

1 Physical model for multi-point normalization of dual-inlet isotope  
2 ratio mass spectrometry isotope data

3

4

5 Abneesh Srivastava

6 *abneesh.srivastava@nist.gov*

7 *Chemical Sciences Division, Materials Measurement Laboratory, National Institute of Standards  
8 and Technology (NIST), 100 Bureau Drive, Gaithersburg, Maryland 20899, United States*

9 **Abstract**

10 A simple model is presented for multi-point normalization of dual-inlet isotope ratio mass  
11 spectrometry (DI-IRMS) isotope data. The model incorporates scale contraction coefficient and  
12 the normalized working reference gas isotope delta value as its two physical parameters. The  
13 model allows the full use of isotope measurement data and outputs the normalized sample isotope  
14 delta value along with the mentioned parameters. The model reduces to the expected linear  
15 behavior on application to natural range CO<sub>2</sub> isotopic composition sample, under typically  
16 observed scale contraction levels. Next, DI-IRMS measurements of the NIST CO<sub>2</sub> gas isotopic  
17 reference materials (RMs) 8562, 8563, and 8564 are used to construct a three-point linear  
18 calibration, spanning 40 ‰ for the  $\delta^{45}\text{CO}_2$  and 20 ‰ for the  $\delta^{46}\text{CO}_2$  raw data. Accuracy of the  
19 regression at the 0.009 ‰ level for  $\delta^{13}\text{C}$  and 0.01 ‰ for  $\delta^{18}\text{O}$  is observed for the three NIST RMs.  
20 The model derived scale contraction term is found to be a more accurate measure of the cross-  
21 contamination in contrast to its end of day measurements by the enriched sample method. The

22 constructed multi-point normalization model is next used to assign  $\delta^{13}\text{C}_{\text{VPDB-CO}_2}$  and  $\delta^{18}\text{O}_{\text{VPDB-CO}_2}$   
23 isotope delta values on the Vienna PeeDee Belmnite-CO<sub>2</sub> (VPDB-CO<sub>2</sub>) scale, for pure CO<sub>2</sub> gas  
24 samples in the natural isotopic range. A Monte Carlo analysis of the uncertainty, including  
25 estimates for the normalization step, is provided to assist future multi-point normalization with  
26 more than three reference points.

27 Keywords: Linear scale normalization; Scale contraction; cross-contamination correction; one-  
28 point; two-point; three-point; multi-point; isotopic reference material; VPDB-CO<sub>2</sub> traceability;  
29  $\delta^{13}\text{C}$ ;  $\delta^{18}\text{O}$ ; DI-IRMS; Monte Carlo simulation; uncertainty; metrology.

## 30 Introduction

31 Scale normalization[1] is routinely used for translating the raw isotope delta measurement  
32 scale to the reference scale. This calibration exercise is necessitated to correct the measurement  
33 bias present in differential (delta) methods on count of instrumental factors.[2] The measured  
34 "scale" is stretched or contracted to match the reference scale and made amenable to comparisons  
35 across measurement campaigns. For the case of CO<sub>2</sub> (carbon dioxide) dual-inlet isotope ratio mass  
36 spectrometry (DI-IRMS) has been the gold standard in relating  $\delta^{45}(\text{CO}_2)_{\text{s/WRG}}$  and  $\delta^{46}(\text{CO}_2)_{\text{s/WRG}}$   
37 (isotope delta of 45/44 and 46/44 isotope ratios) of sample versus working reference gas (WRG)  
38 to sample versus VPDB-CO<sub>2</sub> scale,  $\delta^{45}(\text{CO}_2)_{\text{VPDB}}$  and  $\delta^{46}(\text{CO}_2)_{\text{VPDB}}$  [3]. Normalization schemes  
39 based on single- or multi-point are utilized for data treatment[1]. Each method comes with its  
40 advantages and limitations. While single-point requires a single reference material, it is strongly  
41 dependent on scale correction. The required correction, in the case of CO<sub>2</sub> DI-IRMS measurement  
42 is dominated by scale contraction caused by the cross-contamination of the sample and reference  
43 gas during the differential measurement.[4] Multi-point scale normalization, in practice does not

44 require independent measurement of scale correction but depends on the availability of multiple  
45 RMs across a wide  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$  range. In this regard two carbonate based RMs, NBS-19 ( $\text{CaCO}_3$   
46 with a  $\delta^{13}\text{C}_{\text{VPDB}} = +1.95 \text{‰}$ ) and NIST RM 8545 ( $\text{Li}_2\text{CO}_3$  referred to as LSVEC[5] with a  $\delta^{13}\text{C}_{\text{VPDB}}$   
47  $= -46.6 \text{‰}$ ), have been used historically to achieve two-point normalization for  $\delta^{13}\text{C}_{\text{VPDB-CO}_2}$  [6].  
48 The NIST NBS-19, RM 8545 two-point normalization was adopted in 2006[6] to improve the  
49 consistency in  $\delta^{13}\text{C}$  measurements, and is referred to as the VPDB2006 scale realization.  
50 Limitation on availability (of NBS-19) and stability (of LSVEC) have resulted in efforts to find  
51 appropriate replacements. These include development of a high-purity  $\text{CaCO}_3$  based RM,  
52 USGS44, with a high negative  $\delta^{13}\text{C}_{\text{VPDB2006}}$  value of  $-42.21 \text{‰}$ [7] for  $\delta^{13}\text{C}$  standardization work.  
53 Efforts at the International Atomic Energy Agency (IAEA) have resulted in the development of a  
54 metrologically traceable[8] primary reference material[9] IAEA-603( $\delta^{13}\text{C} = +2.46 \text{‰}$ ,  $\delta^{18}\text{O} = -$   
55  $-2.37 \text{‰}$ ) along with three[10] anchors in the form of IAEA-610, -611 and -612, respectively  
56 (covering  $\delta^{13}\text{C}$  from  $-9.109 \text{‰}$  to  $-36.722 \text{‰}$  and  $\delta^{18}\text{O}$  from  $-4.224 \text{‰}$  to  $-18.834 \text{‰}$ ). (This is  
57 referred to as the VPDB2020[11] scale realization.) In an example of utilizing multiple stable RMs,  
58 the IAEA RMs have been recently used to value assign 25 isotope reference materials. Notably, a  
59 discontinuity of  $0.18 \text{‰}$  at the negative end was seen between the VPDB2020[11] and  
60 VPDB2006 (NBS-19, LSVEC) and scale realizations.

