

1 *In situ* speciation of dissolved inorganic antimony in
2 surface waters and sediment porewaters: development
3 of a thiol-based diffusive gradients in thin films
4 technique for Sb^{III}

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22 **Abstract**

23 Antimony is a priority environmental contaminant typically present as either the trivalent (Sb^{III}) or the
24 pentavalent (Sb^{V}) oxidation state in aquatic systems. Both the toxicity and mobility of antimony are
25 affected by its speciation, and thus the accurate measurement of antimony speciation is essential for
26 investigating the behaviour of this contaminant in aquatic systems. Here we present a diffusive
27 gradients in thin films (DGT) technique, which utilises a binding layer containing a thiol-based
28 adsorbent (3-mercaptopropyl functionalised silica gel), for the selective measurement of Sb^{III} in surface
29 waters and sediment porewaters. We also evaluated the Metsorb DGT technique, which has been
30 previously reported to accurately measure Sb^{V} , for its ability to accumulate Sb^{III} and thus allow the
31 measurement of total inorganic antimony. Both the mercapto-silica and Metsorb DGT techniques
32 showed a high affinity for Sb^{III} , with uptake efficiencies $>97\%$. Elution efficiencies of $86.9 \pm 2.6\%$ and
33 $88.1 \pm 1.2\%$ were obtained for mercapato-silica and Metsorb, respectively, with $1 \text{ mol L}^{-1} \text{ H}_2\text{O}_2$ in 1
34 $\text{mol L}^{-1} \text{ NaOH}$. The accumulation of Sb^{III} by these DGT techniques was linear with time ($R^2 > 0.99$)
35 and unaffected by pH (4.07 – 8.05), ionic strength ($0.001 \text{ mol L}^{-1} - 1.0 \text{ mol L}^{-1} \text{ NaCl}$), bicarbonate (1
36 $\text{mmol L}^{-1} - 15 \text{ mmol L}^{-1}$), and an artificial seawater matrix (pH 8.34; salinity 34.8). Finally, the
37 mercapto-silica DGT technique was applied to measure Sb^{III} and As^{III} in the porewater of a
38 contaminated freshwater sediment at high-resolution.

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47 **Introduction**

48 Antimony is a metalloid widely exploited for industrial applications, with global mine production in
49 2013 estimated at 159,000 metric tons.¹ The extensive industrial use of antimony has resulted in it
50 being listed as a priority pollutant by the United States Environmental Protection Agency.² Antimony
51 is primarily used in flame retardants, lead alloys for lead-acid batteries, and the production of glass,
52 ceramics, and plastics.¹ There have also been numerous reports of severe antimony contamination
53 surrounding mining and processing facilities, and at shooting ranges (see Filella *et al.*³ and Wilson *et*
54 *al.*⁴ for comprehensive reviews on antimony occurrence in the environment). Ultimately, inorganic
55 contaminants like antimony are transported to aquatic systems, where they often accumulate in
56 sediments and are subject to complex biogeochemical processes that determine their environmental
57 behaviour and fate. The study of antimony biogeochemistry is complicated by the fact that it exists in
58 different oxidation states; typically neutral $\text{Sb}(\text{OH})_3$ (Sb^{III}) or oxyanionic $\text{Sb}(\text{OH})_6^-$ (Sb^{V}), which can
59 impact both its affinity for various solid-phases and its toxicity to biota.⁵

60

61 The most reliable way to determine antimony speciation in natural waters is by on-site or *in situ*
62 separation of the species of interest. On-site approaches are limited to solid phase extraction (SPE) of
63 the sample, typically with SPE cartridges through which the sample is passed.⁶ Such methods have the
64 advantage of avoiding sample preservation, storage, and transport, and can be done relatively quickly
65 and inexpensively. Unfortunately, the methods that are available are not suitable for marine waters and
66 require volumes of sample (typically millilitres) not often obtainable when extracting sediment
67 porewater.⁶ Passive *in situ* sampling methods for antimony speciation could provide the same benefits
68 as on-site methods, with the additional advantage of integrating concentrations over the deployment
69 time of the sampling device, thereby providing a more representative measurement of antimony
70 concentrations in dynamic systems.⁷

