

Selective separation of arsenic species from aqueous solutions with immobilized macrocyclic material containing solid phase extraction columns

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1 **Selective Separation of Arsenic Species from Aqueous Solutions with**
2 **Immobilized Macrocyclic Material Containing Solid Phase Extraction**
3 **Columns**

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23 **Abstract**

24 A combination of solid phase extraction (SPE) columns was used for selective separation
25 of water-soluble arsenic species: arsenite, arsenate, monomethylarsonic acid (MMA) and
26 dimethylarsinic acid (DMA). The SPE columns, namely AnaLig TE-01 (TE-01), AnaLig
27 AN-01 Si (AN-01) and AnaLig As-01 PA (As-01), contain immobilized macrocyclic material
28 as the sorbent and commonly known as molecular recognition technology (MRT) gel. The
29 retention, extraction and recovery behavior of the MRT gel SPE columns were studied at pH
30 4–10. Fortified deionized water spiked with 100 μM of arsenic species were treated at the
31 flow rate of 0.2 mL min^{-1} . HNO_3 (1.0 and 6.0 M) was used as eluent to recover the retained
32 arsenic species from TE-01 and AN-01 SPE columns. Arsenic species retained in the As-01
33 column were eluted with HNO_3 (0.1 M) followed by NaOH (2.0 M). Likely interference from
34 the various coexisting ions (Na^+ , K^+ , Ca^{2+} , Mg^{2+} , Cl^- , NO_3^- , CH_3COO^- , PO_4^{3-} , SO_4^{2-} , ClO_4^-)
35 (10 mM) were negligible. Quantitative separation of As(III), As(V), MMA and DMA was
36 achieved based on the differences in extraction and recovery behavior of the MRT gel SPE
37 columns with pH for different arsenic species. Complexation between arsenic species and
38 MRT gel is the core phenomenon of the proposed technique as the complexation of MRT
39 gels is expected to be stronger than the resin-based separation processes. MRT gel SPE
40 columns are advantageous as compared with other reported SPE columns in terms of its
41 performance with As(III). Effortless regeneration and unaltered separation performance of
42 the sorbent materials for more than 100 loading and elution cycles are other sturdy
43 characteristics to consider the MRT gel SPE columns for sensitive and selective arsenic
44 species separation.

45

46 **Keywords:** Solid phase extraction; Molecular recognition technology gel; water-soluble
47 arsenic; selective separation; pH

48 **1.0 Introduction**

49 Arsenic, a ubiquitous toxic trace element, has raised a major toxicological and
50 environmental concerns (WHO, 2001). The concentration levels, oxidation and binding states,
51 ionic and molecular forms and metabolic pathways of As vary strongly in different
52 environmental compartments, food chains and ultimately in humans (Mandal and Suzuki,
53 2002). Widespread human exposure to high levels of As is reported to occur via drinking
54 water and contaminated water irrigated food causing both cancerous and non-cancerous
55 health effects (Karim, 2000; Rahman et al., 2008).

56 Arsenite (oxidation state + III), arsenate (oxidation state + V), monomethylarsonic acid
57 (MMA) and dimethylarsinic acid (DMA) are common water-soluble arsenic species existing
58 in natural water systems-a major pathway of arsenic ingestion to humans (Smedley and
59 Kinniburgh, 2002). Arsenic toxicity in human depends strongly on its chemical form. As(III)
60 is 10 times more toxic than As(V) while almost 70 times more toxic than the methylated
61 forms, MMA and DMA (Squibb and Fowler, 1983). As(III), having successive acid
62 dissociation constants (pK_a) of 9.2, 12.2 and 13.4, is not dissociated at neutral pH and is
63 present as a neutral species. As(V) and MMA has a wide range of pK_a values [As(V): 2.2, 6.9,
64 11.5; MMA: 4.1, 8.7], and exist mainly as anionic species at almost all pH. DMA with a pK_a
65 value of 6.2 subsists as a cation in acidic medium (Committee on Medical and Biologic
66 Effects of Environmental Pollutants, 1977). The United States Environmental Protection
67 Agency proposed a maximum contaminant level of $10 \mu\text{g L}^{-1}$ arsenic for the community
68 water systems (USEPA, 2002). Because of increasingly stringent environmental regulations,
69 selective and accurate measurement of arsenic species is required for precise monitoring and
70 understanding the extent of arsenic contamination.

71 In natural waters, As usually exists at trace levels and several techniques are proposed for
72 selective quantification and speciation analysis of arsenic species at trace levels (Barra et al.,

73 2000; Munoz and Palmero, 2005; Terlecka, 2005; Kumar and Riyazuddin, 2007; Mays and
74 Hussam, 2009). Ion chromatography and high performance liquid chromatography separation
75 followed by sensitive detection such as inductively coupled plasma mass spectrometry
76 (Lintschinger et al., 1998; Bissen and Frimmel, 2000), atomic absorption spectrometry (AAS)
77 with hydride generation interface (Hasegawa et al., 1999; Kumar and Riyazuddin, 2007) and
78 electrospray/nanospray mass spectrometry (Pergantis et al., 1997; Ritsema et al., 1998) are
79 some potential techniques. However, concerns related to the use of element-selective
80 detectors to interface the chromatographic methods limit the efficiency of these techniques
81 (Yu et al., 2003).

82 Separation and preconcentration of contaminant ions using solid sorbent materials, known
83 as solid phase extraction (SPE) systems, have increased in popularity since the 1980s (Hosten
84 and Welz, 1999). The technique has been developed as a cost- and time-saving alternative to
85 the traditional extraction techniques featuring the capability to interact with a variety of metal
86 ions including the fairly specific selectivity to a particular ion (Nickson et al., 1995; Ghaedi
87 et al., 2008). Ion exchange resins (Leal et al., 2004; Jitmanee et al., 2005), silica gel bonded
88 with octadecyl functional groups (Pozebon et al., 1998), yeast immobilized on controlled
89 pore glass (Koh et al., 2005), activated alumina (Karthikeyan et al., 1999), open tubes knotted
90 reactors (Yan et al., 2002; Herbello-Hermelo et al., 2005), polytetrafluoroethylene turnings-
91 packed micro-columns (Anthemidis et al., 2010) have been employed as SPE sorbent
92 material. One group of SPE materials includes the macrocyclic chelants, such as crown ethers,
93 immobilized on a silica or polymer support (Hosten and Welz, 1999). Ion-selective behavior
94 of SPE-type systems with immobilized macrocyclic materials has been mentioned for
95 preconcentration and separation of metals (Bradshaw et al., 1988; Izatt et al., 1994;
96 Hasegawa et al., 2010). SPE techniques have been applied for the quantitative
97 analysis/speciation/separation of various trace elements including arsenic (Yalcin and Le,

98 2001; Yu et al., 2003; Liang et al., 2004; Long et al., 2006; Sanchez et al., 2009). Reports on
99 the retention behavior of different arsenic species with some SPE systems (silica-based or
100 resin-based) at pH 5.5 (Yalcin and Le, 2001) and pH 5.6 (Yu et al., 2003) were available. It
101 was observed that the hydrophobic interaction of the arsenic species with the SPE materials,
102 pK_a values and ionic characters are important factors which may control the retention
103 efficiency of the SPE columns (Yu et al., 2003). Though quantitative retention was achieved
104 with the SPE columns for the water-soluble arsenic species (As(III), As(V), MMA and
105 DMA), elution of the retained species was quiet difficult or sometimes unachievable for some
106 species particularly with As(III) (Yalcin and Le, 2001; Yu et al., 2003).