61 Compared to carbonates[12] pure  $\text{CO}_2$  gas[13] isotope reference materials do not require  
62 additional preparations, making them easier to use and adopt for VPDB scale realization. The  
63 availability of multiple pure  $\text{CO}_2$  gas isotope reference materials is currently in the form of NIST  
64 RMs 8562, 8563, and 8564. These RMs originate from three distinct natural sources- limestone,  
65 petroleum, and biomass, and cover  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$  in the  $(-3.72$  to  $-41.59) \text{‰}$  and  $(-33.52$  to  $-10.09) \text{‰}$

66 range, respectively on the VPDB-CO<sub>2</sub> scale[14,15,13]. The NIST RMs, albeit in short supply, are  
67 particularly useful for VPDB scale realization of a range of pure CO<sub>2</sub> gases, for subsequent use in  
68 the development of CO<sub>2</sub> in Air (CO<sub>2</sub>-free) isotope reference mixtures[16]. Such mixtures are  
69 critical for monitoring[17] global trends in the CO<sub>2</sub> isotope delta value using field-based optical  
70 isotope ratio analyzers. In this regard, ongoing comparison studies (CCQM-P204) of CO<sub>2</sub> isotope  
71 ratio standards, coordinated jointly by BIPM (Bureau International des Poids et Mesures) and  
72 IAEA, is an important step[18].

73 Single-point normalization requires scale contraction correction. The correction can be  
74 minimized by a selection of reference material that lies close to the sample isotope delta value, and  
75 instrument parameters optimized to reduce the cross-contamination coefficient. For multi-point  
76 normalization a measurement of the cross-contamination is not required but is inherently present  
77 in the linear statistical model[1], commonly employed for three or more points. However, a  
78 formulation of the multi-point normalization from first principles incorporating a non-linear cross  
79 contamination coefficient term can lead to a fuller representation of the physico-chemical  
80 processes present in the DI-IRMS measurement. Such a treatment is surprisingly missing and easy  
81 to construct for routine use as a quality control and best practice. In this work such a multi-point  
82 normalization model is built, for the case of CO<sub>2</sub> DI-IRMS, from the definition of isotope ratio  
83 and expressed in terms of a non-linear cross-contamination coefficient and working reference gas  
84 isotope delta value on the reference scale. The model is applied to NIST RMs 8562, 8563, and  
85 8564 for a three-point normalization. Cross-contamination coefficients are obtained and compared  
86 to traditional approaches of determining them. The model is then applied to make value  
87 assignments of eight isotopically distinct samples on the VPDB-CO<sub>2</sub> scale, to assist future multi-  
88 point normalization implementation with more than three standards. Additionally, uncertainty

89 evaluation, including its estimation for the normalization step, is provided using Monte Carlo  
90 simulation.

## 91 Experiment

92 The experimental details for DI-IRMS measurements are identical to Srivastava and  
93 Verkouteren[16] and only salient points are emphasized. NIST CO<sub>2</sub> isotope reference materials,  
94 RMs 8562, 8563 and 8564 were cryogenically transferred from respective glass ampoules to  
95 glass bulbs for experimental usage. The nominal isotope delta value for the CO<sub>2</sub> RMs ranged  
96 between (-4 to -42) ‰ for  $\delta^{13}\text{C}_{\text{VPDB-CO}_2}$  and (-10 to -34) ‰ for  $\delta^{18}\text{O}_{\text{VPDB-CO}_2}$ , respectively (for  
97 reference values, see Table SAIV) (for reference values, see Table SAIV). (Note[16]  
98  $\delta^{13}\text{C}_{\text{VPDB-CO}_2} = \delta^{13}\text{C}_{\text{VPDB}}$  and  $\delta^{45}(\text{CO}_2)_{\text{VPDB-CO}_2} = \delta^{45}(\text{CO}_2)_{\text{VPDB}}$ .) One ampoule was used per  
99 reference material. Two sets of four pure CO<sub>2</sub> gas samples each, (all with isotope delta values in  
100 the natural isotope range) designated as  $N_i$  and  $B_i$  ( $i=1$  to 4), respectively were prepared at NIST  
101 (procured from commercial sources) and BIPM. The nominal isotope delta value for the CO<sub>2</sub>  
102 samples ranged between (-1 to -50) ‰ for  $\delta^{13}\text{C}_{\text{VPDB-CO}_2}$  and (-9 to -41) ‰ for  $\delta^{18}\text{O}_{\text{VPDB-CO}_2}$ ,  
103 respectively. The samples were contained in separate single-ended 50 cm<sup>3</sup> volume stainless steel  
104 sample cylinders. The CO<sub>2</sub> was cryogenically transferred from the sample cylinder to the sample  
105 glass bulb, attached to the dual-inlet system manifold. Adequate precaution was taken to  
106 maintain purity between sample handling steps, using helium pressure-vacuum purge cycles and  
107 tight vacuum levels. The experimental measurement configuration and daily measurement  
108 sequence are summarized in Table I and Table II, respectively.

