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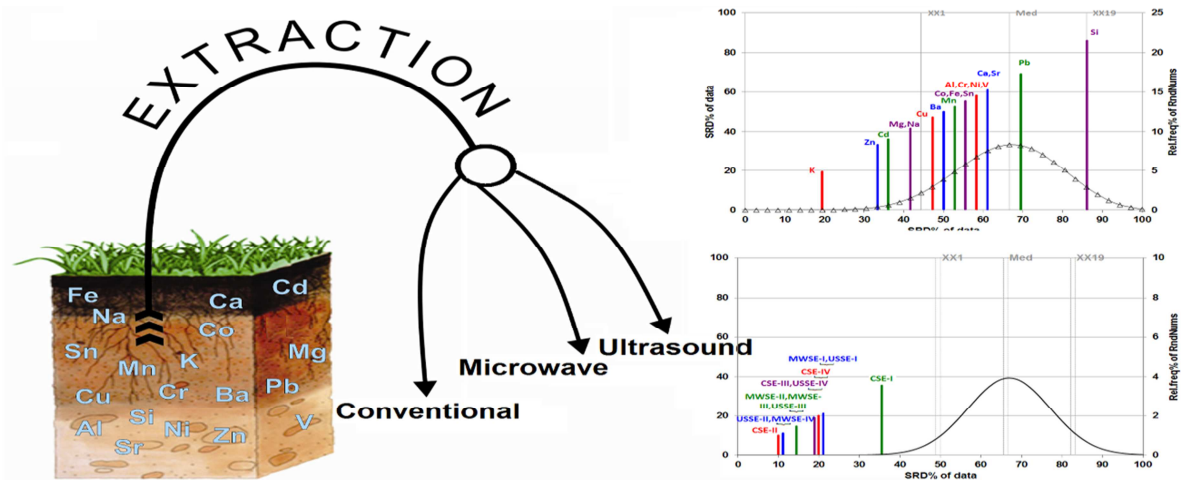
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1 **Ranking and similarity of conventional, microwave and ultrasound element**
2 **sequential extraction methods**

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20
21 **Abbreviations:** **SE**, sequential extraction; **BCR**, Community Bureau of References; **CSE**,
22 conventional sequential extraction; **MWSE**, microwave-assisted sequential extraction; **USSE**,
23 ultrasound-assisted sequential extraction; **SRD**, sum of the ranking differences; **CRRN**,
24 comparison of ranks with random numbers.

25 **Abstract**

26 This study aims to compare three extraction techniques of four sequential element extraction
27 steps from soil and sediment samples that were taken from the location of the Pančevo
28 petrochemical industry (Serbia). Elements were extracted using three different techniques:
29 conventional, microwave and ultrasound extraction. A novel procedure – sum of the ranking
30 differences (SRD) – was able to rank the techniques and elements, to see whether this method is
31 a suitable tool to reveal the similarities and dissimilarities in element extraction techniques,
32 provided that a proper ranking reference is available. The concentrations of the following
33 elements Al, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Na, Ni, Pb, Si, Sn, Sr, V and Zn were
34 determined through ICP OES. The different efficiencies and recovery values of element
35 concentrations using each of the three extraction techniques were examined by the CRM BCR-
36 701. By using SRD, we obtained a better separation between the different extraction techniques
37 and steps when we rank their differences among the samples while lower separation was
38 obtained according to analysed elements. Applying this method for ordering the elements could be
39 useful for three purposes: (i) to find possible associations among the elements; (ii) to find
40 possible elements that have outlier concentrations or (iii) detect differences in geochemical
41 origin or behaviour of elements. Cross-validation of the SRD values in combination with cluster
42 and principal component analysis revealed the same groups of extraction steps and techniques.

43

44 **Keywords:** sequential extraction, steps and techniques comparison, ordering, pattern recognition

45

46 **1 Introduction**

47 The most important analytical tool for obtaining information on element mobility is the
48 sequential extraction (SE) technique. The outcome of SE can provide information on the
49 bioavailability and possible toxicity of hazardous elements in the environment. Soil and sediment
50 samples received much scientific attention because of the significance of element pollution and
51 the threat pollution poses to human health. There are many available SE techniques, with the
52 currently most commonly used variety being the SE technique proposed by the European
53 Community Bureau of References (BCR SE) (Alonso Castillo et al., 2011; Bacon and Davidson,
54 2008; Canepari et al., 2005; Ciceri et al., 2008; De Andrade Passos et al., 2011; Jamali et al.,
55 2009; Martínez-Fernández et al., 2011; Nemati et al., 2011; Passos et al., 2010; Pérez-Cid et al.,
56 1999; Relić et al., 2013a, 2013b; Tokalioğlu et al., 2010). With used SE we determined acid
57 soluble, reducible, oxidizable and residual fractions of elements in four subsequent steps from
58 soil and sediment samples.

59 Typically BCR SE applies an overhead rotary mixing technique. Due to its common usage, this
60 approach is also referred to as the conventional SE (CSE). As this method is time-consuming,
61 alternative sources of energy (microwaves, ultrasound) are often used to reduce treatment times
62 and obtain greater reproducibility. Some authors have achieved encouraging results either
63 ultrasound probes to successfully reduce the time required for the SE of metals from sediments
64 (Davidson and Delevoye, 2001; Kazi et al., 2006; Pérez-Cid et al., 1998; Remeteiová et al.,
65 2008), or ultrasound baths to reduce the extraction time of each BCR step (30 min per step)
66 (Arain et al., 2008; Kazi et al., 2006; Leśniewska et al., 2016). In some studies, good recovery of
67 investigated metals was observed with microwave power lower than 170 W (Arain et al., 2008;
68 Leśniewska et al., 2016; Pérez-Cid et al., 2001; Relić et al., 2013a, 2013b), while other reports
69 indicate the need for higher value ranging from 180 W to (the more common) 560 W (Jamali et
70 al., 2009; Real et al., 1994; Reid et al., 2011).

71 Our study focussed on the area the Pančevo petrochemical plant and surroundings, located in the
72 town of Pančevo, Vojvodina, the northernmost province of the republic of Serbia, about 4 km
73 from the Danube River and 18 km (north-eastern) from the capital Belgrade.