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72 Diffusive gradients in thin films (DGT) is an *in situ*, passive sampling technique that provides a time-
73 weighted average analyte concentration in surface and interstitial waters (see Zhang and Davison⁸ for a
74 recent review on the application of DGT for measurements in water, sediment and soils).^{9, 10} Solutes
75 diffuse through a diffusive layer of known thickness, typically a polyacrylamide hydrogel overlain by a
76 protective membrane, and are immobilised within a binding layer (often hydrogel-based) that contains
77 a binding agent with high affinity for the analyte of interest. The analyte concentration in the bulk
78 solution is calculated from the mass of analyte accumulated by the binding layer (determined after
79 elution and analysis of the analyte), the deployment time, the area of sampler exposed to the bulk
80 solution, the thickness of the diffusive layer, and the diffusion coefficient of the analyte in the diffusive
81 layer.¹⁰ Numerous binding agents have been used in DGT to permit the measurement of a wide variety
82 of analytes, including, for example: cationic trace metals,¹¹ oxyanionic metalloids,^{12, 13} sulfide,¹⁴
83 phosphate,^{15, 16} and ammonium.¹⁷ Additionally, the application of selective binding agents that only
84 accumulate a specific analyte oxidation state, have expanded the DGT technique to allow the *in situ*
85 measurement of chemical speciation. Recently, Fan *et al.*¹⁸ described the application of a 3-
86 mercaptopropyl functionalised silica-based DGT technique for measuring Sb^{III} in wastewater and
87 freshwater, although this method was not tested in seawater and required a complicated elution
88 procedure. Panther *et al.*¹³ described a Metsorb DGT technique for measuring Sb^V in natural waters,
89 although the ability of this technique to measure Sb^{III} was not tested. Bennett *et al.*¹⁹ described the use
90 of a binding layer containing 3-mercaptopropyl functionalised silica gel that selectively accumulated
91 As^{III} in the presence of As^V. When combined with a DGT technique for measuring total inorganic
92 arsenic, such as that described by Panther *et al.*²⁰ or Bennett *et al.*,²¹ this approach allowed the
93 quantification of As^{III} and As^V in surface waters and sediment porewaters.²²

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95 Here we present a diffusive gradients in thin films (DGT) technique for the selective *in situ*
96 measurement of Sb^{III} in fresh and marine surface waters and sediment porewaters. The method utilizes

97 a thiol-based binding layer (3-mercaptopropyl functionalized silica gel immobilized in a
98 polyacrylamide gel matrix) with high selectivity for Sb^{III} over Sb^V. The existing Metsorb DGT
99 technique that was previously evaluated for measuring Sb^V,¹³ was also tested for its ability to
100 accurately measure Sb^{III}, which would allow this technique to be used to measure total inorganic
101 antimony. The mercapto-silica and Metsorb DGT methods were tested at a range of pH, ionic strength,
102 bicarbonate concentration, and in artificial seawater. Finally, the new mercapto-silica DGT method was
103 applied to determining high-resolution porewater profiles of Sb^{III} and As^{III} in a contaminated
104 freshwater sediment.

105

106 **Experimental**

107 **Reagents, materials and solutions.** All plasticware was washed in 5-10% HNO₃ or HCl and then
108 thoroughly rinsed in deionized water (18.2 MΩcm⁻¹; Millipore) before use. All reagents were analytical
109 reagent grade or better. Stock solutions (100 mg L⁻¹) of antimonite (Sb^{III}) and antimonate (Sb^V) were
110 prepared by dissolution of potassium antimony(III) tartrate and potassium hexahydroxoantimonate(V),
111 respectively, in deionized water. Stock solutions were stored at 4°C and diluted immediately before
112 use. DGT sampler housings were purchased from DGT Research Ltd (<http://www.dgtresearch.com>). 3-
113 mercaptopropyl functionalised silica gel (SiliaMetS Thiol) was purchased from Silicycle (Quebec,
114 Canada), and Metsorb HMRP (50 μm powder) was obtained from Graver Technologies (Delaware,
115 United States of America).

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117 **Antimony analysis.** Antimony (m/z 121) was measured by inductively coupled plasma – mass
118 spectrometry (ICP-MS; Agilent 7900). The instrument was equipped with an octopole
119 collision/reaction cell, which was operated in helium collision mode to reduce potential polyatomic
120 interferences. All solutions for analysis were prepared in 2% (v/v) ultrapure HNO₃ (sub-boiling
121 distilled). External calibration solutions and quality control solutions were prepared from separate

122 commercially available standard solutions (High Purity Standards). A certified reference material
123 (Riverine Water, SLRS-5; National Research Council Canada) was analysed in each analytical run and
124 had an average recovery of 102.1 ± 6.0 % ($n = 9$). The stability of inorganic antimony species in the
125 experimental solutions was monitored using a solid phase extraction (SPE) procedure followed by ICP-
126 MS analysis, as described previously,⁶ Speciation changes were negligible over the experimental
127 durations used in this study. Samples taken for speciation analysis were processed immediately, to
128 avoid possible speciation changes due to storage.

129

130 **DGT samplers.** DGT samplers with binding layers containing either 3-mercaptopropyl functionalized
131 silica gel (mercapto-silica) or Metsorb were prepared as described previously.^{19, 21} The diffusive layers
132 of the samplers consisted of a polyacrylamide hydrogel, prepared as described previously,^{19, 21} and a
133 protective $0.45 \mu\text{m}$ pore-size cellulose nitrate (Merck Millipore Ltd.) or polysulfone (Pall Corp.) filter
134 membrane, with a total thickness of 0.088 ± 0.04 cm. Samplers were stored in sealed polyethylene bags
135 at 4°C prior to use. Following retrieval of DGT samplers from deployment solutions they were
136 thoroughly rinsed in deionised water and stored at 4°C prior to elution. DGT binding gels were eluted
137 in 1 mL of $1 \text{ mol L}^{-1} \text{H}_2\text{O}_2 / 1 \text{ mol L}^{-1} \text{NaOH}$ for at least 24 hours, before being diluted 20-fold in 2%
138 HNO_3 for ICP-MS analysis, except where stated otherwise.