107 The objective of the work is to investigate the feasibility of an ion-selective immobilized
108 macrocyclic material attached to a solid phase, commonly known as a molecular recognition
109 technology (MRT) gel, for the selective separation of As(III), As(V), MMA and DMA from
110 aqueous solutions followed by graphite furnace AAS determination. We used following MRT
111 gel SPE columns: AnaLig TE-01, AnaLig AN-01 Si and AnaLig As-01 PA. Specific MRT
112 gel SPE columns have the advantage of the selective retention of the mentioned arsenic
113 species followed by quantitative recovery. Most importantly, As(III) was quantitatively
114 retained and recovered with the AnaLig As-01 PA SPE column.

115 **2.0 Experimental**

116 **2.1 Instruments**

117 A PerkinElmer model AAnalyst 600 AAS (PerkinElmer, Massachusetts, USA) including
118 the AS-800 autosampler equipped with a transverse-heated graphite atomizer with integrated,
119 pyrolytic graphite coated platform (THGA) and longitudinal Zeeman-effect background
120 corrector was used. End-capped THGA tubes were used for better sensitivity and improved
121 precision. An electrodeless discharge lamp (EDL) powered by EDL System II operated at

122 380 mA was employed as light source. The wavelength was set at the 193.7 nm resonance
123 line and the monochromator spectral bandpass at 0.7 nm. Baseline offset correction time was
124 set to 2.0 s and the read delay at 0.0 s. Argon was used as purge gas and the flow rate was set
125 to 250 mL min⁻¹. A temperature program was performed and the different steps were: first
126 and second dry at 110 and 130 °C, ashing at 1200 °C and atomization at 2000 °C held at 30,
127 30, 20 and 5 s respectively. After a calibration with 5 standards (0.5–2.5 μM), 20 μL of
128 sample and 10 μL of Pd–Mg matrix modifier were introduced in the graphite furnace with
129 three replicates of each measurement. The pH of the sample solutions was measured with a
130 Navi F-52 pH meter (Horiba Instruments, Japan) and a combination electrode.

131 ***2.2 Reagents and materials***

132 Analytical grade commercial products were used. Stock solutions (10 mM) of As(III),
133 As(V), MMA and DMA were prepared from sodium arsenite (NaAsO₂) (Kanto Chemical,
134 Japan), sodium arsenate heptahydrate (Na₂HAsO₄·7H₂O), monomethylarsonic acid
135 (CH₃AsO(OH)₂), dimethylarsinic acid sodium salt trihydrate (C₂H₆AsNaO₂·3H₂O) (Nacali
136 Tesque, Japan). Working standards of metal solutions in the range of μM to mM were
137 prepared by dilution on a weight basis. Deionized water prepared with a Barnstead 4 housing
138 E-Pure systems was used to prepare all solutions and is referred to as EPW hereafter.

139 The experimental pH range was 4–10, and adjusted using either 1 M HCl or 1 M NaOH.
140 MES (2-(*N*-morpholino) ethanesulfonic acid monohydrate, C₆H₁₃NO₄S·H₂O) (Sigma–Aldrich,
141 USA), HEPES (N-2-Hydroxyethylpiperazine-N'-2-ethanesulfonic acid, C₈H₁₈N₂O₄S) (Nacali
142 Tesque, Japan), and TAPS (N-Tris(hydroxymethyl)methyl-3-aminopropanesulfonic acid,
143 C₇H₁₇NO₆S) (MP Biomedicals, USA) were used as buffer reagents for pH 4–6, 7–8 and 9–10,
144 respectively.

145 NaCl, KCl, CaCl₂, MgCl₂ were used as a source of cations while the Na-salt of anions
146 (Cl⁻, NO₃⁻, CH₃COO⁻, PO₄³⁻, SO₄²⁻, ClO₄⁻) (Nacali Tesque, Japan) were used to study the

147 effect of coexisting ions. Working solutions of 10 mM concentration were prepared in H₂O
148 matrix and pH was maintained to 7.0. The final solutions were allowed to equilibrate for 24 h
149 before use. The interference study were carried out in a non-competitive environment by
150 applying 4 mL of fortified deionized water at the optimized flow rate with subsequent
151 collection using appropriate eluent.

152 Experimental variables, *e.g.* sample loading flow rate, selection of eluent and eluent
153 concentration were optimized using As(V) spiked solutions (100 μM) in H₂O matrix with pH
154 maintained at 5.0. The MRT gel SPE columns were fed with 4 mL of sample solutions at
155 varying flow rates, and the retention percentage of the As-species into the columns was
156 determined. Different eluent (individual or combinations), 0.1–6.0 M HNO₃ and 0.1–4.0 M
157 NaOH, was checked to select the most appropriate eluent or eluent combinations that were
158 suitable for optimum recovery of the ‘captured’ species.

159 Certified reference materials (CRMs): BCR-713 (effluent wastewater) and BCR-610
160 (groundwater) from EC-JRC-IRMM (European Commission Joint Research Centre, Institute
161 of Reference Materials and Measurements), fortified samples of ‘real’ waters: tap water
162 sample from our laboratory in Kakuma campus, Kanazawa University (Kanazawa, Japan)
163 and river water sample from Asano River (Kanazawa, Japan) were analyzed to validate the
164 proposed separation process. Each of the real water samples was filtered through the cellulose
165 membrane filter of 0.45 μm pore size (Advantec, Japan) before the analysis.

166 Low-density polyethylene bottles (Nalge, USA), perfluoroalkoxy (PFA) tubes and
167 micropipette tips (Nichiryo, Japan) were used throughout. The laboratory wares were cleaned
168 following the sequence: (a) soaking in an alkaline detergent (Scat 20X-PF, Nacali Tesque,
169 Japan) overnight, (b) rinsed with EPW, (c) soaking in 4 M HCl overnight, and (d) rinsed with
170 EPW. PFA tubes and micropipette tips were cleaned according to the procedure described by
171 Sohrin et al. (1998).

172 **2.3 Separation procedure**

173 *2.3.1 Column cleaning and conditioning*

174 MRT gel SPE columns: AnaLig TE-01 (TE-01), AnaLig AN-01 Si (AN-01), AnaLig As-
175 01 PA (As-01) were purchased from GL Sciences, Japan. The SPE sorbents are proprietary
176 polymeric organic materials comprised of ion-selective sequestering property. The sorption
177 ability of the SPE materials is based on the molecular recognition and macrocyclic chemistry.
178 SPE materials packed in 3 mL columns were used in the experiments. Column cleaning was
179 conducted with 8 mL of 1.0 M HNO₃ and 6 mL of EPW. Appropriate buffer solution (5 mL)
180 was allowed to follow through the column to ensure the desired pH condition (4–10).

181 *2.3.2 Retention, extraction and recovery of arsenic species with MRT gel columns*

182 The work-flow sequence for the separation of As(III), As(V), MMA and DMA using
183 MRT gel SPE columns followed by GF-AAS determination is summarized in Table 1.
184 Sample solution (4 mL) was passed through the SPE column at the optimized pre-set flow
185 rate of 0.2 mL min⁻¹. The pH of the sample solution was pre-adjusted with 0.1 M buffer
186 solution (MES, HEPES or TAPS, whichever appropriate). The column effluent was collected.
187 The MRT gel SPE columns were then washed with EPW to remove the analyte that is not
188 captured by the immobilized macrocyclic material in SPE columns. The total analyte
189 concentration in the column effluent and EPW wash solution represent the unretained
190 concentration of analyte in the SPE system. The final step is the elution of analyte from the
191 SPE systems. HNO₃ (1.0 and 6.0 M) was used to elute the arsenic species retained in TE-01
192 and AN-01 SPE columns, and analytes retained in As-01 column were eluted with 0.1 M
193 HNO₃ followed by 2.0 M NaOH. The arsenic concentrations in the sample, effluents and
194 eluent solutions were measured with GF-AAS. Three replicate measurements per sample
195 were made in all instances. The peak height of the reported signal was proportional to the
196 concentration of the respective arsenic species and was used for all measurements.