74 In previous studies we have already demonstrated that the area of the Pančevo petrochemical
75 plant is a moderately polluted (Relić et al., 2013a, 2013b, 2011). In the present study, after
76 determining the elemental concentrations in the soil and sediment samples from our study area

77 we applied a novel procedure for determining similarities and differences in the behavior of
78 elements extracted by conventional sequential extraction (CSE), microwave-assisted sequential
79 extraction (MWSE) and ultrasound-assisted sequential extraction (USSE). This novel procedure
80 is based on the sum of the (absolute) ranking differences (SRD) and is capable of ordering and
81 grouping the techniques and elements, e.g., the concentration of elements obtained in samples
82 after four SE steps and three extraction techniques.

83 SRD has already been applied successfully in various scientific disciplines for solving different
84 method and model comparison problems. It's applications include: column selection in
85 chromatography (the original problem for which it was developed by Héberger (2010),
86 comparing of quantitative structure–*activity* relationship (QSAR) models (Vračko et al., 2010),
87 descriptor selection in QSAR (Kar and Roy, 2012; Ojha and Roy, 2011), comparing quantitative
88 structure–*retention* relationship models (Djaković-Sekulić et al., 2012; Garkani-Nejad and
89 Ahmadvand, 2011; Héberger, 2010; Héberger and Škrbić, 2012), comparing of quantitative
90 structure–*property* relationship models and, the predicting NMR chemical shifts (Liu et al.,
91 2011), predicting solubility (Bolboaca and Jantschi, 2010), comparing computer procedures for
92 estimating octanol–water partition coefficients (Ačanski et al., 2011; Andrić et al., 2016; Andrić
93 and Héberger, 2015a, 2015b; Héberger and Kollar-Hunek, 2011), clustering polarity measures
94 (Héberger and Zenkevich, 2010) including Hansen's solubility parameters (Bielicka-
95 Daszkiewicz et al., 2010), checking evaluation panels in food chemistry (Kollar-Hunek et al.,
96 2008; Sipos et al., 2011), ranking sensory-principal component 1 scores (Wood et al., 2010),
97 performance testing of Raman spectral resolution (Vajna et al., 2012), comparing chemometric
98 methods in near infra-red spectroscopy (Gowen et al., 2011), comparing curve resolution
99 techniques (Vajna et al., 2011), ranking of polycyclic aromatic hydrocarbons (PAHs) and
100 polluted environmental sites (Rocha et al., 2011), biochemical assay comparisons (ELISA
101 Veratox), and liquid chromatography for determining mycotoxin content (Tangni et al., 2011).

102 The calculations (ordering) were made using an Excel macro freely downloadable from
103 <http://aki.ttk.mta.hu/srd> together with sample input and output files (Héberger and Kollar-Hunek,
104 2011).

105 Finally, we applied our method for the first time to this data to rank and group three extraction
106 techniques based on the similarities in extraction capacities and to rank and groups elements to

107 recognise similar of different geochemical origin and behaviour in analysed samples. The
108 clustering pattern was justified by cluster and principal component analysis.

109 **2 Materials and Methods**

110 **2.1 Description of the study area and samples**

111 A total of 41 samples of soil and sediments were collected from the area of the Pančevo
112 petrochemical industry (Relić et al., 2013a, 2013b, 2011). .

113 The samples were packed in pouches and stored at 4 °C in order to prevent changes in the
114 chemical composition of samples. The elements for which we analysed using inductively
115 coupled plasma/optical emission spectrometry (ICP OES) were: Al, Ba, Ca, Cd, Co, Cr, Cu, Fe,
116 K, Mg, Mn, Na, Ni, Pb, Si, Sn, Sr, V and Zn.

117

118 **2.2 Sequential extraction techniques**

119 Solutions for extraction were prepared using analytical grade reagents (Carlo Erba). The
120 conditions applied in each of the three steps of the BCR extraction protocol, as well as the
121 pseudo-total quantity of elements in solid residues after three BCR steps were presented
122 previously (Relić et al., 2013a, 2013b). CSE was obtained during 16 h through over-head
123 extraction of solids, requiring the use of reagent for BCR extraction. The ratio solid to solution
124 was 1:40, also for MWSE and USSE. During the first three BCR steps of MWSE, the applied
125 microwave power was 90W, applied during 120 s. This microwave radiation was applied after 2h
126 of sample digestion with hydrogen-peroxide, for determining the oxidizable fraction. For USSE,
127 ultrasonic waves at a power of 100 W for 30 minutes for each of the first two BCR steps
128 (determining acid soluble and residual fraction) and again after sample digestion with hydrogen-
129 peroxide in the third BCR step. The fourth step, *aqua regia* digestion, was identical for all
130 sequential extraction techniques: digestion of residues with a mixture of mineral acids on water
131 bath.

132

133

134

135 **2.3 Instrumentation**

136 The elements were determined using an iCAP 6500 Duo ICP OES (Thermo Scientific, United
137 Kingdom). The detector was an RACID86 charge injector device (CID). External standard
138 solutions were prepared from 1000 mg L⁻¹ stock metal solutions. For minimal interference, a
139 multi-element standard stock solution was prepared in which the ratios of the metals were the
140 same as in the samples. These multi-element standards were prepared in the same matrix as the
141 extracting reagents to minimize matrix effects. Blanks were prepared for background correction.
142 Reagent blanks for all extractants were analyzed in parallel with all samples and found to have
143 negligible levels of the studied elements. The instrumental calibration was checked after every
144 10–12 samples.

145 Analytical wavelengths for each element were optimized daily before calibrating the instrument.
146 The ICP OES was calibrated using an acid blank and metal standard. For all trace elements
147 calibration curves the square of the correlation coefficient (R^2) was ≥ 0.995 .

148 The acid matrix baseline correction wavelengths for each metal were selected by comparing the
149 observed signal intensities with the acid blank, analyte standard and sample digestion solutions.
150 The following sequence of analyses was adhered to: first the blank, then the standards and at the
151 end the samples in sequence. The blank intensity was subtracted from both the standard and the
152 sample intensities. All elements were measured at the most appropriate wavelength, which was
153 determined by the estimated composition. The sensitivity was maximized and the spectral
154 overlap of elements was avoided as much as possible.

155 A microwave oven (Electrolux model 2100 S, 800 W power) was used as a microwave radiation
156 source. For extraction with ultrasound, we used a J.P. Selecta ultrasonic bath, with a maximum
157 frequency of 42 kHz applied over a period of 30 minutes during extraction, and an effective
158 power of 100W, without heating. A centrifuge (Tehtnica, Železniki) was used during 10 min at
159 3000 x g separate the extracts.

160

161 **2.4 Certified reference materials**

162 Certified reference material was supplied by The Community Bureau of Reference Samples
163 (BCR): BCR-701. The certified material was handled according to the supplier's specifications.