109 Table I Experimental measurement configuration

Parameter	Value
-----------	-------

Instrument	MAT 253 (Thermo)
MS High Voltage	9500 V
Filament Emission Current	1.5 mA
Configuration	Dual-Inlet, CO <sub>2</sub>
Integration time	16 s
Idle Time	15 s
Cycles per acquisition	8
Typical acquisition time	13 min
Bellow/Bellow Master	Sample
VISC* valve turns open	1.5 out of 6
Cup 1 (m/z=44) Resistor	3 x10 <sup>8</sup> Ω
Cup 2 (m/z=45) Resistor	3 x10 <sup>10</sup> Ω
Cup 3 (m/z=46) Resistor	1 x10 <sup>11</sup> Ω
Bellow pressure	30 mbar
Typical Signal level, Cup 2	8500 mV
Typical Background	< 5 mV

\*VISC = variable ion source conductance

110 For each sample-RM sequence run, three continuous acquisitions were measured (each  
 111 comprising 8 sample-reference cycles). To ensure maximum repeatability peak center,  
 112 background, and 'pressure adjust' steps were included in each acquisition. The internal precision  
 113 (standard deviation) over repeated cycles of a single acquisition is 0.008 ‰ for  $\delta^{45}\text{CO}_2$  and  
 114 0.012 ‰ for  $\delta^{46}\text{CO}_2$ ) and found to be consistent with shot-noise predictions. The typical run  
 115 time for a complete sample-RM sequence run was close to 40 minutes.

116

117

118 Table II Daily measurement sequence of dual-inlet sample-reference bellow

Sequence Run No.	Sample bellow	Reference bellow	Acquisitions*
1	WRG <sup>§</sup>	WRG	2
2	Sample1	WRG	3
3	RM1=RM8563	WRG	3
4	Sample2	WRG	3
5	RM2=RM8564	WRG	3
6	Sample3	WRG	3
7	RM3=RM8562	WRG	3
8	Sample4	WRG	3
9	ES <sup>†</sup>	WRG	2

\*Each acquisition has 8 cycle runs, §WRG = Working reference gas, †ES = Enriched sample  
 119 The measured isotope delta values ( $\delta^p(\text{CO}_2)_{\text{sam}/\text{WRG}}$ ,  $\delta^p(\text{CO}_2)_{\text{RM}/\text{WRG}}$ , sam = sample, RM =  
 120 reference material versus WRG= working reference gas, p = 45,46) of the individual cycles were  
 121 pooled (across the constituent acquisitions) to obtain the mean and standard deviation for a given  
 122 sample-RM sequence run. At the end of each sample-RM sequence run, an 'end of day' enriched  
 123 sample, ES (nominal  $\delta^{13}\text{C} = +948 \text{‰}$ ,  $\delta^{18}\text{O} = +1498 \text{‰}$ ) versus working reference gas run  
 124 (Sequence run No. 9) was conducted to obtain the cross-contamination coefficient,  $\eta_{\text{ES}}$  [16].  
 125 The VISC (variable ion source conductance) valve opening, MS high voltage, filament emission  
 126 current values (see Table I) are optimized to maximize ion sensitivity (near 1000 molecules per  
 127 ion for listed conditions). The measurement sequence for each sample set ( $N_i$ ,  $B_i$ ) was repeated  
 128 over 3 days.

## 129 Results & Discussion

### 130 A. Multi-point normalization model

131  
 132 To convert from the raw isotope delta scale (sample versus WRG) to the VPDB-CO<sub>2</sub> isotope  
 133 delta scale (sample versus VPDB-CO<sub>2</sub>) scale a multi-point normalization model is constructed  
 134 from first principles. The stable isotopologue-amount ratios for CO<sub>2</sub> can be expressed as

$$\frac{R_s^p}{R_{\text{VPDB-CO}_2}^p} = \frac{R_s^p}{R_{\text{WRG}}^p} \frac{R_{\text{WRG}}^p}{R_{\text{VPDB-CO}_2}^p} \quad (1)$$

135 where,  $R$ ,  $p$ ,  $s$ ,  $\text{WRG}$ , refer to stable isotopologue-amount ratio relative to mass number 44, mass  
 136 numbers 45 and 46, sample, and working reference gas, respectively. In isotope delta notation  
 137 Eqn. (1) becomes,

$$\delta^p(\text{CO}_2)_{s/\text{VPDB-CO}_2} = \left[ 1 + \delta^p(\text{CO}_2)_{\text{WRG}/\text{VPDB-CO}_2} \right] \delta^p(\text{CO}_2)_{s/\text{WRG}} + \delta^p(\text{CO}_2)_{\text{WRG}/\text{VPDB-CO}_2} \quad (2)$$

138 to give a one parameter linear relationship between  $\delta^p(\text{CO}_2)_{s/\text{VPDB-CO}_2}$  and  $\delta^p(\text{CO}_2)_{s/\text{WRG}}$ . (Eqn.

139 (2) is also used for single-point referencing when working gas isotopic composition,

140  $\delta^p(\text{CO}_2)_{\text{WRG}/\text{VPDB-CO}_2}$  is known[1].) To account for scale contraction in the sample to working

141 reference gas isotope delta value, cross-contamination correction for DI-IRMS proposed by Meijer

142 et al.[4] is used according to

$$\delta_c = \frac{\delta_m}{1 - 2\eta - \eta\delta_m}. \quad (3)$$

143 Here  $\delta_c$  and  $\delta_m$  refer to the corrected and measured isotope delta value, while  $\eta$  is the cross-

144 contamination coefficient. The  $\eta\delta_m$  term imparts non-linearity to the cross-contamination

145 correction and is relevant only for significantly enriched samples. Inserting the correction Eqn. (2)