197 The terms, retention, extraction and recovery, were used to explain the separation
198 performance of the SPE systems. The retention ratio was calculated comparing the analyte
199 concentration in the sample solution loaded in SPE columns with that in the solution passed
200 through the columns providing only the information about the concentration of analyte sorbed
201 in the SPE systems. On the other hand, the analyte concentrations in the column effluent,
202 EPW wash solution and eluent were measured to understand the extraction and recovery
203 behavior of the SPE columns. The extraction ratio of each column for the individual species
204 was calculated by comparing the numbers of mol of analyte in the eluent with the cumulative
205 number of mol of analyte present in the total effluents. Numbers of mol of analyte recovered
206 in all fractions were compared with the numbers of mol of analyte in the solution loaded to
207 the column to calculate the recovery ratio.

208 **3.0 Results and discussion**

209 *3.1 Optimization of variables*

210 *3.1.1 Sample loading flow rate*

211 The flow rate of the metal-rich sample solution has a reasonable impact on the metal
212 retention rate in SPE columns (Bag et al., 1998). Effect of sample loading flow rates adjusted
213 in the range of 0.2–4.0 mL min⁻¹ (Table 2) was checked at the optimum conditions.
214 Quantitative retention up to the flow rates of 0.25 mL min⁻¹ was observed. The retention
215 capacities decrease gradually with the increase of flow rates above 0.25 mL min⁻¹. Such
216 behavior indicates the constant retaining capability of the MRT gel at the initial loading
217 period. Therefore, a flow rate of 0.2 mL min⁻¹ was applied to ensure maximum retention of
218 the analyte from MRT Gel SPE columns.

219 3.1.2 Selection of eluent and eluent concentration

220 The eluent should be able to extract the analyte without affecting the quantitative
221 determination of analytes (Chen et al., 2009). Analytes retained in the TE-01 and AN-01 SPE
222 columns were eluted with HNO₃ (4 mL) of varying concentrations (0.1–6.0 M). The recovery
223 patterns were similar and the recovery rates became constant for the eluent concentration
224 above 0.5 M (Figs. 1a and 1b). However, greater than or equal to 5.0 M acids were
225 recommended for the elution of bound ions in TE-01 and AN-01 SPE columns (IBC
226 Advanced Technologies, 2007, 2009). Hence, a combination of 1.0 M HNO₃ (2 mL) and 6.0
227 M HNO₃ (1 mL) was selected as eluent for the subsequent experiments to ensure the
228 complete elution of the analyte when treated with TE-01 or AN-01. On the other hand, only
229 0.1–4.0 M NaOH (2 mL) or 0.1–6.0 M HNO₃ (2 mL) was found unsuitable for the elution of
230 analytes from As-01. Combinations of 0.1–4.0 M NaOH (1 mL) followed by 2.0 M HNO₃ (1
231 mL) and vice-versa were used to check the elution of arsenic species from the As-01 column
232 (Figs. 1c and 1d). The recovery was achieved at quantitative maximum for the following
233 eluent combination: 0.1 M HNO₃ (1 mL) + 2.0 M NaOH (1 mL), and was applied for the next
234 experiments with As-01 MRT gel column.

235 3.2 Retention behavior of the MRT gel SPE columns

236 The retention efficiency of the MRT gel SPE columns for different arsenic species at
237 varying pH is illustrated in Fig. 2. The retention (%) of As(III) was negligible with TE-01 and
238 AN-01 SPE columns. Average retention efficiency (%) of 92 ± 3.7 was observed with As-01
239 column at the pH 4 to 10 while it was highest at pH 7 (96 ± 1.2). As(III) mainly exists as a
240 neutral species, As(OH)₃, at the entire range of the studied pH. Thus, the macrocyclic
241 materials immobilized in the TE-01 and AN-01 columns were not capable of retaining the
242 neutral form of As(III). Almost complete retention of As(V) and MMA was achieved at pH 4
243 to 7 with all the MRT gel SPE columns. As(V) and MMA remain in the anionic form within

244 that pH range, as evident from the corresponding pK_a values. Therefore, all of the MRT gel
245 columns investigated can retain the anionic form of As(V) and MMA. DMA, which exists as
246 a cation in the acidic medium, was retained at an average efficiency (%) of 94 ± 3.3 with As-
247 01 column between pH 4 and 6 while the retention was not that notable with TE-01 and AN-
248 01 columns.

249 Data evaluation showed that the most significant finding of our work was with As(III).
250 Yu et al. (2003) checked 11 SPE systems at pH 5.6 and found that none of them were capable
251 of retaining As(III) quantitatively. Yalcin and Le (2001) worked with 7 SPE systems and
252 reported that Alumina-A, -B and -N (normal phase in acidic, basic, and neutral activity; from
253 Millipore-Waters, Missisauga, ON, Canada) and silica-based LC-SCX (sulfonic acid-bonded;
254 from Supelco, Bellefonte, PA, USA) columns can retain As(III) at the pH of 5.5. None of
255 those SPE systems were recommended for As(III) separation considering the difficulty in
256 elution. In our study, at pH 7, As(III) was completely retained at As-01 SPE column followed
257 by quantitative recovery.

258 ***3.3 Extraction and recovery behavior of the MRT gel SPE columns***

259 The extraction behavior of the MRT gel SPE columns with four arsenic species is
260 illustrated in Fig. 3. A similar extraction pattern was observed with TE-01 and AN-01 SPE
261 columns; As(III) was not captured, As(V) was captured at an average rate (%) of 99 ± 0.5 until
262 pH 8, MMA extracted at an average percent rate of 99 ± 0.60 at pH 6 and 7, and the highest
263 extraction (%) of 71 ± 4.6 was observed at pH 7 for DMA. With As-01 SPE columns, the
264 average extraction (%) was 96 ± 3.2 at pH 4–6 for As(V), MMA and DMA, while it was
265 99 ± 1.1 at pH 7–9 for As(III).

266 Recovery (%) of the arsenic species with the MRT gel SPE columns is shown in Fig. 4.
267 TE-01 SPE columns showed quantitative recovery performance at the entire studied pH range
268 for all the arsenic species. AN-01 SPE columns showed similar behavior with As(III), As(V)

269 and DMA while fluctuating recovery was achieved for MMA at different pH. A gradual
270 increase in the recovery (%) was observed from pH 4 to pH 10 with As-01 SPE columns, and
271 expected maximum recoveries were achieved for all the arsenic species at pH 7.

272 The extraction and recovery behavior of the MRT gel SPE columns leads us to the
273 following assumptions: (i) TE-01 and AN-01 columns are not effective for As(III) separation
274 but can be used to separate other target species (As(V), MMA and DMA) quantitatively at
275 varying pH conditions; (ii) selective separation and complete elution of As(III) is possible
276 with the As-01 column; (iii) the As-01 column can also be used to preconcentrate the targeted
277 water-soluble arsenic species for the determination of total arsenic content in the samples, if
278 selective separation is not desired; and (iv) column regeneration process is simple because the
279 retained analytes are completely eluted.