164

165 **2.5 Ranking and pattern recognition methods**

166 The SRD procedure is entirely general, and it is supervised in the sense that a reference
167 (benchmark) ranking should be available. The data should be arranged in matrix form. We
168 arranged two matrices consisting of objects and variables, with the objects placed in the rows and
169 the variables in the columns of the input matrix. The first matrix contained 19 objects, which
170 represented the averaged element concentrations, while the 12 variables represented the BCR
171 steps with adequate extraction techniques. The second matrix contained 41 objects,
172 representative of 41 samples and again 12 variables were the BCR steps with adequate extraction
173 techniques. After arranging the matrices, the second important step is the selection of a reference
174 (benchmark) for ranking. We chose to use the average of all objects as a reference for the
175 ranking in order to obtain a consensus ranking; random errors cancel each other out, and
176 systematic errors of different laboratories and/or various measurement methods also cancel each
177 other. All samples were extracted at the same time for each extraction technique and all element
178 concentrations were measured on the same scale; hence, no data pre-processing was necessary.
179 For the matrices, an averaged value was calculated for each row. These averaged values were
180 added as an additional column after the last column in each matrix. However, the average is not
181 necessarily an unbiased solution; also minimum or maximum value, or some read value can be
182 used as reference value for ranking. Every variable in the two matrices was ranked and compared
183 to the known reference (average) values. The absolute differences between the average and
184 individual rankings were then calculated and summed for each technique.

185 The closer the SRD value is to zero the closer the variables are to the reference value. If
186 techniques have similar SRD values this means that those techniques are similar to each other, in
187 the sense that they could have a similar impact on the extraction of elements.

188 The calculations (ordering) were made using an Excel macro freely downloadable from
189 <http://aki.ttk.mta.hu/srd> together with sample input and output files (Héberger and Kollar-Hunek,
190 2011).

191 Cluster, principal component and correlation analyses were also used to reveal and so validate
192 the grouping pattern in the data. All the calculations were made using Statistica v7.0 (Tulsa
193 Oklahoma, USA).

194

195 **3 Results and Discussion**

196 **3.1 Sequential extraction of soil and sediment samples**

197 In Table S1, we present concentrations of elements obtained after four SE steps using three
198 extraction techniques. According to obtained results, it can be seen that the highest
199 concentrations of analysed elements were detected after the fourth SE step, while for the
200 majority of them the lowest values were detected in the first SE step, after extraction of an acid
201 soluble fraction. Ca and Sr behaviours differently, these metals were extracted in highest
202 quantities in an acid soluble fraction, because of carbonate dissolution. Comparing different
203 techniques in each step, within the first SE, for a majority of elements, CSE was the technique
204 that obtains the highest element concentrations. After the second and the third step, the highest
205 values were detected after CSE also for the majority of elements.

206 **3.2 Certified material**

207 The concentrations of metals in the certified BCR material is presented in Relić et al. (2013a) as
208 a mean value with one standard deviation, accuracy, precision and recovery values for all four
209 SE steps and three extraction techniques. CSE yield a good accuracy for certified reference
210 metals for all BCR steps. Also USSE gave good results there, except for the extracted quantities
211 after the second BCR step. The MWSE technique had the lowest agreement with certificate
212 concentrations among all BCR steps (Relić et al., 2013a).

213 In Table 1 we present the sum total extracted quantities of certified reference metals, obtained by
214 CSE, MWSE and USSE after four SE steps and the direct pseudo-total quantity of BCR 701 by
215 *aqua regia* from Sutherland (2010), to calculate the recovery values for each technique using this
216 equation:

$$217 \text{ Recovery} = ([\text{step 1} + \text{step 2} + \text{step 3} + \text{step 4}] / \text{pseudo-total concentration}) \times 100 \quad (1)$$

218 The lowest recovery values were obtained for Cr: for CSE 41%, for MWSE 42% and for USSE
219 47% and for Ni: for CSE 74.0 %, for MWSE 73.2 % and for USSE 63.8 % (Table 1), the other
220 metals have accuracy values above 80%.

221 Apart from Cr and Ni, Cu was the third metal to have recovery values of less than 90% for all
222 techniques (Relić et al., 2013b). Each of Cr and Ni concentrations were recalculated according to
223 the discrepancy in the measured values from its relevant value in the reference material. For all

224 certified reference metals, the highest recovery values were obtained after CSE with values for
225 Cd and Pb close to or or higher than 100% (Table 1), while for the other metals and techniques,
226 these values were lower. This agrees with recovery 1 and 2 from Relić et al. (2013a), which were
227 also lower than 100%. The highest metal quantities were thus extracted by CSE, and more
228 closely to certified values, rather than using the faster techniques, while the closest recovery
229 values were obtain for time saving techniques, for most of metals except for Cr and Ni. Lead was
230 the only metal that has microwave and ultrasound extraction quantities more closely to certified
231 reference value.

232 3.3 SRD ranking

233 3.3.1 SRD ranking – techniques and BCR steps vs. elements

234 In our matrices, the rows contained the element concentrations (the averaged value of their
235 quantities from all samples for an adequate extraction step and technique) while the columns
236 contained the four SE steps which we will refer to SE-I, SE-II, SE-III and SE-IV, as well as the
237 three extraction methods: using microwaves (MW), ultrasound (US) and conventional rotary
238 mixing (C). A notation with Roman numerals next to abbreviations of the extraction technique
239 represents the corresponding step in the BCR SE.

240 A simple SRD ranking is shown in Figure 1.

241
242 Figure 1

243 CSE-II was most similar to the average of all techniques; the elemental concentrations extracted
244 using this technique and after the second SE step were hardly different from the averaged values,
245 and the sum of all SRD values was the lowest overall. The techniques with artificial sources of
246 energy applied in the second and fourth sequential steps were ranked in the first subgroup
247 (USSE-II and MWSE-IV, both are indistinguishable from one another through SRD ranking).
248 The next grouping had three indistinguishable extraction techniques: MWSE-II, MWSE-III and
249 USSE-III, and the fourth grouping consisted of CSE-III and USSE-IV. CSE-IV fairly belongs to
250 either of groups and MWSE-I and USSE-I is indistinguishable as well. When techniques were
251 indistinguishable it could indicate the existence of similarity in the rank differences of extraction
252 element concentrations after the usage of these techniques. CSE-I was farthest from the other
253 techniques but below the theoretical distribution function of random numbers (black curve in

254 Figure 1). This indicates that there should not be significant difference between this and other
255 techniques. When we compared techniques applied in the first BCR step, MWSE-I and USSE-I
256 were closer to each other than to CSE-I (Figure 1), i.e. the SRD for MWSE-I and USSE-I was
257 lower than for CSE-I. This similar SRD ranking of MWSE-I and USSE-I (Figure 1) indicates
258 that the effect of two artificial sources of energy on the most mobile fraction of elements were
259 similar, considering they differed from the averaged values in the same way. The closest and
260 farthest techniques were conventional ones according to SRD ranking. Considering that CSE is
261 the recommended technique for BCR SE together with fact that those techniques which use
262 artificial sources of energy were located between CSE-II and CSE-I, confirms their applicability
263 for this kind of analysis.