146 becomes

$$\delta^p(\text{CO}_2)_{s/\text{VPDB-CO}_2} = \left[ 1 + \delta^p(\text{CO}_2)_{\text{WRG}/\text{VPDB-CO}_2} \right] \delta^p(\text{CO}_2)_{s/\text{WRG-c}} + \delta^p(\text{CO}_2)_{\text{WRG}/\text{VPDB-CO}_2} \quad (4)$$

$$\delta^p(\text{CO}_2)_{s/\text{VPDB-CO}_2} = (1 + \delta^p(\text{CO}_2)_{\text{WRG}/\text{VPDB-CO}_2}) \left( \frac{\delta^p(\text{CO}_2)_{s/\text{WRG-m}}}{1 - 2\eta - \eta\delta^p(\text{CO}_2)_{s/\text{WRG-m}}} \right) + \delta^p(\text{CO}_2)_{\text{WRG}/\text{VPDB-CO}_2} \quad (5)$$

147 where  $\delta^p(\text{CO}_2)_{s/\text{WRG-c}}$ ,  $\delta^p(\text{CO}_2)_{s/\text{WRG-m}}$  are the corrected and measured values of the sample versus

148 working reference gas isotope delta,  $\delta^p(\text{CO}_2)_{s/\text{WRG}}$ . It is to be noted that  $\delta^p(\text{CO}_2)_{s/\text{VPDB-CO}_2}$  is non-

149 linear with respect to the measured isotope delta value,  $\delta^p(\text{CO}_2)_{s/\text{WRG-m}}$ .

150 Eqn. (5) represents a complete model for multi-point scale normalization and includes two physical  
 151 parameters,  $\delta^p(\text{CO}_2)_{\text{WRG/VPDB-CO}_2}$  and  $\eta$  which can be obtained by fitting the model to the ' $N$ '  
 152 reference material  $ref_j$  data points, ( $\delta^p(\text{CO}_2)_{ref_j/\text{WRG-m}} = x_j = \text{measured}$ ;  $\delta^p(\text{CO}_2)_{ref_j/\text{VPDB-CO}_2} = y_j$   
 153 = known,  $j=1\dots N$ ). This treatment makes use of the available reference material measurement  
 154 data and does not rely on separate measurements of the cross-contamination coefficient. A  
 155 simulation of the model is presented in Fig. 1 for  $\delta^p(\text{CO}_2)_{ref_j/\text{WRG-m}} = x_j$ ,  $\delta^p(\text{CO}_2)_{ref_j/\text{VPDB-CO}_2} = y_j$   
 156,  $\eta = 0.3$ ,  $\delta^p(\text{CO}_2)_{\text{WRG/VPDB-CO}_2} = -20\text{‰}$ ,  $N = 101$  points between  $\delta^p(\text{CO}_2)_{ref_j/\text{WRG-m}} -500\text{‰}$  to 500  
 157 ‰. Scale contraction is clearly visible on the raw measurement isotope scale when comparing (  
 158  $\delta^p(\text{CO}_2)_{ref_j/\text{WRG-m}} = x_j$ ;  $\delta^p(\text{CO}_2)_{ref_j/\text{VPDB-CO}_2} = y_j$ ) and ( $\delta^p(\text{CO}_2)_{ref_j/\text{WRG-c}} = x_j$ ;  
 159  $\delta^p(\text{CO}_2)_{ref_j/\text{VPDB-CO}_2} = y_j$ ) plots.

160 When the term  $\eta\delta_m^p$  in Eqn. (3) becomes negligible, as in the case of natural samples, contraction  
 161 correction becomes linear[16,4] in  $\delta$  (see SC, supporting information) and Eqn. (5) reduces to

$$\delta^p(\text{CO}_2)_{s/\text{VPDB-CO}_2} = \left(1 + \delta^p(\text{CO}_2)_{\text{WRG/VPDB-CO}_2}\right) (1 + 2\eta) \delta^p(\text{CO}_2)_{s/\text{WRG-m}} + \delta^p(\text{CO}_2)_{\text{WRG/VPDB-CO}_2} \quad (6)$$

162 This is similar in form to the linear relationship used for multi-point normalization,

$$\delta^p(\text{CO}_2)_{s/\text{VPDB-CO}_2} = m\delta^p(\text{CO}_2)_{s/\text{WRG}} + b \quad (7)$$

163, where  $m$  is the slope and  $b$  the intercept =  $\delta^p(\text{CO}_2)_{\text{WRG/VPDB-CO}_2}$ . The slope is referred to as the  
 164 'expansion factor' and the intercept, as the 'additive correction factor'[1]. For flow based  
 165 differential measurements, as in continuous-flow and elemental-analyzer isotope ratio mass  
 166 spectrometry (CF-IRMS and EA-IRMS),  $m$  and  $b$  in Eqn. (7) are treated as independent. The linear

167 multi-point normalization serves as a statistical model. However, as Eqn. (8) shows, in the case of  
168 DI-IRMS, the terms  $m$  and  $b$  are related as,

$$(1 + b)(1 + 2\eta) = m \quad (8)$$

169 In this study the constraint introduced in Eqn. (8) is included to maximize the information from  
170 the DI-IRMS measurement data, lending a physical meaning to the calibration slope. The linear  
171 slope is influenced by both the isotope delta value (of the working gas) and the measurement of  
172 the cross-contamination coefficient. While the former, for typically used depleted (negative valued  
173  $\delta^{13}\text{C}_{\text{WRG/VPDB-CO}_2}, \delta^{18}\text{O}_{\text{WRG/VPDB-CO}_2}$ ) working reference gases, leads to shrinking, the second factor  
174 leads to stretching of the measurement scale when anchored to the VPDB scale.