280 ***3.4 Interference studies***

281 Cations of alkaline and alkaline earth metals are always found in water samples and have
282 the capability to compete with the target metal ions during the binding with the SPE material,
283 and common anions have the ability to bind with the target metal ions. In their presence, the
284 efficiency of the SPE material to bind the target ions may be reduced resulting in a reduction
285 of the recovery. The effects of matrix ions in water samples on the recovery of the spiked
286 sample solutions of 100 μM As(III), As(V), MMA and DMA were investigated. The
287 recovery of different arsenic species in the presence of 10 mM of different ions (Na^+ , K^+ ,
288 Ca^{2+} , Mg^{2+} , Cl^- , NO_3^- , CH_3COO^- , PO_4^{3-} , SO_4^{2-} , ClO_4^-) in the water samples were observed
289 in the range of 95 ± 2.7 – $100\pm 3.2\%$. Therefore, there is limited possibility of the interference
290 from the matrix ions commonly found in sample waters, which is may be due to the selective
291 separation capability of the MRT gel SPE materials.

292 **3.5 Retention capacity and regeneration of the SPE columns**

293 Retention capacity of the MRT gel SPE columns is important for determining the stability
294 of the MRT gel SPE columns during the separation process. Analyte concentration and
295 breakthrough volume (the volume of sample that causes the target analyte to be eluted from
296 the SPE columns) were used to find out the retention capacity (Yu et al., 2003). After arsenic-
297 spiked sample solutions were passed through the SPE columns, the retention capacity was
298 expressed in terms of mmol of analyte captured in one gram of SPE material. The retention
299 capacities of the MRT gel SPE columns at pH 7 were calculated as follows: 0.40 ± 0.02 mmol
300 g^{-1} TE-01, 0.39 ± 0.02 mmol g^{-1} AN-01 and 0.31 ± 0.01 mmol g^{-1} As-01 (sample solution– 10
301 mM of As(V), matrix– H_2O , flow rate– 0.2 mL min^{-1} , elution– 2 mL of HNO_3 + 1 mL of 6
302 M HNO_3 + 1 mL of EPW, for TE-01 and AN-01 SPE columns and 1 mL of 0.1 M HNO_3 + 1
303 mL of 2.0 M NaOH + 2 mL of EPW). The result was in good agreement with the certified
304 values for the MRT gel SPE columns (IBC Advanced Technologies, 2006, 2007, 2009). The
305 regeneration ability of the MRT gel SPE columns was investigated, and it was observed that
306 more than 100 loading and elution cycles can be performed without the loss of analytical
307 performance. SPE systems with macrocycles attached onto solid supports allow selective
308 separation of analytes from matrix facilitating the repeated use of the macrocycles (Bradshaw
309 et al., 1988; Horwitz et al., 1992; Izatt, 1997).

310 **3.6 Scheme for selective separation of arsenic species**

311 The differences in extraction and recovery pattern of MRT gel SPE columns for different
312 arsenic species enabled us to propose a selective separation method. The method is based on
313 the selective retention of the arsenic species followed by quantitative selective recovery at the
314 elution step. Retention, extraction and recovery behavior of three MRT gel SPE columns:
315 TE-01, AN-01 and As-01 were studied and combined to design a multi-step separation
316 technique for quantitative measurement of As(III), As(V), MMA and DMA. Another MRT

317 gel SPE column, AnaLig AN-02, was also checked. The retention, extraction and recovery
318 behaviors of the AN-02 column were somewhat similar with those of AN-01 column.
319 Therefore, AN-02 column can be considered as an alternative of AN-01 column in the
320 separation process. The scheme for selective separation with subsequent quantitative
321 measurement of the arsenic species by GF-AAS technique is shown in Fig. 5.

322 At pH 5, As(V) and MMA were quantitatively retained in the TE-01 SPE column while
323 As(III) and DMA remained in the column effluent. The captured species was eluted with
324 HNO₃. The eluted solution was separated into two equal portions, and pH was adjusted to 5
325 and 8 respectively. When each of the pH-adjusted portions independently treated with AN-01
326 SPE columns, As(V) and MMA were quantitatively extracted and recovered from the eluted
327 solution, respectively, at pH 5 and pH 8. The column effluent containing As(III) and DMA
328 were adjusted to pH 9, and treated with As-01 SPE column. DMA remained in the solution
329 that passed through the SPE material while As(III) was selectively captured. Captured As(III)
330 was eluted with the eluent combination of 0.1 M HNO₃ followed by 2.0 M NaOH. GF-AAS
331 were used to determine the concentration of the individual arsenic species.

332 **3.7 Analytical characteristics**

333 The concentrations of As(III), As(V), MMA and DMA in the treated solutions from MRT
334 gel SPE columns were measured using GF-AAS. At optimum conditions, the linear range
335 was found to be 0.01–0.32 µg mL⁻¹ As(III), 0.01–0.78 µg mL⁻¹ As(V), 0.01–0.35 µg mL⁻¹
336 MMA and 0.01–0.54 µg mL⁻¹ DMA. The method detection limits were calculated by three
337 times the standard deviation ($n = 15$) of the blank. The values were 0.06 µg L⁻¹ for As(III)
338 and As(V), and 0.05 µg L⁻¹ for MMA and DMA. The precision of the method was also
339 studied. The repeatability, as relative standard deviation, was 0.65, 2.93, 2.25 and 1.20%,
340 calculated from 10 replicate measurements at the 1.0 µM of As(III), As(V), MMA and DMA
341 respectively.

342 **3.8 Accuracy and applications**

343 The accuracy of the proposed separation scheme was evaluated by analyzing two EC-
344 JRC-IRMM CRMs, namely BCR-713 (effluent wastewater) and BCR-610 (groundwater)
345 (Table 3). None of the arsenic species measured in this work has either certified or literature
346 values. Our values for the total of all arsenic species determined for both BCR-713 and BCR-
347 610 were in good agreement with the certified value, the calculated recoveries, 97% for BCR-
348 713 and 94% for BCR-610, were satisfactory. The proposed separation scheme was also
349 applied to the analysis of local natural water samples (tap water and river water) and was
350 validated by spiking the samples with known amounts of As(III), As(V), MMA and DMA
351 (Table 4). The recoveries from spiked solutions were varied in the range 98 ± 1.6 – $102\pm 1.7\%$.

352 **4.0 Conclusions**

353 The application of three MRT gel SPE columns (TE-01, AN-01 and As-01) for selective
354 separation of four different arsenic species (As(III), As(V), MMA and DMA) was
355 demonstrated. Retention behaviors of the arsenic species were varied with the change of pH
356 at the range of 4 to 10. TE-01 and AN-01 SPE columns were unable to retain As(III) while
357 As-01 showed the ability to retain all the species at a certain pH quantitatively. Either HNO₃
358 or a combination of HNO₃ and NaOH were used as eluent to recover the ‘captured’ species
359 from the MRT gel structure. However, the recovery ratio was also found to depend on the pH.
360 pH-dependent retention and recovery behavior of the MRT gel SPE columns were used to
361 design a selective separation scheme for quantitative determination of a particular arsenic
362 species in the sample solution. It is possible to overcome the tedious preconcentration process
363 by following the proposed selective separation technique. To the best of our knowledge, it is
364 the first ever report dealing with SPE columns equipped with immobilized macrocyclic
365 material as sorbent material for selective determination of arsenic in water. In addition,
366 quantitative retention followed by recovery of As(III) was achieved with As-01 column

367 which was previously not achieved with any other reported SPE systems. Easy operation,
368 virtually unlimited loading and elution capability of the sorbent material without losing the
369 analytical performance and high-sensitive separation ability can make the proposed technique
370 as a useful one for selective separation of arsenic species from natural waters.

