all reported values



Food Analysis using Isotopic Techniques (FIT) – PTS V

Z-score







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Thank you for your attention

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