175 Next, the residual error introduced by the linear approximation of the cross-contamination  
176 correction is simulated in Fig. 2, for  $\text{CO}_2$  isotope delta values in the natural range as a function of  
177 three cross-contamination coefficient values ( $\eta = 2 \times 10^{-2}, 2 \times 10^{-3}, 2 \times 10^{-4}$ , chosen to capture their  
178 observable range reported in literature[7,10,19,3]). The simulated data is derived using the full  
179 non-linear model given in Eqn. (5). Residual values for the predicted isotope delta values approach  
180 0.01 ‰, 0.001 ‰ and 0.0001 ‰ for  $\eta = 2 \times 10^{-2}, 2 \times 10^{-3}$ , and  $2 \times 10^{-4}$ , respectively at  $\delta^p(\text{CO}_2)_{\text{WRG-m}}$   
181  $= \pm 50$  ‰ for linear approximation (Eqn. (6)) of the simulated data. At  $\eta$  levels  $< 2 \times 10^{-3}$  linear  
182 approximation of the model, used in rest of the paper, is adequate to achieve 0.001 ‰ accuracy for  
183  $\delta^{45}\text{CO}_2$  and  $\delta^{46}\text{CO}_2$  in the natural isotopic range.

## 184 B. Three-point scale normalization of NIST RMs: 8562, 8563, 8564

185

186 Six independent determinations of the three-point normalization of NIST RMs are made in  
187 terms of the described model. As illustrated in Fig. 3 the  $\delta^{45,46}\text{CO}_2$  values lie in the natural

188 isotopic range and the model falls within the linear regime. The absolute value of the residuals  
 189 for  $\delta^{45}\text{CO}_2$  and  $\delta^{46}\text{CO}_2$  are seen to be within 0.004 ‰ and 0.009 ‰, respectively, representing  
 190 excellent fit. The fit residuals provide a quantitative measure of the normalization accuracy  
 191 relative to the best estimate of the NIST RM true values (see Table SAIV, supporting  
 192 information). The predicted  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  NIST RM values are within 0.009 ‰ and 0.01‰ of  
 193 their true values, well within the reported standard uncertainties of the RMs. Consistent with  
 194 calibration using Eqn. 6 the cross-contamination coefficient,  $\eta^p = \eta_{\text{model}}^p$  and the isotope delta  
 195 value of the working reference gas relative to the VPDB- $\text{CO}_2$ ,  $\delta^p(\text{CO}_2)_{\text{WRG/VPDB-CO}_2}$  is obtained  
 196 for  $p = 45$  and  $46$ . (Complete regression parameters, its residuals are provided in the supporting  
 197 information SA.) The cross-contamination coefficient values obtained in the fit are tabulated in  
 198 Table III for further discussion.

199 Table III Cross-contamination values for  $\delta^{45}\text{CO}_2$ ,  $\delta^{46}\text{CO}_2$  using three approaches, with  
 200 uncertainties in brackets for the model method

$\eta_{45}/10^{-3}$			$\eta_{46}/10^{-3}$		
Model	PW*	ES <sup>§</sup>	Model	PW	ES
2.12(0.02)	2.13	0.64	0.34(0.15)	0.36	2.21
2.19(0.06)	2.22	0.67	0.65(0.27)	0.60	2.21
2.10(0.00)	2.10	0.69	0.85(0.25)	0.89	2.32
2.12(0.10)	2.16	0.69	1.97(0.24)	2.01	2.28
2.04(0.04)	2.05	0.68	1.95(0.35)	2.01	2.25
2.13(0.04)	2.15	0.69	2.51(0.08)	2.52	2.26

\*PW = Pairwise is calculated value for RM pair with the largest isotope delta difference. <sup>§</sup>ES= Enriched sample values are based on measurement of enriched sample versus working reference gas.

201  
 202 Two other determinations of the cross-contamination coefficient are provided. One is based on  
 203 measurements of the enriched sample versus working reference gas method ( $\eta^p = \eta_{ES}^p$ ) at the end

204 of the day. The second value is the calculated pairwise value for RM pair with the largest isotope  
 205 delta value difference. This value is computed to mimic the pairwise measurement method of  
 206 cross-contamination determination between two isotopically different CO<sub>2</sub> gases with large  
 207 isotope delta value difference for  $p = 45, 46$ . The model derived cross-contaminations are observed  
 208 to be a close match to the pairwise calculated values, within the fitting errors of the  $\eta_{\text{model}}^p$ . In the  
 209 limit, of the fit residuals approaching zero the two would become identical. In contrast the model  
 210 and pairwise determined cross-contamination coefficients are 3.1(0.1) and 0.6(0.4) times relative  
 211 to the enriched sample method, expressed as mean(uncertainty) for  $\eta_{45}$  and  $\eta_{46}$ , respectively.  
 212 Additionally, the model scale-contraction parameters show larger variability for  $\delta^{46}\text{CO}_2$   
 213 compared to  $\delta^{45}\text{CO}_2$  with mean (standard deviation) values of  $\eta_{\text{model}}^{46} = 1.38(0.80) \times 10^{-3}$  and  $\eta_{\text{model}}^{45}$   
 214  $= 2.12(0.05) \times 10^{-3}$  across the six measurement sequences. This suggests scale contraction during  
 215 sample and RM measurement is not entirely represented by the end of day enriched sample cross-  
 216 contamination correction and would require consideration of additional sampling, memory effects  
 217 during the measurement sequence. Rather, the model derived value is a more realistic capture of  
 218 the contraction during the sample-RM measurement sequence.