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389 **References**

- 390 Anthemidis, A.N., Zachariadis, G.A., Stratis, J.A., 2010. On-line preconcentration and determination
391 of nickel and zinc in natural water samples by flow injection - flame atomic absorption
392 spectrometry using PTFE-turnings for column packing. *Int. J. Environ. An. Ch.* 90, 127-136.
- 393 Bag, H., Lale, M., Turker, A.R., 1998. Determination of iron and nickel by flame atomic absorption
394 spectrophotometry after preconcentration on *Saccharomyces cerevisiae* immobilized sepiolite.
395 *Talanta* 47, 689-696.
- 396 Barra, C.M., Santelli, R.E., Abrao, J.J., de la Guardia, M., 2000. Arsenic speciation - A review. *Quim.*
397 *Nova* 23, 58-70.
- 398 Bissen, M., Frimmel, F.H., 2000. Speciation of As(III), As(V), MMA and DMA in contaminated soil
399 extracts by HPLC-ICP/MS. *Fresen. J. Anal. Chem.* 367, 51-55.
- 400 Bradshaw, J.S., Bruening, R.L., Krakowiak, K.E., Tarbet, B.J., Bruening, M.L., Izatt, R.M.,
401 Christensen, J.J., 1988. Preparation of silica gel-bound macrocycles and their cation-binding
402 properties. *J. Chem. Soc. Chem. Comm.* 812-814.
- 403 Chen, D., Huang, C., He, M., Hu, B., 2009. Separation and preconcentration of inorganic arsenic
404 species in natural water samples with 3-(2-aminoethylamino) propyltrimethoxysilane
405 modified ordered mesoporous silica micro-column and their determination by inductively
406 coupled plasma optical emission spectrometry. *J. Hazard. Mater.* 164, 1146-1151.
- 407 Committee on Medical and Biologic Effects of Environmental Pollutants, 1977. Arsenic: Medical and
408 Biologic Effects of Environmental Pollutants. National Academy of Sciences, Washington,
409 D.C.
- 410 Ghaedi, M., Shokrollahi, A., Kianfar, A.H., Mirsadeghi, A.S., Pourfarokhi, A., Soylak, M., 2008. The
411 determination of some heavy metals in food samples by flame atomic absorption
412 spectrometry after their separation-preconcentration on bis salicyl aldehyde, 1,3 propan
413 diimine (BSPDI) loaded on activated carbon. *J. Hazard. Mater.* 154, 128-134.
- 414 Hasegawa, H., Matsui, M., Okamura, S., Hojo, M., Iwasaki, N., Sohrin, Y., 1999. Arsenic speciation
415 including 'hidden' arsenic in natural waters. *Appl. Organomet. Chem.* 13, 113-119.
- 416 Hasegawa, H., Rahman, I.M.M., Kinoshita, S., Maki, T., Furusho, Y., 2010. Non-destructive
417 separation of metal ions from wastewater containing excess aminopolycarboxylate chelant in
418 solution with an ion-selective immobilized macrocyclic material. *Chemosphere* 79, 193-198.
- 419 Herbello-Hermelo, P., Barciela-Alonso, M.C., Bermejo-Barrera, A., Bermejo-Barrera, P., 2005. Flow
420 on-line sorption preconcentration in a knotted reactor coupled with electrothermal atomic
421 absorption spectrometry for selective AS(III) determination in sea-water samples. *J. Anal.*
422 *Atom. Spectrom.* 20, 662-664.

423 Horwitz, E., Dietz, M., Chiarizia, R., 1992. The application of novel extraction chromatographic
424 materials to the characterization of radioactive waste solutions. *J. Radioanal. Nucl. Ch.* 161,
425 575-583.

426 Hosten, E., Welz, B., 1999. Evaluation of an immobilised macrocyclic material for on-line column
427 preconcentration and separation of cadmium, copper and lead for electrothermal atomic
428 absorption spectrometry. *Anal. Chim. Acta* 392, 55-65.

429 IBC Advanced Technologies, 2006. AnaLig® Data Sheet: As-01 PA. IBC Advanced Technologies,
430 Inc., Utah, USA.

431 IBC Advanced Technologies, 2007. AnaLig® Data Sheet: TE-01 and TE-02. IBC Advanced
432 Technologies, Inc., Utah, USA.

433 IBC Advanced Technologies, 2009. AnaLig® Data Sheet: AN-01 Si. IBC Advanced Technologies,
434 Inc., Utah, USA.

435 Izatt, R.M., 1997. Review of selective ion separations at BYU using liquid membrane and solid phase
436 extraction procedures. *J. Incl. Phenom. Macro.* 29, 197-220.

437 Izatt, R.M., Bradshaw, J.S., Bruening, R.L., Bruening, M.L., 1994. Solid phase extraction of ions of
438 analytical interest using molecular recognition technology. *Am. Lab.* 26, 28C-28M

439 Jitmanee, K., Oshima, M., Motomizu, S., 2005. Speciation of arsenic(III) and arsenic(V) by
440 inductively coupled plasma-atomic emission spectrometry coupled with preconcentration
441 system. *Talanta* 66, 529-533.

442 Karim, M., 2000. Arsenic in groundwater and health problems in Bangladesh. *Water Res.* 34, 304-310.

443 Karthikeyan, S., Prasada Rao, T., Iyer, C.S.P., 1999. Determination of arsenic in sea water by sorbent
444 extraction with hydride generation atomic absorption spectrometry. *Talanta* 49, 523-530.

445 Koh, J., Kwon, Y., Pak, Y.-N., 2005. Separation and sensitive determination of arsenic species
446 ($\text{As}^{3+}/\text{As}^{5+}$) using the yeast-immobilized column and hydride generation in ICP-AES.
447 *Microchem. J.* 80, 195-199.

448 Kumar, A.R., Riyazuddin, P., 2007. Non-chromatographic hydride generation atomic spectrometric
449 techniques for the speciation analysis of arsenic, antimony, selenium, and tellurium in water
450 samples - a review. *Int. J. Environ. An. Ch.* 87, 469-500.

451 Leal, L.O., Semenova, N.V., Forteza, R., Cerdà, V., 2004. Preconcentration and determination of
452 inorganic arsenic using a multisyringe flow injection system and hydride generation-atomic
453 fluorescence spectrometry. *Talanta* 64, 1335-1342.

454 Liang, P., Liu, Y., Guo, L., Zeng, J., Lu, H.B., 2004. Multiwalled carbon nanotubes as solid-phase
455 extraction adsorbent for the preconcentration of trace metal ions and their determination by
456 inductively coupled plasma atomic emission spectrometry. *J. Anal. Atom. Spectrom.* 19,
457 1489-1492.

458 Lintschinger, J., Schramel, P., Hatalak-Rauscher, A., Wendler, I., Michalke, B., 1998. A new method
459 for the analysis of arsenic species in urine by using HPLC-ICP-MS. *Fresen. J. Anal. Chem.*
460 362, 313-318.

461 Long, X.B., Miro, M., Hansen, E.H., Estela, J.M., Cerda, V., 2006. Hyphenating multisyringe flow
462 injection lab-on-valve analysis with atomic fluorescence spectrometry for on-line bead
463 injection preconcentration and determination of trace levels of hydride-forming elements in
464 environmental samples. *Anal. Chem.* 78, 8290-8298.

465 Mandal, B.K., Suzuki, K.T., 2002. Arsenic round the world: A review. *Talanta* 58, 201-235.

466 Mays, D.E., Hussam, A., 2009. Voltammetric methods for determination and speciation of inorganic
467 arsenic in the environment-A review. *Anal. Chim. Acta* 646, 6-16.