139

140 **Uptake and elution.** Mercapto-silica and Metsorb binding gel discs were exposed in triplicate to
141 known volumes of $100 \mu\text{g L}^{-1} \text{Sb}^{\text{III}}$ (pH 6, $0.01 \text{ mol L}^{-1} \text{NaCl}$) for at least 12 hours. Samples of the
142 solution before and after exposure of the binding gel discs were collected for ICP-MS analysis to
143 determine the mass of antimony accumulated by the binding gels. Elution of antimony from the binding
144 gels was tested by exposing them to 1 mL of one of the following solutions for at least 24 h: 0.1 mol L^{-1}
145 KIO_3 in $1 \text{ mol L}^{-1} \text{HNO}_3$ and $1 \text{ mol L}^{-1} \text{HCl}$; or $1 \text{ mol L}^{-1} \text{H}_2\text{O}_2$ in $1 \text{ mol L}^{-1} \text{NaOH}$. Eluents were
146 diluted and analysed by ICP-MS to determine the efficiency of elution procedures.

147

148 **Accumulation over time.** To determine if the accumulation of Sb^{III} by Mercapto-silica and Metsorb
149 DGT samplers was linear, three sets of triplicate samplers were exposed to a well-stirred solution
150 containing 100 µg L⁻¹ Sb^{III} in 0.01 mol L⁻¹ NaCl (buffered at pH 7.79 ± 0.05 with 5.0 x 10⁻⁴ mol L⁻¹
151 NaHCO₃) for 10, 18, and 26 hours. A similar experiment was done to determine if Sb^V was
152 accumulated by mercapto-silica DGT, where a single set of triplicate samplers was exposed to a well-
153 stirred solution of 100 µg L⁻¹ Sb^V in 0.01 mol L⁻¹ NaCl (buffered at pH 7.90 ± 0.02 with 5.0 x 10⁻⁴ mol
154 L⁻¹ NaHCO₃) for 26 hours. The average temperature of the experimental solutions was 24.8 ± 0.4°C.
155 Samples of the solution were taken at the beginning of the experiment, and each time DGT samplers
156 were removed, for ICP-MS analysis of total antimony and for antimony speciation analysis by SPE-
157 ICP-MS.

158

159 **Effect of pH, ionic strength, bicarbonate concentration, and artificial seawater.** The performance
160 of mercapto-silica and Metsorb DGT were assessed for measuring Sb^{III} over a range of pH, ionic
161 strengths, bicarbonate concentrations, and in artificial seawater. DGT samplers of each type were
162 deployed in triplicate in well-stirred solutions containing 100 µg L⁻¹ Sb^{III} for 24 – 26 hours (exact time
163 recorded). Solutions of different pH were prepared in 0.01 mol L⁻¹ NaCl through the addition of HCl
164 (for pH < 6) or by buffering with 5.0 x 10⁻⁴ mol L⁻¹ NaHCO₃ (for pH > 7). Solutions of different ionic
165 strength were prepared with NaCl at 0.001, 0.01, 0.1, and 1.0 mol L⁻¹ (buffered at pH 7.82 ± 0.12 with
166 5.0 x 10⁻⁴ mol L⁻¹ NaHCO₃). Solutions of different bicarbonate concentration were prepared in 0.01
167 mol L⁻¹ NaCl with NaHCO₃ at 0.001, 0.005, 0.01 and 0.015 mol L⁻¹. Due to the buffering of solution
168 pH by NaHCO₃, the pH of each solution was slightly different: pH 8.20 ± 0.03 for 0.001 mol L⁻¹
169 NaHCO₃; pH 8.65 ± 0.22 for 0.005 mol L⁻¹ NaHCO₃; pH 8.81 ± 0.18 for 0.010 mol L⁻¹ NaHCO₃; and
170 pH 8.79 ± 0.18 for 0.015 mol L⁻¹ NaHCO₃. Artificial seawater was prepared as described by Grasshoff
171 *et al.*,²³ and had a pH of 8.34 ± 0.01 and a salinity of 34.8.

172

173 **Capacity.** The linear accumulation capacity of mercapto-silica DGT and Metsorb DGT samplers was
174 determined by exposing ten samplers of each type to a well-stirred solution of $22 \text{ mg L}^{-1} \text{ Sb}^{\text{III}}$ and
175 removing one sampler of each type every 30 minutes. The mass accumulated by each sampler was
176 compared to the theoretical mass calculated from the diffusion coefficient of Sb^{III} , the deployment time,
177 the area of sampler exposed to solution and the diffusive layer thickness (as per the DGT equation).¹⁰
178 Deviation of the measured mass from the theoretical mass indicates that the linear accumulation
179 capacity has been exceeded, and the sampler is no longer accumulating Sb^{III} quantitatively.