### 219 C. Sample VPDB-CO<sub>2</sub> scale realization

220

221 The model regression parameters are used to obtain VPDB-CO<sub>2</sub> scale isotope delta values in  
 222 the analyzed samples ( $N_i, B_i, i = 1$  to 4). Sample  $\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$ ,  $\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$  values are obtained  
 223 using appropriate <sup>17</sup>O interference correction parameters ( $^{13}R_{\text{VPDB}} = 0.011180(28)$ ,  $^{17}R_{\text{VPDB-CO}_2} =$   
 224  $0.0003931(9)$ ,  $\lambda = 0.528$ ,  $K = 0.01022461$ ,  $^{18}R_{\text{VPDB-CO}_2} = 0.00208835$ , uncertainties reported at 95%  
 225 confidence interval) and  $\delta^{45}(\text{CO}_2)_{\text{s/VPDB-CO}_2}$ ,  $\delta^{46}(\text{CO}_2)_{\text{s/VPDB-CO}_2}$  to  $\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$ ,  $\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$

226 linearization approximation provided by Brand, Assonov, Coplen[20]. The uncertainty due to 17-  
 227 O correction parameter selection was negligible,  $\leq \pm 0.001$  ‰ across the sample range. As shown  
 228 in Fig. 4, the derived values span a wide range, (-50 to -1) ‰, (-41 to -9) ‰ for  $\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$  and  
 229  $\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$ , respectively. The  $\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$ ,  $\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$  values obtained involve  
 230 extrapolation of  $(N_1, N_4, B_1, B_2)$ ,  $(N_4, B_1, B_2)$  for  $\delta^{45}\text{CO}_2$ ,  $\delta^{46}\text{CO}_2$  normalization. The uncertainty  
 231 of each sample comprises Type A and Type B and is tabulated along with the mean isotope delta  
 232 values in Table IV.

233 Table IV. Mean and uncertainty values of the sample  $\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$  and  $\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$

Sample	$\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$ ‰	$\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$ ‰	$u_A^p$ ‰		$u_B^p$ ‰		$u_c^p$ ‰	
	(mean of 3 days)		$p=13$	$p=18$	$p=13$	$p=18$	$p=13$	$p=18$
$N_1$	-2.85	-16.51	0.012	0.014	0.03	0.13	0.03	0.13
$N_2$	-10.98	-12.27	0.012	0.019	0.02	0.16	0.03	0.16
$N_3$	-39.89	-33.73	0.013	0.024	0.04	0.23	0.04	0.23
$N_4$	-50.42	-41.33	0.015	0.027	0.06	0.32	0.06	0.32
$B_1$	-1.48	-9.43	0.011	0.021	0.02	0.15	0.03	0.15
$B_2$	-43.11	-36.00	0.021	0.030	0.04	0.24	0.05	0.24
$B_3$	-8.84	-14.34	0.011	0.020	0.03	0.19	0.04	0.19
$B_4$	-34.30	-30.18	0.017	0.033	0.03	0.18	0.04	0.18

$u_{A(B)}^p$  = Type A(B) standard uncertainty,  $u_c^p = \sqrt{(u_A^p)^2 + (u_B^p)^2}$  = combined standard uncertainty,  $p=13,$   
 18

234  
 235 The Type A standard uncertainty contribution forms the minor component of the overall budget  
 236 and lies in the (0.011 to 0.021) ‰ and (0.014 to 0.030) ‰ range for  $\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$  and  $\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$   
 237 , respectively. The reported uncertainty estimates are obtained by the Monte Carlo simulation  
 238 method and include its evaluation for the normalization step (see supporting information, SB).

## 239 Conclusion

240 A simple two parameter physical measurement model is presented for multi-point  
241 normalization of DI-IRMS isotope data. The contribution of scale contraction, in terms of cross-  
242 contamination coefficient, is explicitly included to derive its value from the model regression  
243 under experimental conditions. This approach represents the full use of the measurement data and  
244 gives the working gas isotope delta on the VPDB-CO<sub>2</sub> scale as its second parameter, for the case  
245 of CO<sub>2</sub> DI-IRMS. The commonly used linear model for continuous-flow isotope ratio  
246 measurement calibration is refined, in terms of cross-contamination correction, to obtain a physical  
247 interpretation of the model parameters for the case of DI-IRMS. In contrast to the end of day  
248 measurement of cross-contamination coefficient by enriched sample method the model derived  
249 values mimic experimental scale contraction conditions better. Consequently, a pairwise method  
250 or a multi-point normalization model-based method, as established in this work, is potentially a  
251 better alternative to the end of day enriched sample method for correcting the cross-contamination  
252 introduced bias in DI-IRMS measurements.

253 The model is applied to the NIST RMs 8562, 8563, and 8564 to construct scale normalization  
254 with accuracy at the 0.009 ‰ level for  $\delta^{13}\text{C}$  and 0.01 ‰ for  $\delta^{18}\text{O}$ . The three-point normalization  
255 is used to make VPDB-CO<sub>2</sub> isotope delta value assignments of eight isotopically distinct CO<sub>2</sub>  
256 samples in the natural range. A full uncertainty analysis using the Monte Carlo method is also  
257 provided. Future studies will allow usage of these CO<sub>2</sub> samples as standards for multi-point  
258 normalization with more than three points.

## 259 Supporting information

SA Three-point Normalization model regression results

SB Uncertainty calculation using Monte Carlo simulation

SC Cross-contamination linear approximation

## 260 Acknowledgement

261 Kimberly Harris (NIST) and Dr. Joële Viallon (BIPM) are thanked for the CO<sub>2</sub> samples.

262 Disclaimer: Commercial instrument identified as part of the experimental procedure is not a  
263 recommendation nor an endorsement by the National Institute of Standards and Technology.