468 Munoz, E., Palmero, S., 2005. Analysis and speciation of arsenic by stripping potentiometry: a review.
469 *Talanta* 65, 613-620.

470 Nickson, R.A., Hill, S.J., Worsfold, P.J., 1995. Analytical perspective. Solid phase techniques for the
471 preconcentration of trace metals from natural waters. *Anal. Proc.* 32, 387-395.

472 Pergantis, S.A., Winnik, W., Betowski, D., 1997. Determination of ten organoarsenic compounds
473 using microbore high-performance liquid chromatography coupled with electrospray mass
474 spectrometry mass spectrometry. *J. Anal. Atom. Spectrom.* 12, 531-536.

475 Pozebon, D., Dressler, V.L., Gomes Neto, J.A., Curtius, A.J., 1998. Determination of arsenic(III) and
476 arsenic(V) by electrothermal atomic absorption spectrometry after complexation and sorption
477 on a C-18 bonded silica column. *Talanta* 45, 1167-1175.

478 Rahman, I.M.M., Nazim Uddin, M., Hasan, M.T., Hossain, M.M., 2008. Assimilation of arsenic into
479 edible plants grown in soil irrigated with contaminated groundwater. In: Bundschuh, J.,
480 Armienta, M.A., Birkle, P., Bhattacharya, P., Matschullat, J., Mukherjee, A.B. (Eds.). *Natural*
481 *Arsenic in Groundwaters of Latin America*. CRC Press/Balkema, Leiden, The Netherlands,
482 pp. 351-358.

483 Ritsema, R., Dukan, L., Navarro, T.R.I., van Leeuwen, W., Oliveira, N., Wolfs, P., Lebret, E., 1998.
484 Speciation of arsenic compounds in urine by LC-ICP MS. *Appl. Organomet. Chem.* 12, 591-
485 599.

486 Sanchez, W.M., Zwicker, B., Chatt, A., 2009. Determination of As(III), As(V), MMA and DMA in
487 drinking water by solid phase extraction and neutron activation. *J. Radioanal. Nucl. Ch.* 282,
488 133-138.

489 Smedley, P.L., Kinniburgh, D.G., 2002. A review of the source, behaviour and distribution of arsenic
490 in natural waters. *Appl. Geochem.* 17, 517-568.

491 Sohrin, Y., Iwamoto, S.-i., Akiyama, S., Fujita, T., Kugii, T., Obata, H., Nakayama, E., Goda, S.,
492 Fujishima, Y., Hasegawa, H., Ueda, K., Matsui, M., 1998. Determination of trace elements in
493 seawater by fluorinated metal alkoxide glass-immobilized 8-hydroxyquinoline concentration

494 and high-resolution inductively coupled plasma mass spectrometry detection. *Anal. Chim.*
495 *Acta* 363, 11-19.

496 Squibb, K.S., Fowler, B.A., 1983. The toxicity of arsenic and its compounds. In: Fowler, B.A. (Ed.).
497 *Biological and Environmental Effects of Arsenic*. Elsevier Science Publishers B.V., New
498 York, pp. 233-269.

499 Terlecka, E., 2005. Arsenic speciation analysis in water samples: A review of the hyphenated
500 techniques. *Environ. Monit. Assess.* 107, 259-284.

501 USEPA, 2002. Implementation Guidance for the Arsenic Rule - Drinking Water Regulations for
502 Arsenic and Clarifications to Compliance and New Source Contaminants Monitoring (EPA-
503 816-K-02-018). United States Environmental Protection Agency (USEPA), Washington, DC.

504 WHO, 2001. Environmental Health Criteria 224: Arsenic and Arsenic Compounds World Health
505 Organization (WHO), Geneva.

506 Yalcin, S., Le, X.C., 2001. Speciation of arsenic using solid phase extraction cartridges. *J. Environ.*
507 *Monit.* 3, 81-85.

508 Yan, X.-P., Yin, X.-B., He, X.-W., Jiang, Y., 2002. Flow injection on-line sorption preconcentration
509 coupled with hydride generation atomic fluorescence spectrometry for determination of
510 (ultra)trace amounts of arsenic(III) and arsenic(V) in natural water samples. *Anal. Chem.* 74,
511 2162-2166.

512 Yu, C.H., Cai, Q.T., Guo, Z.X., Yang, Z.G., Khoo, S.B., 2003. Inductively coupled plasma mass
513 spectrometry study of the retention behavior of arsenic species on various solid phase
514 extraction cartridges and its application in arsenic speciation. *Spectrochim. Acta B* 58, 1335-
515 1349.

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525 Table 1. Separation process of As(III), As(V), MMA and DMA using MRT gel SPE columns

Step	Function	Solution	Volume (mL)	Flow rate (mL min ⁻¹)
1	Rinsing 1	0.1 M HNO ₃	8	0.5
2	Rinsing 2	EPW	6	0.5
3	Conditioning	200 mM NaNO ₃ + 0.1 M buffer solution*	32–40	0.2–0.5
4	Collection	100 μM As-species spiked sample solution	4	0.2
5	Washing	EPW	4	0.2
6	Elution 1	<i>For TE-01 or AN-01 SPE columns</i> 1 M HNO ₃	2	0.2
		<i>For As-01 SPE column</i> 0.1 M HNO ₃	1	0.2
7	Elution 2	<i>For TE-01 or AN-01 SPE columns</i> 6 M HNO ₃	1	0.2
		<i>For As-01 SPE column</i> 2.0 M NaOH	1	0.2
8	Elution 3	<i>For TE-01 or AN-01 SPE columns</i> EPW	1	0.2
		<i>For As-01 SPE column</i> EPW	2	0.2

526 *MES Buffer (pH 4–6), HEPES Buffer (pH 7–8), TAPS Buffer (pH 9–10)

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535 Table 2. Effect of the sample loading flow-rates on the retention capacities (%) of the MRT
536 gel SPE columns

Flow rate (mL min ⁻¹)	TE-01	AN-01	As-01
0.20	101±3.8	100±3.7	101±4.6
0.25	99±3.0	100±3.4	100±4.3
0.30	88±2.8	82±2.6	92±3.8
0.50	75±2.4	71±2.7	87±2.9
1.00	74±1.8	68±3.2	82±2.7
2.00	65±2.6	62±1.6	71±3.4
4.00	62±3.2	59±1.8	68±2.2

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551 Table 3. Analysis of EC-JRC-IRMM CRMs for arsenic species

Arsenic species	Effluent Wastewater CRM		Groundwater CRM	
	BCR-713 ($\mu\text{g L}^{-1}$)		BCR-610 ($\mu\text{g L}^{-1}$)	
	This work	Certified value	This work	Certified value
As(III)	1.9±0.3	NR	3.3±0.6	NR
As(V)	7.1±1.2	NR	6.9±1.1	NR
MMA	BDL	NR	BDL	NR
DMA	0.4 ±0.1	NR	BDL	NR
Σ (As-species)	9.4±1.4	9.7±1.1	10.2±1.6	10.8±0.4

552 *'BDL' – Below Detectable Limit; 'NR'– Not reported

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567 Table 4. Determination of arsenic species in the fortified samples of 'real' waters