180

181 **High-resolution, porewater measurements of Sb^{III} and As^{III} with mercapto-silica DGT.** Sediment
182 was collected from an antimony and arsenic contaminated wetland in Urunga, New South Wales,
183 Australia ($30^{\circ}30'12.4''\text{S } 153^{\circ}00'46.1''\text{E}$), and transported to the laboratory. The sediment was sieved to
184 $< 1 \text{ mm}$ and homogenised, before being placed into a large Perspex mesocosm and allowed to stabilise
185 for at least one month in a constant temperature room at $21 \pm 0.2^{\circ}\text{C}$. The overlying water of the
186 mesocosm was constantly sparged with air to ensure oxic conditions. A DGT sediment sampler
187 containing a mercapto-silica binding layer for the selective measurement of Sb^{III} and As^{III} was prepared
188 with a diffusive layer consisting of a 0.08 cm -thick bisacrylamide-crosslinked polyacrylamide hydrogel
189 covered with a 0.01 cm -thick $0.45 \mu\text{m}$ -poresize cellulose nitrate filter membrane. The sampler was
190 deoxygenated in $0.01 \text{ mol L}^{-1} \text{ NaNO}_3$ sparged with nitrogen gas for at least 2 hours prior to
191 deployment. The sampler was deployed in the sediment for 24 hours, after which time it was removed,
192 thoroughly rinsed with deionised water, and stored at 4°C prior to processing and analysis. The
193 mercapto-silica binding gel was cut from the DGT sampler and washed for at least 1 hour in 50 mL of
194 deionised water to remove unbound solutes, before being laterally sliced at 2.5 mm intervals and each
195 slice eluted in 0.5 mL of $0.01 \text{ mol L}^{-1} \text{ KIO}_3$ in $1 \text{ mol L}^{-1} \text{ HNO}_3$ and HCl for 24 hours. A sub-sample of

196 each eluent was diluted 20-fold in 2% HNO₃ prior to analysis by ICP-MS. A blank DGT sampler was
197 processed in the same way to determine the limit of detection (3σ).

198

199 **Results and Discussion**

200 **Uptake and elution.** Uptake efficiencies of Sb^{III} by mercapto-silica and Metsorb binding gels were
201 99.5% and 97.9%, respectively, which provided initial verification that these binding gels had a high
202 affinity for Sb^{III} and were suitable for further testing. Elution of Sb^{III} from Metsorb was done using 1
203 mol L⁻¹ H₂O₂ / 1 mol L⁻¹ NaOH, as previously described for the elution of Sb^V from Metsorb binding
204 gels,¹³ and resulted in an Sb^{III} elution efficiency of 88.1 ± 1.2%. The elution of Sb^{III} from mercapto-
205 silica binding gels was tested with 0.1 mol L⁻¹ KIO₃ / 1 mol L⁻¹ HNO₃ / 1 mol L⁻¹ HCl (which is similar
206 to the procedure previously reported for the elution of As^{III} from the same binding layer¹⁹); and 1 mol
207 L⁻¹ H₂O₂ / 1 mol L⁻¹ NaOH, which resulted in elution efficiencies of 62.6 ± 1.4% and 86.9 ± 2.6%,
208 respectively. The comparatively high elution efficiency obtained with 1 mol L⁻¹ H₂O₂ / 1 mol L⁻¹
209 NaOH may be due to the complete oxidation of the thiol functional groups on the mercapto-silica under
210 these conditions. These elution procedures are much simpler than that reported by Fan *et al.*¹⁸ for the
211 same binding gel, which required microwave-assisted extraction in concentrated HNO₃, HCl, and HF.
212 Subsequent elution of Sb^{III} from DGT binding layers in this study was done with 1 mol L⁻¹ H₂O₂ / 1
213 mol L⁻¹ NaOH, except where otherwise stated. Depending on the analytical equipment being used for
214 the quantification of antimony in the eluent solutions, it may be more appropriate to use the 0.1 mol L⁻¹
215 KIO₃ / 1 mol L⁻¹ HNO₃ / 1 mol L⁻¹ HCl eluent solution to minimise the dilution required before
216 analysis. The Agilent 7900 ICP-MS used in this study was tuned to have a robust plasma (i.e. CeO:Ce
217 < 0.2%) that was capable of analysing 1 mol L⁻¹ H₂O₂ / 1 mol L⁻¹ NaOH eluent solutions after only a 10
218 or 20-fold dilution, with negligible ionisation suppression due to high sodium ion concentrations.