264 However, the ordering process comes with some uncertainties that can significantly affect the
265 grouping pattern. A sevenfold cross-validation (approximately seven-fold split) is suitable for
266 assigning uncertainties to the rank numbers: approximately one-seventh of the elements were left
267 out and the ranking was completed in a similar way to the full data set. The procedure was
268 repeated seven times, yielding seven rankings with six-sevenths of the data and one full ranking.
269 Uncertainties were assigned to the SRD values. The next figure (Figure S1) shows the SRD
270 values with uncertainties in increasing order.

271
272 Figure S1

273 The sevenfold cross-validation distributed the sample extraction methods into four groups and all
274 techniques were grouped according to their significance (Figure 2). These groups are as follows:
275 CSE-II, USSE-II and MWSE-IV from group I; MWSE-II, MWSE-III and USSE-III from group
276 II; group III contains CSE-III, USSE-IV, CSE-IV, MWSE-I and USSE-I; and CSE-I can be
277 considered as 'group IV' (Figure S1). Comparing these groups there are two extraction
278 techniques for each step in each group: first step, MWSE-I and USSE-I (group III); second step,
279 CSE-II and USSE-II (group I); third step, MWSE-III and USSE-III (group I) and fourth step,
280 CSE-IV and USSE-IV (group III).

281 Figure S2 shows a hierarchical cluster analysis of the data matrix that was used in case of SRD
282 ranking).

283
284 Figure S2

285 The interpretation of results from a cluster analysis always has some arbitrary element. A
286 horizontal line below 10000 (arbitrary distance units) defines four clusters. The left clusters
287 consist of the techniques of the fourth BCR step (MWSE-IV, USSE-IV and CSE-IV), while the
288 next two sub-clusters include the techniques after the second and third BCR steps. The right
289 cluster has a sub-cluster that is below 10000 and which consists of MWSE-I and USSE-I while
290 CSE-I is attached to this above a linkage distance of 10000 (Figure S2). A cluster analysis
291 provides similar groupings to SRD clustering because of the proximity of the lines for CSE-IV
292 and USSE-IV, and also of the lines for MWSE-III and USSE-III, and USSE-I and MWSE-I
293 (Figure S1 and Figure S2).

294 The grouping pattern is similar to that of Figure S2 when we consider the score plot of a
295 principal component analysis for the same input data (Figure S3). The most influential points in
296 the top-left part of the figure panel belong to CSE-IV, USSE-IV, MWSE-IV and CSE-III, and on
297 the other side, we find CSE-I, MWSE-I, USSE-I, MWSE-II, USSE-II, MWSE-III, USSE-III and
298 CSE-II.

299

300 Figure S3

301 Based on the cluster and principal component analysis, a more similar distribution of extracted
302 element concentrations was achieved by using the techniques that used artificial sources of
303 energy after the second and third BCR steps (Figure S2 and Figure S3) which is also visible from
304 the CRRN and SRD results (Figure 1 and Figure S1). In this sense the hierarchical cluster and
305 principal component analysis both validate the SRD and CRRN findings and we can conclude
306 that these latter two techniques provide a ‘natural’ ordering of techniques and not just groupings.

307 **3.3.2 SRD ranking – elements vs. techniques and BCR steps**

308 To illustrate the versatility and usefulness of the SRD methodology, we transposed the first
309 matrix in order to examine the similarity and dissimilarity between the elements from the point
310 of view of SE steps and techniques. Figure 2 shows the grouping of the extracted elements by
311 SRD.

312

313 Figure 2

314 A hypothetical ‘average concentration of the extractable elements after all steps and techniques’
315 was calculated as the row average of the transposed matrix and chosen as a reference for
316 ranking. Again, SRD values (x -axis and left y -axis) were scaled to range from 0 to 100. The right
317 y -axis of Figure 2 contains the relative frequencies for the theoretical distribution function.
318 Potassium was the closest to the ‘average’ metal; its quantities obtained after all techniques and
319 all steps were the least different from the ranking order of average element concentrations. The
320 metals that were below the XX1 (5% probability line) value were Zn, Cd as well as Mg and Na.
321 The other group consists of Cu, Ba and Mn and three sub-groups consist of elements Co, Fe and
322 Sn; Al, Cr, Ni and V; and Ca and Sr were the sub-groups closest to a median value of averaged
323 metal concentrations (Figure 2). These proximity to median value of averaged metal
324 concentrations could indicate similar a geochemical origin or association between elements. The
325 correlation coefficients for all analyzed are shown in Table S2. Commensurate with findings in
326 previous studies (Alfaro et al., 2015; Sipos et al., 2014) we also found an association between
327 Fe, Sn and Co. The associations between Al, Cr, Ni and V (Tumuklu et al., 2007 and Table S2)
328 can indicate similar geochemical behaviour, while the association between Ca and Sr (Table S2)
329 could represent the carbonate fraction in analyzed samples. The difference between these three
330 sub-groups of elements is that for the case of Ca and Sr, the highest concentrations were obtained
331 after the first BCR step (Table S1), which could indicate a carbonate origin for these two metals,
332 while for the other two sub-groups, the highest concentrations were obtained after the fourth
333 BCR step.

334 The remaining metals (Pb and Si) were located on the tail-end of a Gaussian-like curve, i.e., they
335 were ranked contrary to all other elements. Standing isolated Zn, Cd, Cu, Ba, Mn and Pb could
336 indicate that these metals behave differently from the macro-elements (Al, Ca, Mg, Fe, Si and
337 K). Not only a similarity in the CRRN analysis indicates associations or similar geochemical
338 behaviour: if elements are isolated (Figure 2), this could be because of the existence of outliers;
339 some of the analysed samples had higher concentrations of these metals, which was shown in our
340 previous work (Relić et al., 2011, 2013a) especially for Zn, Cu, Pb and Ba. A significant
341 positive correlations exists between Zn, Cu, Mn and Ba (Table S2). Pb, on the other hand, it is
342 not correlated to these metals (Table S2), and has more similarities with the macro-elements Al
343 and Fe. Pb is closer to the Fe and Al groups than to Zn, Cu, Mn or Ba (Figure 2).