Arsenic species	Tap water			River water		
	Added ($\mu\text{g L}^{-1}$)	Found ($\mu\text{g L}^{-1}$)	Recovery (%)	Added ($\mu\text{g L}^{-1}$)	Found ($\mu\text{g L}^{-1}$)	Recovery (%)
As(III)	0	BDL	–	0	0.7±0.12	–
	19.5	19.3±0.10	99±0.5	20	19.4±0.41	99±2.1
As(V)	0	BDL	–	0	1.3±0.15	–
	31.2	31.3±0.48	100±1.5	31.2	30.5±0.51	98±1.6
MMA	0	BDL	–	0	BDL	–
	21.0	21.3±0.34	102±1.7	21.0	20.7±0.43	99±2.0
DMA	0	BDL	–	0	0.1±0.01	–
	32.1	32.3±0.27	101±0.8	32.1	31.9±0.60	99±1.9

568 *'BDL' – Below Detectable Limit

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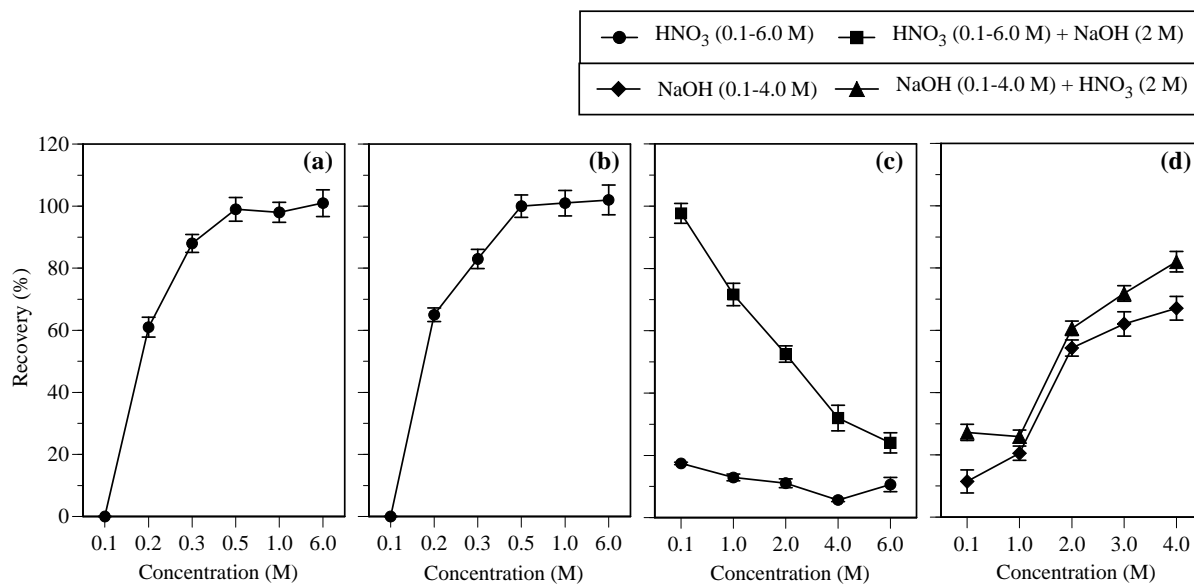
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581 Figure 1: Selection of eluent and eluent concentration: (a) AnaLig TE-01 (HNO₃–
 582 0.1/0.2/0.3/0.5/1.0/6.0 M) (b) AnaLig AN-01 Si (HNO₃– 0.1/0.2/0.3/0.5/1.0/6.0 M) (c)
 583 AnaLig As-01 PA (HNO₃– 0.1/1.0/2.0/4.0/6.0 M + NaOH– 2.0 M) (d) AnaLig As-01 PA
 584 (NaOH– 0.1/1.0/2.0/3.0/4.0 M + HNO₃– 2.0 M). Sample solution– As(V) (100 μM), matrix–
 585 H₂O, pH– 5, sample volume– 4 mL, flow rate– 0.2 mL min⁻¹ (n =3).

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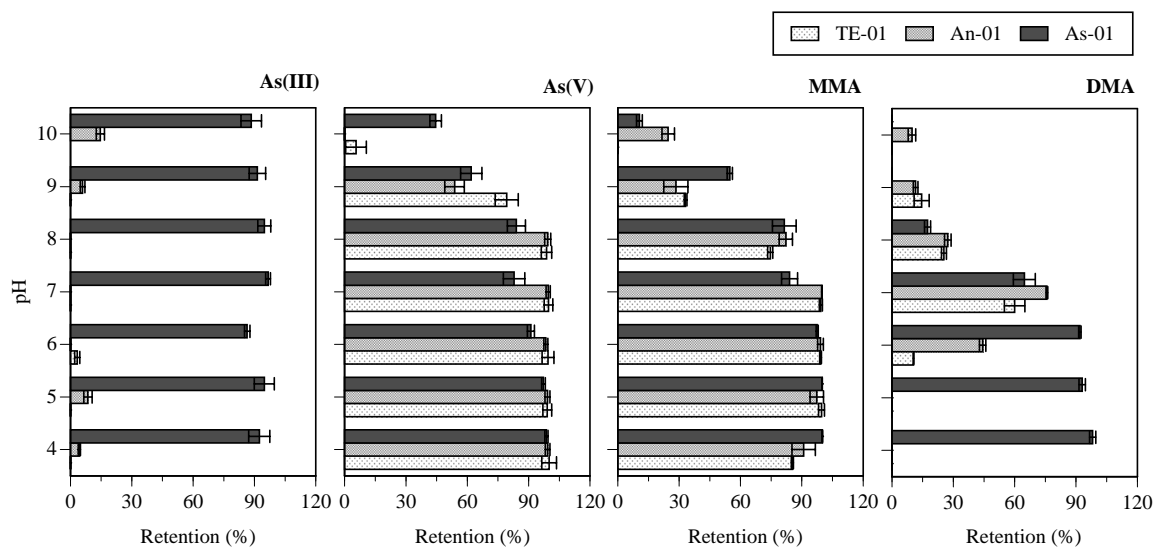
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595 Figure 2: Retention behavior of the MRT gel SPE columns. Sample solution– As(III), As(V),
 596 MMA and DMA (100 μ M), matrix– H₂O, pH– 4 to 10, sample volume– 4 mL, flow rate– 0.2
 597 mL min⁻¹, elution– 1.0 M HNO₃ (2 mL) + 6.0 M HNO₃ (1 mL) + EPW (1 mL), for TE-01
 598 and AN-01 SPE columns and 0.1 M HNO₃ (1 mL) + 2.0 M NaOH (1 mL) + EPW (2 mL), for
 599 As-01 SPE column ($n=3$).

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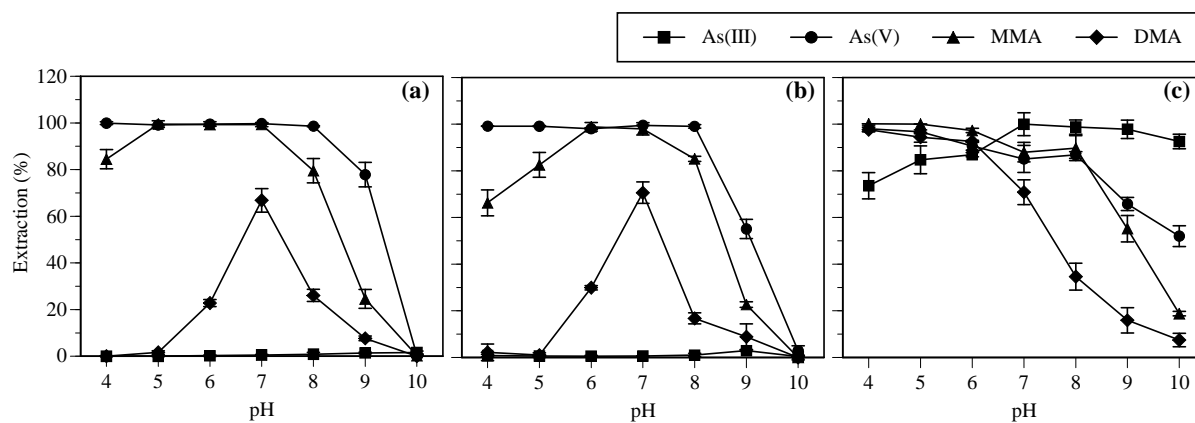
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611 Figure 3: Extraction behavior of the MRT gel SPE columns: (a) AnaLig TE-01, (b) AnaLig
 612 AN-01 Si and (c) AnaLig As-01 PA. Sample solution– As(III), As(V), MMA and DMA (100
 613 μM), matrix– H_2O , pH– 4 to 10, sample volume– 4 mL, flow rate– 0.2 mL min^{-1} , elution–
 614 1.0 M HNO_3 (2 mL) + 6.0 M HNO_3 (1 mL) + EPW (1 mL), for TE-01 and AN-01 SPE
 615 columns and 0.1 M HNO_3 (1 mL) + 2.0 M NaOH (1 mL) + EPW (2 mL), for As-01 SPE
 616 column ($n = 3$).