219

220 **Accumulation over time.** The accumulation of Sb^{III} by mercapto-silica DGT and Metsorb DGT was
221 tested by deploying samplers of each type in triplicate for 10, 18, and 26 hours. Unfortunately, the
222 instability of Sb^{III} in laboratory solutions prevented longer deployment times. Linear regression of the
223 mass of Sb^{III} accumulated by the samplers over time confirmed that the uptake process was linear, with
224 R² values of 0.999 and 0.991 for mercapto-silica DGT and Metsorb DGT, respectively (Figure S1).
225 Effective diffusion coefficients were calculated as described previously¹⁹ from the slope of the
226 regression line, the ICP-MS-measured concentration of antimony in the deployment solution, the
227 thickness of the diffusive layer, and the area of the sampler exposed to the solution. The effective
228 diffusion coefficients of Sb^{III} estimated from the mercapto-silica DGT and Metsorb DGT data were
229 $9.42 \times 10^{-6} \text{ cm}^2 \text{ s}^{-1}$ and $8.23 \times 10^{-6} \text{ cm}^2 \text{ s}^{-1}$ at 25°C, respectively. These values are similar to the
230 diffusion coefficient previously reported for As^{III} ($9.04 \times 10^{-6} \text{ cm}^2 \text{ s}^{-1}$), which is a similar neutral
231 oxyanion.¹⁹ The diffusion coefficient of Sb^{III} reported by Fan *et al.*¹⁸ is only 32% of the value
232 determined in this study. This discrepancy is unlikely to be due to the difference in binding layer
233 materials, as suggested by Fan *et al.*,¹⁸ because the polyethersulfone membrane used in their study has
234 been used as a component of DGT diffusive layers previously without retarding diffusion to such a
235 large extent.²⁴ To ensure accurate results, we recommend that laboratories independently determine
236 diffusion coefficients with their specific DGT configuration.

237

238 The accumulation of Sb^V by mercapto-silica DGT was also tested over 26 hours, and was found to be
239 negligible (mass of Sb^V accumulated was ~2% of the mass of Sb^{III} accumulated over the same time)
240 (Figure S1). This provides initial verification that mercapto-silica DGT can be used to selectively
241 measure Sb^{III} in the presence of Sb^V, thus providing speciation information. Furthermore, previous
242 research by Panther *et al.*¹³ has demonstrated that Metsorb DGT quantitatively measures Sb^V, which in
243 association with this work, which demonstrates its capability to measure Sb^{III}, indicates that Metsorb
244 DGT can be used to measure total inorganic antimony (Sb^{III} + Sb^V). The simultaneous application of

245 both mercapto-silica DGT, for selectively measuring Sb^{III} ; and Metsorb DGT, for measuring both Sb^{III}
 246 and Sb^{V} , will allow the determination of inorganic antimony speciation in natural waters. Such an
 247 approach to *in situ* speciation using DGT has been described in detail previously for inorganic
 248 arsenic.^{19, 25}

249

250 **Effect of pH, ionic strength, bicarbonate concentration, and artificial seawater.** The performance
 251 of mercapto-silica DGT and Metsorb DGT under various solution conditions is summarised in Table 2.
 252 Both mercapto-silica DGT and Metsorb DGT show acceptable performance under all tested conditions,
 253 indicating that these techniques can be deployed in a variety of natural water types with confidence.
 254 Importantly, this is the first time that a thiol-based DGT technique has been tested for its ability to
 255 measure Sb^{III} in a seawater matrix and in the high concentrations of bicarbonate often found in
 256 sediment porewaters due to the microbial metabolism of organic matter. Previous work by Panther *et*
 257 *al.*²⁶ demonstrated the importance of considering bicarbonate as an interference, particularly for the
 258 measurement of oxyanionic species by DGT techniques with metal oxide-based binding gels.

259

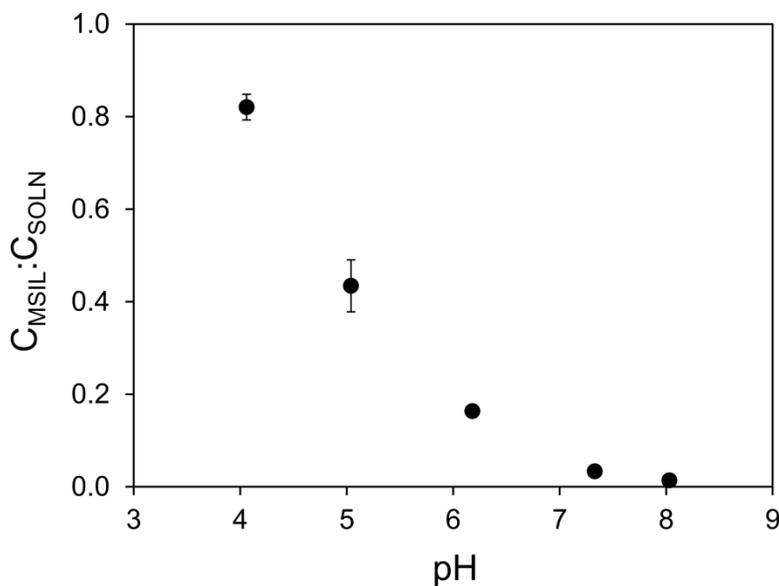
260 **Table 2. Effect of pH, ionic strength, bicarbonate concentration, and artificial seawater on the**
 261 **measurement of Sb^{III} by mercapto-silica DGT (C_{MSIL}) and Metsorb DGT (C_{MET}). The**
 262 **concentration of antimony in solution (C_{SOLN}) was measured by ICP-MS.**