344 In the cross-validation of SRD values that left out one seventh of the elements (Figure S4) we
345 observed a quasi-continuous increase of the SRD values for elements, but the borderlines can be
346 defined easily. The borderlines were not set arbitrarily but using a Wilcoxon matched pair test
347 for the cross-validated rankings. A significant difference (at the 5 % level) was found for Pb and
348 Si, and also between K, Zn, Cd, Na and Mg, and between Cu and Ba, whereas the rest of the
349 elements are statistically indistinguishable (Figure S4).

350

351 Figure S4

352 The dendrogram of a cluster analysis of cross-validated SRD values reveal the same pattern
353 (Figure S5). Three groups can be identified below a horizontal line at 200 arbitrary distance
354 units. The group of K, Na, Mg, Zn and Cd, elements that were closest to the average element
355 concentrations, are grouped in a sub-cluster.

356

357 Figure S5

358 3.3.3 SRD ranking – techniques and BCR steps vs. samples

359 We applied the SRD ranking method also to the matrix that contained samples in 41 rows and
360 the BCR steps and techniques in 12 columns. The numerical values assigned to each sample
361 represented the sum of its element concentrations, while the columns contained the four SE steps
362 of the three extraction methods: microwaves, ultrasound and conventional rotary mixing.

363 The SRD ranking can be seen in Figure 3.

364

365 Figure 3

366

367 A slightly different situation was observed when SRD ranking was performed on the samples.
368 CSE-II was closest to the average value (as was observed when ranking averaged concentrations
369 of each element) i.e., the sum of all extracted elemental quantities was closest to the averaged
370 values, and the sum of SRD values was the lowest. In comparison to Figure 1, there are non-
371 overlaying techniques and BCR steps, as was the case for the first data matrix, with a better
372 separation having been achieved between the techniques and steps. All techniques in the first and
373 fourth extraction steps were below the XX1 value with USSE-III, CSE-III and USSE-II closest

374 to, but still below XX1. MWSE-III and MWSE-II were the farthest from the averaged reference
375 values. This statistical difference of MWSE-II and MWSE-III from the rest of the techniques
376 and BCR steps is in good agreement with the results from the extraction of BCR certified
377 material, where the lowest precision was obtained for these two microwave extractions (Relić et
378 al., 2013a). In order to understand why these two microwave techniques were so different from
379 the averaged values, we correlated the ranking difference of their technique and step with the
380 ranking differences of each element extracted for that same technique and step.

381 We calculate the element ranking difference by determining the absolute difference between the
382 ranking values of element concentrations and the ranking value for each sample for an adequate
383 step and technique. This was performed for MWSE-II and CSE-II, and for MWSE-III and CSE-
384 III with results shown in Table S3. The ranking difference for MWSE-II was positively (and
385 significantly) correlated with the ranking differences for Ca and Mg, while for the majority of the
386 remaining elements the correlation was negatively and non-significant. In the case of MWSE-III,
387 the ranking differences was positively and significant correlated with Ca, Sr, and Mg, while the
388 remaining elements showed no significant correlation. This absence of a significant positive
389 correlation indicates that the conditions applied in the second and the third BCR steps influenced
390 the element concentrations in ways that were different from other techniques and BCR steps. It
391 can therefore be concluded that the conditions applied in MWSE-II and MWSE-III should be
392 changed in order to achieve a more uniform distribution. A good example for how the conditions
393 of a technique in a step could be in agreement with extracted element concentrations was shown
394 for the case of CSE-II, and CSE-III (Table S3). The ranking differences for both these techniques
395 were significantly correlated with some of the elements, and, more importantly, they were also
396 positively correlated with the remaining elements (Table S3).

397 **4 Conclusion**

398 For the first time, the SRD procedure was used on this data to order and group extraction
399 techniques in BCR steps according to the elements and samples, providing an overall ordering
400 and clustering pattern. Cross-validation of the SRD values provided a good measure for
401 similarity/dissimilarity thereby enabling the grouping. Cluster and principal component analysis
402 also grouped the extraction techniques according to the sequential steps. A Wilcoxon matched
403 pair test and cross-validation of the SRD values and a cluster and principal component analysis

404 revealed essentially the same four groups (or sub-clusters) based on the steps of the SE
405 (including extraction techniques), although the grouping was not exactly the same.
406 The second BCR sequential step, conducted by conventional extraction, had the lowest ranking
407 differences, i.e., it was close to the average for all techniques and steps. The SRD analysis of
408 samples showed that the techniques involving two microwave extractions after the second and
409 third steps yielded results were significantly different from the averaged values, which was also
410 confirmed using the certified material. A correlation analysis confirmed a lower number of
411 significant positive correlations among ranking differences for these techniques and ranking
412 differences of extracted elements. This indicates that it may be advisable to change the
413 conditions applied for these techniques. Apart from ordering the techniques and BCR steps, the
414 SRD ranking method also ordered the elements. The ordering of elements yields information on
415 whether there are associations among them, or it can point to the existence of certain elements
416 that do not follow the distribution of the macro-elements when analysing a sample. This could
417 also point towards the existence of outliers. Based on our findings, we can conclude that this
418 novel procedure, SRD is a suitable tool for this of data as provides a more detailed analysis tool,
419 in particular when the data are complex. For the future study, we indent to apply SRD method to
420 obtain the ordering of samples, to see how they differs from each other and does the similar
421 geochemical origin or nature group samples or not.