## 264 Compliance with ethical standards

### 265 Conflict of interest

266 The author declares no conflict of interest.

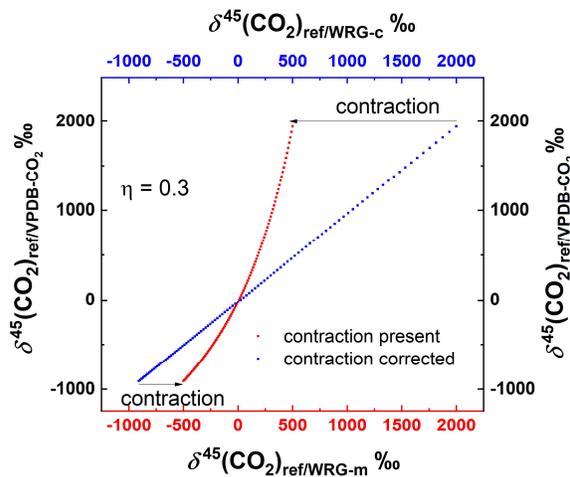
## 267 References

- 268 1. Paul D, Skrzypek G, Forizs I (2007) Normalization of measured stable isotopic compositions to isotope  
269 reference scales - a review. *Rapid Communications in Mass Spectrometry* 21 (18):3006-3014.  
270 doi:10.1002/rcm.3185
- 271 2. Verkouteren RM, Allison CE, Studley SA, Leckrone KJ (2003) Isotopic metrology of carbon dioxide.  
272 I. Interlaboratory comparison and empirical modeling of inlet equilibration time, inlet pressure, and ion  
273 source conductance. *Rapid Communications in Mass Spectrometry* 17 (8):771-776. doi:10.1002/rcm.905
- 274 3. Werner RA, Brand WA (2001) Referencing strategies and techniques in stable isotope ratio analysis.  
275 *Rapid Communications in Mass Spectrometry* 15 (7):501-519. doi:10.1002/rcm.258
- 276 4. Meijer HAJ, Neubert REM, Visser GH (2000) Cross contamination in dual inlet isotope ratio mass  
277 spectrometers. *International Journal of Mass Spectrometry* 198 (1-2):45-61. doi:10.1016/s1387-  
278 3806(99)00266-3
- 279 5. Flesch G, Anderson Jr A, Svec H (1973) A secondary isotopic standard for 6Li/7Li determinations.  
280 *International Journal of Mass Spectrometry and Ion Physics* 12 (3):265-272
- 281 6. Coplen TB, Brand WA, Gehre M, Groning M, Meijer HAJ, Toman B, Verkouteren RM (2006) New  
282 guidelines for delta C-13 measurements. *Analytical Chemistry* 78 (7):2439-2441. doi:10.1021/ac052027c

- 283 7. Qi H, Moossen H, Meijer HA, Coplen TB, Aerts-Bijma AT, Reid L, Geilmann H, Richter J, Rothe M,  
284 Brand WA (2021) USGS44, a new high-purity calcium carbonate reference material for  $\delta^{13}\text{C}$   
285 measurements. *Rapid Communications in Mass Spectrometry* 35 (4):e9006
- 286 8. Assonov S, Fajgelj A, Allison C, Gröning M (2021) On the metrological traceability and hierarchy of  
287 stable isotope reference materials aimed at realisation of the VPDB scale: Revision of the VPDB  $\delta^{13}\text{C}$   
288 scale based on multipoint scale-anchoring RMs. *Rapid Communications in Mass Spectrometry* 35  
289 (8):e9018
- 290 9. Assonov S, Groening M, Fajgelj A, Hélie JF, Hillaire-Marcel C (2020) Preparation and characterisation  
291 of IAEA-603, a new primary reference material aimed at the VPDB scale realisation for  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$   
292 determination. *Rapid Communications in Mass Spectrometry* 34 (20):e8867
- 293 10. Assonov S, Fajgelj A, Hélie JF, Allison C, Gröning M (2021) Characterisation of new reference  
294 materials IAEA-610, IAEA-611 and IAEA-612 aimed at the VPDB  $\delta^{13}\text{C}$  scale realisation with small  
295 uncertainty. *Rapid Communications in Mass Spectrometry* 35 (7):e9014
- 296 11. Hélie J-F, Adamowicz-Walczak A, Middlestead P, Chartrand MM, Mester Z, Meija J (2021)  
297 Discontinuity in the Realization of the Vienna Peedee Belemnite Carbon Isotope Ratio Scale. *Analytical*  
298 *Chemistry* 93 (31):10740-10743
- 299 12. Brand WA, Huang L, Mukai H, Chivulescu A, Richter JM, Rothe M (2009) How well do we know  
300 VPDB? Variability of  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  in  $\text{CO}_2$  generated from NBS19-calcite. *Rapid Communications in*  
301 *Mass Spectrometry: An International Journal Devoted to the Rapid Dissemination of Up-to-the-Minute*  
302 *Research in Mass Spectrometry* 23 (6):915-926
- 303 13. Verkouteren RM (1999) Preparation, characterization, and value assignment of carbon dioxide  
304 isotopic reference materials: RMs 8562, 8563, and 8564. *Analytical Chemistry* 71 (20):4740-4746.  
305 doi:10.1021/ac990233c
- 306 14. Report of Investigation, Reference Materials 8562-8564 (2018). National Institute of Standards and  
307 Technology, Gaithersburg, MD
- 308 15. Verkouteren RM, Klinedinst DB (2004) Value Assignment and Uncertainty Estimation of Selected  
309 Light Stable Isotope Reference Materials: RMs 8543-8545, RMs 8562-8564, and RM 8566. 2004 edn.  
310 NIST Special Publication 260-149 2004 Edition,
- 311 16. Srivastava A, Verkouteren RM (2018) Metrology for stable isotope reference materials: C-13/C-12  
312 and O-18/O-16 isotope ratio value assignment of pure carbon dioxide gas samples on the Vienna PeeDee  
313 Belemnite- $\text{CO}_2$  scale using dual-inlet mass spectrometry. *Analytical and Bioanalytical Chemistry* 410  
314 (17):4153-4163. doi:10.1007/s00216-018-1064-0