<i>Parameter</i>		$C_{\text{MSIL}}:C_{\text{SOLN}}$	$C_{\text{MET}}:C_{\text{SOLN}}$
pH	4.07	0.99 ± 0.10	1.17 ± 0.08
	6.15	1.06 ± 0.04	1.09 ± 0.05
	8.05	0.98 ± 0.04	1.06 ± 0.04
Art. seawater (pH 8.35)		0.93 ± 0.13	0.99 ± 0.03
Ionic	0.001 mol L ⁻¹	1.05 ± 0.03	1.14 ± 0.05

strength (as NaCl)	0.01 mol L ⁻¹	1.00 ± 0.07	1.10 ± 0.05
	0.1 mol L ⁻¹	0.98 ± 0.01	1.14 ± 0.03
	1.0 mol L ⁻¹	0.95 ± 0.01	1.05 ± 0.09
[HCO ₃ ⁻]	1 mmol L ⁻¹	1.08 ± 0.06	1.09 ± 0.08
	5 mmol L ⁻¹	1.04 ± 0.08	1.14 ± 0.11
	10 mmol L ⁻¹	1.15 ± 0.02	1.04 ± 0.10
	15 mmol L ⁻¹	1.12 ± 0.05	1.03 ± 0.05

263

264 As the purpose of the mercapto-silica DGT technique is to selectively measure Sb^{III}, unlike Metsorb
 265 DGT that is designed to measure total inorganic antimony (Sb^{III} + Sb^V), the effect of pH on the
 266 accumulation of Sb^V by mercapto-silica DGT was tested to ensure that this technique remained
 267 selective for Sb^{III} over a range of environmentally relevant pH (Figure 1).



268

269 **Figure 1. Effect of solution pH on the accumulation of Sb^V by mercapto-silica DGT as shown by**
 270 **the ratio of the concentration measured by mercapto-silica DGT (C_{MSIL}) to the concentration**
 271 **measured in solution by ICP-MS (C_{SOLN}). Error bars represent ± 1 standard deviation from the**
 272 **mean (n=3).**

273

274 The solution pH had a strong effect on the accumulation of Sb^V by mercapto-silica DGT. At pH 4.06
275 the accumulation of Sb^V is close to quantitative, as indicated by a C_{MSIL}:C_{SOLN} value of 0.82. The ratio
276 decreased at pH 5.04 to 0.43, and at pH 6.18 it was only 0.16. Above pH 7, the accumulation of Sb^V by
277 mercapto-silica DGT was negligible (C_{MSIL}:C_{SOLN} < 0.03). The most likely explanation for the
278 accumulation of Sb^V by mercapto-silica at lower pH is the reduction of Sb^V to Sb^{III} by the thiol
279 functional groups, a phenomena observed previously in antimonial drug compounds,²⁷ followed by
280 adsorption of Sb^{III} to the mercapto-silica. At pH 4.06, the rate of the reduction reaction appears to be
281 sufficiently rapid to facilitate adsorption of Sb to the mercapto-silica without the accumulation of
282 antimony at the interface of the binding and diffusive layers. As the pH increases, the rate of the
283 reduction reaction may not be sufficiently rapid to allow adsorption of the antimony as it diffuses into
284 the sampler, causing the concentration at the interface of the binding and diffusive layers to increase,
285 retarding further diffusion of Sb^{III} from the bulk solution, causing the accumulation to depart from the
286 linear theoretical response.¹⁰ We recommend that Sb^{III} concentrations measured with the mercapto-
287 silica DGT technique be interpreted with caution when deployed in natural waters below pH 7, as there
288 may be a confounding effect of Sb^V adsorption at lower pH. Future work should examine if this effect
289 is observed in natural waters at low pH to further resolve the boundaries of acceptable deployment
290 conditions for this technique.