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427 **References:**

428 Ačanski, M.M., Vujić, Đ.N., Jovanović-Šanta, S., 2011. Separation and lipophilicity of some
429 new steroid derivatives in normal- and reversed-phase high performance liquid
430 chromatography. *Chem. Ind. Chem. Eng. Q.* 17, 535–542.
431 <https://doi.org/10.2298/CICEQ110506039A>

- 432 Alfaro, M.R., Montero, A., Ugarte, O.M., do Nascimento, C.W.A., de Aguiar Accioly, A.M.,
433 Biondi, C.M., da Silva, Y.J.A.B., 2015. Background concentrations and reference values for
434 heavy metals in soils of Cuba. *Environ. Monit. Assess.* 187, 4198.
435 <https://doi.org/10.1007/s10661-014-4198-3>
- 436 Alonso Castillo, M.L., Vereda Alonso, E., Siles Cordero, M.T., Cano Pavon, J.M., Garcia de
437 Torres, A., 2011. Fractionation of heavy metals in sediment by using microwave assisted
438 sequential extraction procedure and determination by inductively coupled plasma mass
439 spectrometry. *Microchem. J.* 98, 234–239. <https://doi.org/10.1016/j.microc.2011.02.004>
- 440 Andrić, F., Bajusz, D., Rácz, A., Šegan, S., Héberger, K., 2016. Multivariate assessment of
441 lipophilicity scales—computational and reversed phase thin-layer chromatographic indices.
442 *J. Pharm. Biomed. Anal.* 127, 81–93. <https://doi.org/10.1016/j.jpba.2016.04.001>
- 443 Andrić, F., Heberger, K., 2015a. Chromatographic and computational assessment of lipophilicity
444 using sum of ranking differences and generalized pair-correlation. *J. Chromatogr. A* 1380,
445 130–138. <https://doi.org/10.1016/j.chroma.2014.12.073>
- 446 Andrić, F., Heberger, K., 2015b. Towards better understanding of lipophilicity: Assessment of in
447 silico and chromatographic logP measures for pharmaceutically important compounds by
448 nonparametric rankings. *J. Pharm. Biomed. Anal.* 115, 183–191.
449 <https://doi.org/10.1016/j.jpba.2015.07.006>
- 450 Arain, M.B., Kazi, T.G., Jamali, M.K., Jalbani, N., Afridi, H.I., Baig, J.A., 2008. Speciation of
451 heavy metals in sediment by conventional, ultrasound and microwave assisted single
452 extraction methods: A comparison with modified sequential extraction procedure. *J.*
453 *Hazard. Mater.* 154, 998–1006. <https://doi.org/10.1016/j.jhazmat.2007.11.004>
- 454 Bacon, J.R., Davidson, C.M., 2008. Is there a future for sequential chemical extraction? *Analyst*
455 133, 25–46. <https://doi.org/10.1039/b711896a>
- 456 Bielicka-Daszkiwicz, K., Voelkel, A., Pietrzynska, M., Heberger, K., 2010. Role of Hansen
457 solubility parameters in solid phase extraction. *J. Chromatogr. A* 1217, 5564–5570.
458 <https://doi.org/10.1016/j.chroma.2010.06.066>

- 459 Bolboaca, S.D., Jantschi, L., 2010. Diagnostic of a QSPR model: aqueous solubility of drug-like
460 compounds. *Stud. Univ. Babes-Bolyai Chem.* 69–76.
- 461 Canepari, S., Cardarelli, E., Ghighi, S., Scimonelli, L., 2005. Ultrasound and microwave-assisted
462 extraction of metals from sediment: A comparison with the BCR procedure. *Talanta* 66,
463 1122–1130. <https://doi.org/10.1016/j.talanta.2005.01.014>
- 464 Ciceri, E., Giussani, B., Pozzi, A., Dossi, C., Recchia, S., 2008. Problems in the application of
465 the three-step BCR sequential extraction to low amounts of sediments: An alternative
466 validated route. *Talanta* 76, 621–626. <https://doi.org/10.1016/j.talanta.2008.04.006>
- 467 Davidson, C.M., Delevoye, G., 2001. Effect of ultrasonic agitation on the release of copper, iron,
468 manganese and zinc from soil and sediment using the BCR three-stage sequential
469 extraction. *J. Environ. Monit.* 3, 398–403. <https://doi.org/10.1039/b103602p>
- 470 De Andrade Passos, E., Alves, J.D.P.H., Garcia, C.A.B., Costa, A.C.S., 2011. Metal fractionation
471 in sediments of the Sergipe River, Northeast, Brazil. *J. Braz. Chem. Soc.* 22, 828–835.
472 <https://doi.org/10.1590/S0103-50532011000500004>
- 473 Djaković-Sekulić, T., Mandić, A., Trišović, N., Uščumlić, G., 2012. Structure-Retention
474 Relationship Study of HPLC Data of Antiepileptic Hydantoin Analogues 3–9.
- 475 Garkani-Nejad, Z., Ahmadvand, M., 2011. Comparative QSRR modeling of nitrobenzene
476 derivatives based on original molecular descriptors and multivariate image analysis
477 descriptors. *Chromatographia* 73, 733–742. <https://doi.org/10.1007/s10337-011-1969-7>
- 478 Gowen, A.A., Downey, G., Esquerre, C., O'Donnell, C.P., 2011. Preventing over-fitting in PLS
479 calibration models of near-infrared (NIR) spectroscopy data using regression coefficients. *J.*
480 *Chemom.* 25, 375–381. <https://doi.org/10.1002/cem.1349>
- 481 Héberger, K., 2010. Sum of ranking differences compares methods or models fairly. *TrAC -*
482 *Trends Anal. Chem.* 29, 101–109. <https://doi.org/10.1016/j.trac.2009.09.009>
- 483 Héberger, K., Kollar-Hunek, K., 2011. Sum of ranking differences for method discrimination

- 484 and its validation: Comparison of ranks with random numbers. *J. Chemom.* 25, 151–158.
485 <https://doi.org/10.1002/cem.1320>
- 486 Héberger, K., Škrbić, B., 2012. Ranking and similarity for quantitative structure-retention
487 relationship models in predicting Lee retention indices of polycyclic aromatic
488 hydrocarbons. *Anal. Chim. Acta* 716, 92–100. <https://doi.org/10.1016/j.aca.2011.11.061>
- 489 Héberger, K., Zenkevich, I.G., 2010. Comparison of physicochemical and gas chromatographic
490 polarity measures for simple organic compounds. *J. Chromatogr. A* 1217, 2895–2902.
491 <https://doi.org/10.1016/j.chroma.2010.02.037>
- 492 Jamali, M.K., Kazi, T.G., Arain, M.B., Afridi, H.I., Jalbani, N., Kandhro, G.A., Shah, A.Q.,
493 Baig, J.A., 2009. Speciation of heavy metals in untreated sewage sludge by using
494 microwave assisted sequential extraction procedure. *J. Hazard. Mater.* 163, 1157–1164.
495 <https://doi.org/10.1016/j.jhazmat.2008.07.071>
- 496 Kar, S., Roy, K., 2012. First report on development of quantitative interspecies structure-
497 carcinogenicity relationship models and exploring discriminatory features for rodent
498 carcinogenicity of diverse organic chemicals using OECD guidelines. *Chemosphere* 87,
499 339–355. <https://doi.org/10.1016/j.chemosphere.2011.12.019>
- 500 Kazi, T.G., Jamali, M.K., Siddiqui, A., Kazi, G.H., Arain, M.B., Afridi, H.I., 2006. An ultrasonic
501 assisted extraction method to release heavy metals from untreated sewage sludge samples.
502 *Chemosphere* 63, 411–420. <https://doi.org/10.1016/j.chemosphere.2005.08.056>
- 503 Kollar-Hunek, K., Heszberger, J., Kokai, Z., Lang-Lazi, M., Papp, E., 2008. Testing panel
504 consistency with GCAP method in food profile analysis. *J. Chemom.* 22, 218–226.
505 <https://doi.org/10.1002/cem.1135>
- 506 Leśniewska, B., Kisielewska, K., Wiater, J., Godlewska-Żyłkiewicz, B., 2016. Fast and simple
507 procedure for fractionation of zinc in soil using an ultrasound probe and FAAS detection.
508 Validation of the analytical method and evaluation of the uncertainty budget. *Environ.*
509 *Monit. Assess.* 188, 1–13. <https://doi.org/10.1007/s10661-015-5020-6>