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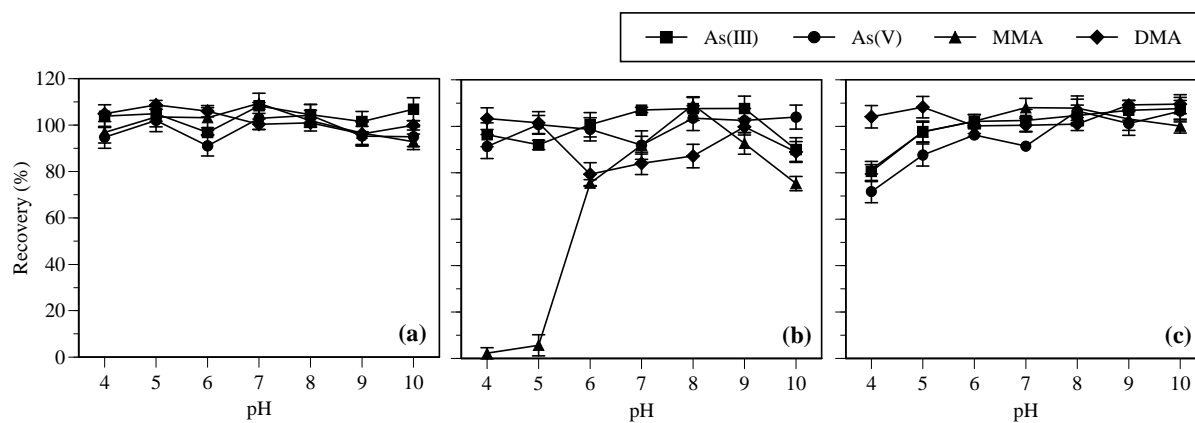
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629 Figure 4: Recovery behavior of the MRT gel SPE columns: (a) AnaLig TE-01, (b) AnaLig
 630 AN-01 Si and (c) AnaLig As-01 PA. Sample solution– As(III), As(V), MMA and DMA (100
 631 μM), matrix– H_2O , pH– 4 to 10, sample volume– 4 mL, flow rate– 0.2 mL min^{-1} , elution–
 632 1.0 M HNO_3 (2 mL) + 6.0 M HNO_3 (1 mL) + EPW (1 mL), for TE-01 and AN-01 SPE
 633 columns and 0.1 M HNO_3 (1 mL) + 2.0 M NaOH (1 mL) + EPW (2 mL), for As-01 SPE
 634 column ($n = 3$).

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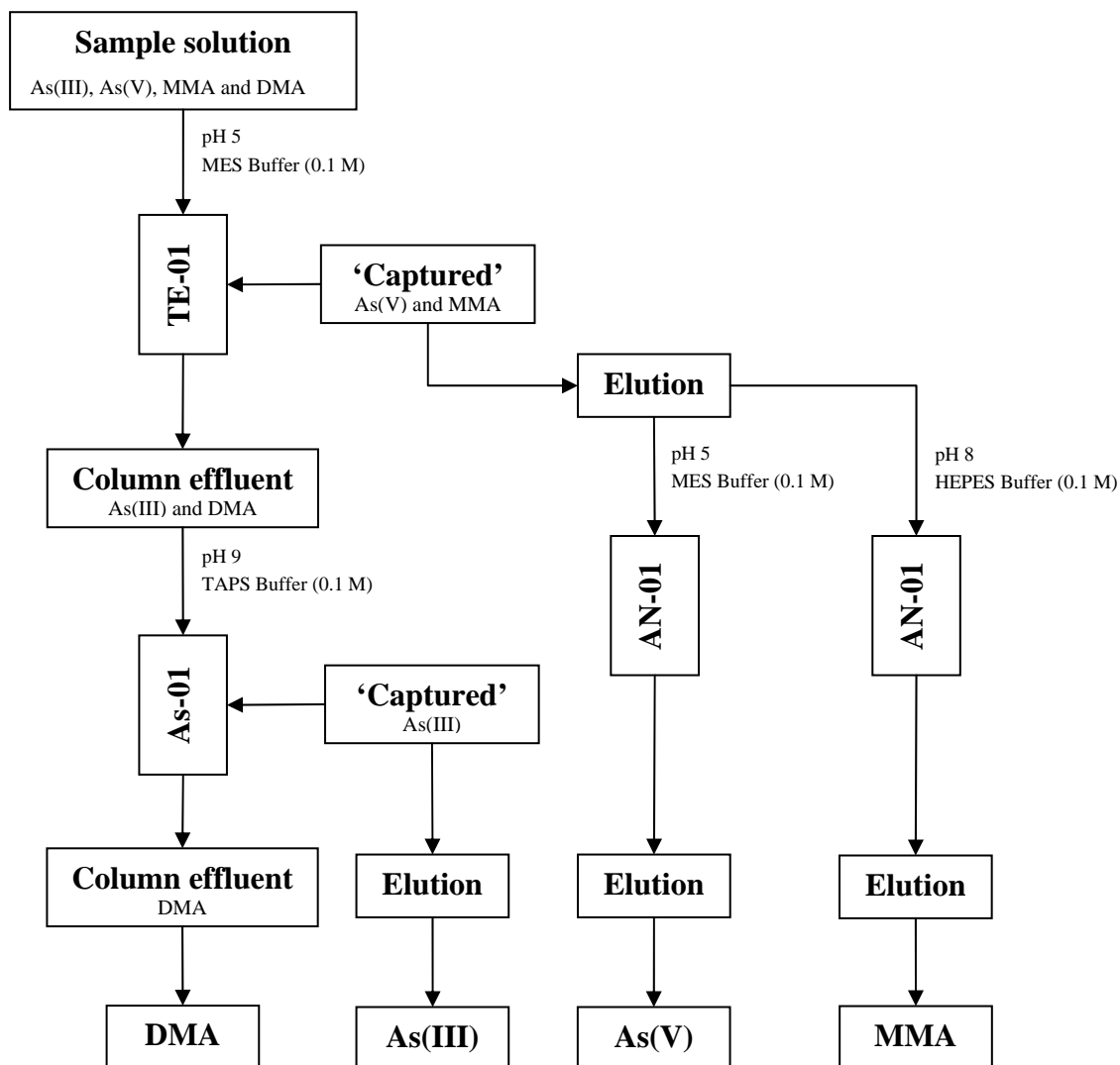


Figure 5: Scheme for selective separation of the arsenic species by MRT gel SPE columns