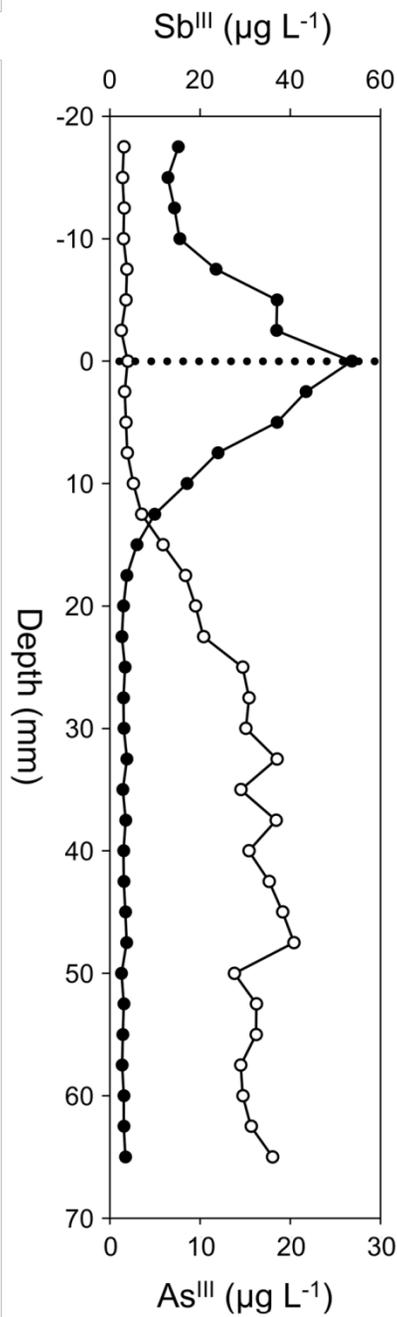
291

292 **Capacity.** The capacity of mercapto-silica DGT and Metsorb DGT for Sb^{III} was tested to ensure that
293 the binding layers have sufficient reactive sites for extended deployments, or deployment at high
294 antimony concentrations (e.g. at contaminated sites). The results of these experiments (Figure S2) show
295 that these DGT techniques exhibit linear accumulation capacity for Sb^{III} in excess of 100 µg of Sb^{III} per
296 binding layer disc. This capacity is sufficient, for example, to allow deployment for over 30 days at an
297 Sb^{III} concentration of 100 µg L⁻¹ (25°C), although biofilm development on the sampler surface
298 typically restricts deployments to shorter durations. The capacity is similar to that reported for As^{III}

299 with mercapto-silica DGT (77.5 μg), but much higher than that reported for Metsorb DGT (8.5 μg).¹⁹
300 The high capacity for antimony indicates that these techniques are suitable for measurements at both
301 contaminated and uncontaminated sites, and for long deployment times. However, we recommend that
302 DGT techniques are evaluated at field-relevant concentrations and deployment times before their
303 application to new deployment conditions. Users of this technique should also account for the
304 possibility of exceeding the analyte capacity due to the adsorption of non-target solutes that are
305 accumulated by the binding layer (e.g. arsenite), particularly at contaminated sites.

306

307 **High-resolution, porewater measurements of Sb^{III} and As^{III} with mercapto-silica DGT.** To
308 demonstrate the capability of the newly developed mercapto-silica DGT technique, it was applied to
309 measuring co-distributions of Sb^{III} and As^{III} in a contaminated freshwater sediment (Figure 2). The
310 calculated DGT detection limits of Sb^{III} and As^{III} were 0.06 $\mu\text{g L}^{-1}$ and 0.04 $\mu\text{g L}^{-1}$, respectively, and all
311 measured concentrations were above these values. It should be noted that the pH of the overlying water
312 in this mesocosm was ~ 5.2 during the DGT deployment, which could result in the measurement of
313 some Sb^V if it were present in the system, as explained earlier (see Figure 1).



314

315 **Figure 2. Porewater profile of Sb^{III} (●) and As^{III} (○) measured by mercapto-silica DGT in a**
 316 **contaminated freshwater wetland sediment at 2.5 mm resolution. The dotted line indicates the**
 317 **position of the sediment-water interface.**

318

319 Sb^{III} exhibits a sharp concentration maximum at the sediment-water interface, followed by a steep
 320 concentration decrease over a depth of 20 mm. Conversely, As^{III} is present at very low concentrations

321 to 10 mm depth, after which point the concentration gradually increases and plateaus at ~30 mm depth.
322 This is the first time that such contrasting porewater concentration profiles of Sb^{III} and As^{III} have been
323 observed at the millimetre scale in aquatic sediments. The steep Sb^{III} concentration gradient across the
324 sediment-water interface is clearly represented by numerous data points, due to the high spatial
325 resolution possible with the DGT technique – such a concentration profile would not be obtainable with
326 conventional sediment core slicing and porewater extraction techniques. Furthermore, the process of
327 porewater extraction, transport and storage could result in antimony and arsenic speciation changes
328 before analysis (e.g. oxidation), thus resulting in an erroneous interpretation of the sediment chemistry
329 of these contaminants. In contrast, the mercapto-silica DGT technique selectively accumulates the
330 reduced oxidation states of antimony and arsenic *in situ*, and thus preserves the speciation information
331 at the time of sampling. The accurate measurement of different oxidation states is integral to
332 investigating the complex biogeochemical processes associated with antimony and arsenic mobilisation
333 and sequestration. Future work should systematically apply the mercapto-silica DGT technique for the
334 selective measurement of Sb^{III} and As^{III}, in association with the Metsorb DGT technique for measuring
335 total Sb and As, in order to further investigate the geochemical behaviour of antimony and arsenic in
336 aquatic sediments.

337

338 **Conclusion.** The mercapto-silica DGT method presented here provides a significant advance in the
339 speciation analysis of antimony in circumneutral natural waters. Combined with the existing Metsorb
340 DGT method, it is possible to individually determine both Sb^{III} and Sb^V under a wide range of
341 environmental conditions, including fresh and marine surface waters and sediment porewaters.
342 Furthermore, this *in situ* approach avoids many of the common issues associated with conventional *ex*
343 *situ* speciation analysis, such as speciation changes during sample collection, transport and storage.