- 510 Liu, X., Ren, Y., Zhou, P., Shang, Z., 2011. Prediction of protein $^{13}\text{C}\alpha$ NMR chemical shifts
511 using a combination scheme of statistical modeling and quantum-mechanical analysis. *J.*
512 *Mol. Struct.* 995, 163–172. <https://doi.org/10.1016/j.molstruc.2011.04.012>
- 513 Martínez-Fernández, M., Barciela-Alonso, M.C., Moreda-Piñeiro, A., Bermejo-Barrera, P., 2011.
514 Matrix solid phase dispersion-assisted BCR sequential extraction method for metal
515 partitioning in surface estuarine sediments. *Talanta* 83, 840–849.
516 <https://doi.org/10.1016/j.talanta.2010.10.035>
- 517 Nemati, K., Bakar, N.K.A., Abas, M.R., Sobhanzadeh, E., 2011. Speciation of heavy metals by
518 modified BCR sequential extraction procedure in different depths of sediments from Sungai
519 Buloh, Selangor, Malaysia. *J. Hazard. Mater.* 192, 402–410.
520 <https://doi.org/10.1016/j.jhazmat.2011.05.039>
- 521 Ojha, P.K., Roy, K., 2011. Comparative QSARs for antimalarial endochins: Importance of
522 descriptor-thinning and noise reduction prior to feature selection. *Chemom. Intell. Lab.*
523 *Syst.* 109, 146–161. <https://doi.org/10.1016/j.chemolab.2011.08.007>
- 524 Passos, E. de A., Alves, J.C., dos Santos, I.S., Alves, J. do P.H., Garcia, C.A.B., Spinola Costa,
525 A.C., 2010. Assessment of trace metals contamination in estuarine sediments using a
526 sequential extraction technique and principal component analysis. *Microchem. J.* 96, 50–57.
527 <https://doi.org/10.1016/j.microc.2010.01.018>
- 528 Pérez-Cid, B., Fernandez Albores, A., Fernandez Gomez, E., Falque Lopez, E., 2001. Use of
529 microwave single extractions for metal fractionation in sewage sludge samples. *Anal. Chim.*
530 *Acta* 431, 209–218. [https://doi.org/10.1016/S0003-2670\(00\)01335-0](https://doi.org/10.1016/S0003-2670(00)01335-0)
- 531 Pérez-Cid, B., Lavilla, I., Bendicho, C., 1999. Application of microwave extraction for
532 partitioning of heavy metals in sewage sludge. *Anal. Chim. Acta* 378, 201–210.
533 [https://doi.org/10.1016/S0003-2670\(98\)00634-5](https://doi.org/10.1016/S0003-2670(98)00634-5)
- 534 Pérez-Cid, B., Lavilla, I., Bendicho, C., 1998. Speeding up of a three-stage sequential extraction
535 method for metal speciation using focused ultrasound. *Anal. Chim. Acta* 360, 35–41.
536 [https://doi.org/10.1016/S0003-2670\(97\)00718-6](https://doi.org/10.1016/S0003-2670(97)00718-6)

- 537 Real, C., Barreiro, R., Carballeira, A., 1994. The application of microwave heating in sequential
538 extractions of heavy metals in estuarine sediments. *Sci. Total Environ.* 152, 135–142.
539 [https://doi.org/10.1016/0048-9697\(94\)90493-6](https://doi.org/10.1016/0048-9697(94)90493-6)
- 540 Reid, M.K., Spencer, K.L., Shotbolt, L., 2011. An appraisal of microwave-assisted Tessier and
541 BCR sequential extraction methods for the analysis of metals in sediments and soils. *J. Soils
542 Sediments* 11, 518–528. <https://doi.org/10.1007/s11368-011-0340-9>
- 543 Relić, D., Đorđević, D., Sakan, S., Anđelković, I., Miletić, S., Đuričić, J., 2011. Aqua regia
544 extracted metals in sediments from the industrial area and surroundings of Pančevo, Serbia.
545 *J. Hazard. Mater.* 186, 1893–1901. <https://doi.org/10.1016/j.jhazmat.2010.12.086>
- 546 Relić, D., Đorđević, D., Sakan, S., Anđelković, I., Pantelić, A., Stanković, R., Popović, A.,
547 2013a. Conventional, microwave, and ultrasound sequential extractions for the fractionation
548 of metals in sediments within the Petrochemical Industry, Serbia. *Environ. Monit. Assess.*
549 185, 7627–7645.
- 550 Relić, D., Đorđević, D., Sakan, S., Anđelković, I., Pantelić, A., Stanković, R., Radojičić, A.,
551 Popović, A., 2013b. An appraisal of conventional , microwave and ultrasound BCR
552 extraction methods for the analysis of metals in sediments of Pančevo , Serbia, in: *E3S Web
553 of Conferences*. pp. 1–4. <https://doi.org/10.1051/e3sconf/20130139002>
- 554 Remeteiová, D., Ružičková, S., Rusnák, R., 2008. Ultrasound-assisted extraction in the
555 fractionation analysis of gravitation dust sediments. *Microchim. Acta* 163, 257–261.
556 <https://doi.org/10.1007/s00604-008-0005-6>
- 557 Rocha, M.J., Rocha, E., Cruzeiro, C., Ferreira, P.C., Reis, P. a, 2011. Determination of
558 polycyclic aromatic hydrocarbons in coastal sediments from the Porto region (Portugal) by
559 microwave-assisted extraction, followed by SPME and GC-MS. *J. Chromatogr. Sci.* 49,
560 695–701. <https://doi.org/10.1093/chrs/49.9.695>
- 561 Sipos, L., Kovács, Z., Szöllosi, D., Kókai, Z., Dalmadi, I., Fekete, A., 2011. Comparison of
562 novel sensory panel performance evaluation techniques with e-nose analysis integration. *J.
563 Chemom.* 25, 275–286. <https://doi.org/10.1002/cem.1391>