- 315 17. Brewer PJ, Kim JS, Lee S, Tarasova OA, Viallon J, Flores E, Wielgosz RI, Shimosaka T, Assonov S,  
 316 Allison CE (2019) Advances in reference materials and measurement techniques for greenhouse gas  
 317 atmospheric observations. *Metrologia* 56 (3):034006
- 318 18. Wielgosz R, Tavella P, Judge S, Stock M, Milton M (2021) News from the BIPM laboratories—2020.  
 319 *Metrologia* 58 (1):015018
- 320 19. Verkouteren RM, Assonov S, Klinedinst DB, Brand WA (2003) Isotopic metrology of carbon  
 321 dioxide. II. Effects of ion source materials, conductance, emission, and accelerating voltage on dual-inlet  
 322 cross contamination. *Rapid Communications in Mass Spectrometry* 17 (8):777-782. doi:10.1002/rcm.906
- 323 20. Brand WA, Assonov SS, Coplen TB (2010) Correction for the O-17 interference in delta(C-13)  
 324 measurements when analyzing CO2 with stable isotope mass spectrometry (IUPAC Technical Report).  
 325 *Pure and Applied Chemistry* 82 (8):1719-1733. doi:10.1351/pac-rep-09-01-05

326



327

328

329

330

331

332 Fig. 1 Simulation of multi-point normalization of reference versus working gas,  $\delta^{45}(\text{CO}_2)_{\text{ref}/\text{WRG}}$ ,

333 scale to the reference versus VPDB-CO<sub>2</sub>,  $\delta^{45}(\text{CO}_2)_{\text{ref}/\text{VPDB-CO}_2}$ , scale. Scale contraction and its

334 correction are shown for an assumed cross-contamination coefficient,  $\eta = 0.3$ . The value of

335  $\delta^{45}(\text{CO}_2)_{\text{WRG/VPDB-CO}_2}$  is assumed to be -20 ‰. The terms  $\delta^{45}(\text{CO}_2)_{\text{ref/WRG-m}}$ ,  $\delta^{45}(\text{CO}_2)_{\text{ref/WRG-c}}$   
 336 represent the measured (bottom x-axis) and scale contraction corrected (top x-axis) isotope delta  
 337 value of reference versus working reference gas. See text for details.

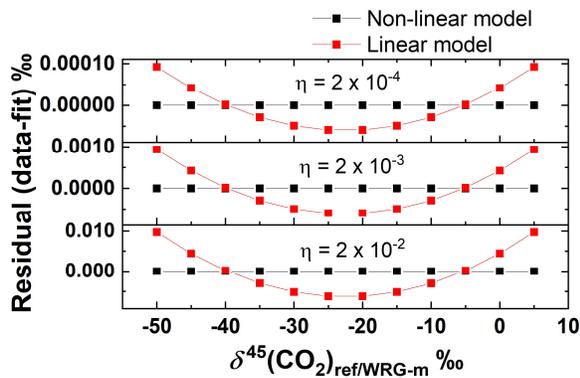
338

339

340

341

342



343

344

345

346

347

348

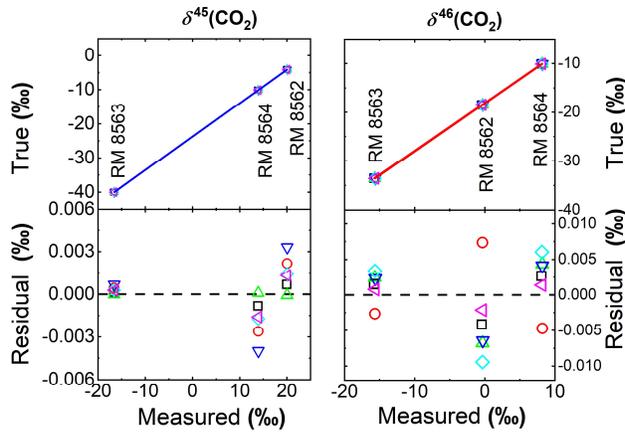
349

350

351

352

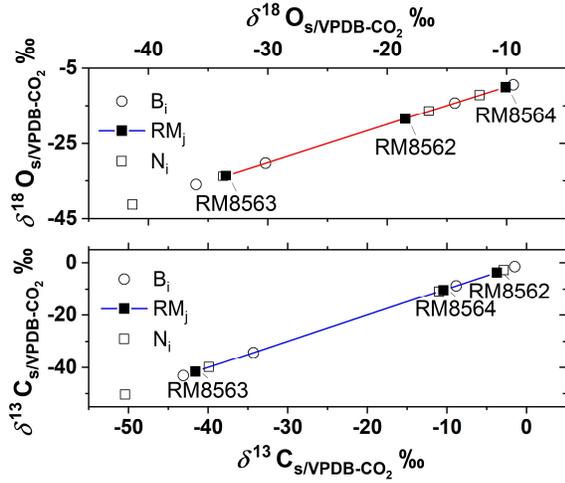
353 Fig. 2 Residual dependence of non-linear and linear fit of the normalization model in the natural  
 354 range of CO<sub>2</sub> isotope delta values as a function of cross-contamination coefficient. See text for  
 355 details.  
 356



357  
 358  
 359  
 360  
 361  
 362  
 363  
 364  
 365 Fig. 3 Multi-point normalization (shown as linear fit) of the measured,  $\delta^p(\text{CO}_2)_{\text{ref}/\text{WRG-m}}$  (x-axis)  
 366 to the true (known best estimate) VPDB-CO<sub>2</sub> isotope scale,  $\delta^p(\text{CO}_2)_{\text{ref}/\text{VPDB-CO}_2}$  (y-axis) for NIST  
 367 RMs 8562, 8563, and 8564 (p = 45, 46). Residuals, representing deviation of the true (known

368 best estimates) from their fit predicted values have a maximum absolute value of 0.004 ‰ and  
369 0.009 ‰ for  $\delta^p(\text{CO}_2)_{\text{ref/VPDB-CO}_2}$ ,  $p = 45$  and 46.

370



371

372

373

374

375

376

377

378

379

380

381

382

383 Fig. 4 Relative position of the sample ( $N_i, B_i$ ) and NIST RMs  $\delta^{13}\text{C}_{\text{s/VPDB-CO}_2}$ ,  $\delta^{18}\text{O}_{\text{s/VPDB-CO}_2}$ . See

384 text for details.

Submitted Version