344

345 The application of these DGT techniques to investigating the sediment biogeochemistry of antimony
346 will allow researchers to selectively measure antimony species in sediment porewaters at higher spatial
347 resolution than previously possible. This is particularly important in productive freshwater and coastal
348 sediments, where changes in biogeochemical zonation, and possibly antimony speciation, can occur
349 over small spatial scales. The insight gained from this approach will allow a greater understanding of
350 the complex biogeochemical processes that govern the mobility of antimony in aquatic systems.

351

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357 **References**

- 358 1. D. E. Guberman, *2013 Minerals Yearbook*, United States Geological Survey, 2015.
- 359 2. USEPA, Priority Pollutant List, <http://www.epa.gov/sites/production/files/2015-09/documents/priority-pollutant-list-epa.pdf>, (accessed 14/12, 2015).
- 360 3. M. Filella, N. Belzile and Y.-W. Chen, *Earth-Sci. Rev.*, 2002, **57**, 125-176.
- 361 4. S. C. Wilson, P. V. Lockwood, P. M. Ashley and M. Tighe, *Environ. Pollut.*, 2010, **158**, 1169-1181.
- 362 5. M. Filella, P. A. Williams and N. Belzile, *Environ. Chem.*, 2009, **6**, 95-105.
- 363 6. C. Yu, Q. Cai, Z.-X. Guo, Z. Yang and S. B. Khoo, *Analyst*, 2002, **127**, 1380-1385.
- 364 7. B. Vrana, G. Mills, I. Allan, E. Dominiak, K. Svensson, J. Knutsson, G. Morrison and R. Greenwood, *Trends Anal. Chem.*, 2005, **24**, 845-868.
- 365 8. H. Zhang and W. Davison, *Environ. Chem.*, 2015, **12**, 85-101.
- 366 9. W. Davison and H. Zhang, *Nature*, 1994, **367**, 546-548.
- 367 10. H. Zhang and W. Davison, *Anal. Chem.*, 1995, **67**, 3391-3400.
- 368 11. O. Garmo, O. Royset, E. Steinnes and T. Flaten, *Anal. Chem.*, 2003, **75**, 3573-3580.
- 369 12. Q. Sun, J. Chen, H. Zhang, S. Ding, Z. Li, P. N. Williams, H. Cheng, C. Han, L. Wu and C. Zhang, *Anal. Chem.*, 2014, **86**, 3060-3067.
- 370 13. J. G. Panther, R. R. Stewart, P. R. Teasdale, W. W. Bennett, D. T. Welsh and H. Zhao, *Talanta*, 2013, **105**, 80-86.
- 371 14. P. Teasdale, S. Hayward and W. Davison, *Anal. Chem.*, 1999, **71**, 2186-2191.
- 372 15. J. G. Panther, P. R. Teasdale, W. W. Bennett, D. T. Welsh and H. Zhao, *Environ. Sci. Technol.*, 2010, **44**, 9419-9424.
- 373 16. S. Ding, D. Xu, Q. Sun, H. Yin and C. Zhang, *Environ. Sci. Technol.*, 2010, **44**, 8169-8174.

- 380 17. J. Huang, W. W. Bennett, D. T. Welsh, T. Li and P. R. Teasdale, *Anal. Chim. Acta*, 2016, **904**,
381 83-91.
- 382 18. H.-T. Fan, A.-J. Liu, B. Jiang, Q.-J. Wang, T. Li and C.-C. Huang, *RSC Advances*, 2016, **6**,
383 2624-2631.
- 384 19. W. W. Bennett, P. R. Teasdale, J. G. Panther, D. T. Welsh and D. F. Jolley, *Anal. Chem.*, 2011,
385 **83**, 8293-8299.
- 386 20. J. G. Panther, K. P. Stillwell, K. J. Powell and A. J. Downard, *Anal. Chim. Acta*, 2008, **622**,
387 133-142.
- 388 21. W. W. Bennett, P. R. Teasdale, J. G. Panther, D. T. Welsh and D. F. Jolley, *Anal. Chem.*, 2010,
389 **82**, 7401-7407.
- 390 22. W. W. Bennett, P. R. Teasdale, J. G. Panther, D. T. Welsh, H. Zhao and D. F. Jolley, *Environ.*
391 *Sci. Technol.*, 2012, **46**, 3981-3989.
- 392 23. K. Grasshoff, M. Ehrhardt, K. Kremling and L. G. Anderson, *Methods of seawater analysis*,
393 Wiley-VCH, 3rd edn., 1999.
- 394 24. O. A. Garmo, W. Davison and H. Zhang, *Anal. Chem.*, 2008, **80**, 9220-9225.
- 395 25. J. Gorny, L. Lesven, G. Billon, D. Dumoulin, C. Noiriel, C. Pirovano and B. Madé, *Talanta*,
396 2015, **144**, 890-898.
- 397 26. J. G. Panther, P. R. Teasdale, W. W. Bennett, D. T. Welsh and H. Zhao, *Anal. Chim. Acta*,
398 2011, **698**, 20-26.
- 399 27. C. dos Santos Ferreira, P. Silveira Martins, C. Demicheli, C. Brochu, M. Ouellette and F.
400 Frézard, *BioMetals*, **16**, 441-446.
- 401