- 564 Sipos, P., Choi, C., Nemeth, T., Szalai, Z., Poka, T., 2014. Relationship between iron and trace
565 metal fractionation in soils. *Chem. Speciat. Bioavailab.* 26, 21–30.
566 <https://doi.org/10.3184/095422914X13887685052506>
- 567 Sutherland, R.A., 2010. BCR®-701: A review of 10-years of sequential extraction analyses.
568 *Anal. Chim. Acta* 680, 10–20. <https://doi.org/10.1016/j.aca.2010.09.016>
- 569 Tangni, E.K., Motte, J.C., Callebaut, A., Chandelier, A., De Schrijver, M., Pussemier, L., 2011.
570 Deoxynivalenol loads in matched pair wheat samples in Belgium: Comparison of ELISA
571 VERATOX kit against liquid chromatography. *Mycotoxin Res.* 27, 105–113.
572 <https://doi.org/10.1007/s12550-010-0081-4>
- 573 Tokalioğlu, Ş., Yilmaz, V., Kartal, Ş., 2010. An assessment on metal sources by multivariate
574 analysis and speciation of metals in soil samples using the BCR sequential extraction
575 procedure. *Clean - Soil, Air, Water* 38, 713–718. <https://doi.org/10.1002/clen.201000025>
- 576 Tumuklu, A., Yalcin, M.G., Sonmez, M., 2007. Detection of heavy metal concentrations in soil
577 caused by Nigde city garbage dump. *Polish J. Environ. Stud.* 16, 651–658.
- 578 Vajna, B., Farkas, A., Pataki, H., Zsigmond, Z., Igricz, T., Marosi, G., 2012. Testing the
579 performance of pure spectrum resolution from Raman hyperspectral images of differently
580 manufactured pharmaceutical tablets. *Anal. Chim. Acta* 712, 45–55.
581 <https://doi.org/10.1016/j.aca.2011.10.065>
- 582 Vajna, B., Patyi, G., Nagy, Z., Bodis, A., Farkas, A., Marosi, G., 2011. Comparison of
583 chemometric methods in the analysis of pharmaceuticals with hyperspectral Raman
584 imaging. *J. Raman Spectrosc.* 42, 1977–1986. <https://doi.org/10.1002/jrs.2943>
- 585 Vračko, M., Minovski, N., Heberger, K., 2010. Ranking of QSAR models to predict minimal
586 inhibitory concentrations toward *Mycobacterium tuberculosis* for a set of fluoroquinolones.
587 *Acta Chim. Slov.* 57, 586–590. <https://doi.org/Vracko-2010-3> [pii]
- 588 Wood, J.E., Allaway, D., Boulton, E., Scott, I.M., 2010. Operationally realistic validation for
589 prediction of cocoa sensory qualities by high-throughput mass spectrometry. *Anal. Chem.*

590 82, 6048–6055. <https://doi.org/10.1021/ac1006393>

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594 **Figure captions**

595 Figure 1. Ordering using sum of ranking differences (SRD) for 12 extraction techniques by
596 consensus (row average was used as reference ranking) by 19 elements. The SRD
597 values (x -axis and left y -axis) were scaled to range from 0 to 100. The theoretical
598 distribution function was approximated by fitting a Gaussian bell curve (mean = 66.95,
599 standard deviation = 10.15). The right y -axis contains the relative frequencies for the
600 theoretical distribution function.

601 Figure 2. Ordering of 19 extracted metals by consensus (row average was used as reference
602 ordering) by 12 extraction techniques. The SRD values (x -axis and left y -axis) were
603 scaled to range from 0 to 100. The right y -axis contains the relative frequencies for the
604 theoretical distribution function. XX1 is the first icosaille (5 % limit), Med means
605 median and XX19 is the last icosaille (95 %).

606 Figure 3. Ordering using the sum of ranking differences for 12 extraction techniques by
607 consensus (row average was used as reference ordering) by 41 samples. The SRD
608 values (x -axis and left y -axis) were scaled to range from 0 to 100. The theoretical
609 distribution function was approximated by fitting a Gaussian bell curve (mean = 66.77,
610 standard deviation = 6.7). The right y -axis contains the relative frequencies for the
611 theoretical distribution function.

612

613 **Figure captions supplementary material**

614 Figure S1. Box and whisker plot of the cross-validated (sevenfold) SRD values for the extraction
615 techniques.

616 Figure S2. Tree diagram of the original data matrix (Euclidian distance and Ward's method were
617 used).

618 Figure S3. Principal component analysis of the original data matrix: score 1 vs. score 2 plot (data
619 pre-treatment: standardization, explained variance can be found in brackets).

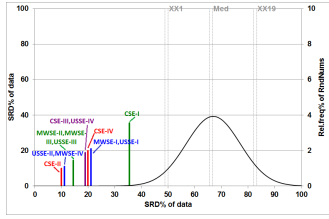
620 Figure S4. Box and whisker plot of the cross-validated (sevenfold) SRD values for extracted
621 metals.

622 Figure S5. Cluster analysis of cross-validated SRD values for metals (linkage rule: Ward's
623 method; distance measure: Euclidian one).

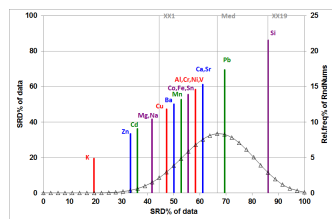
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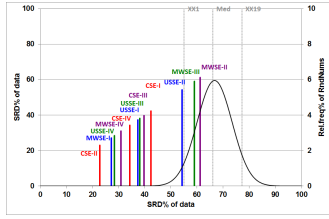
Table 1. Recoveries (%) of reference material BCR-701 after all four SE steps (mean \pm standard deviation, n=2, mg kg⁻¹, dry weight).

Metal	Technique	Sum all four extracted concentrations (3 steps +residual)	Direct aqua regia extraction (Sutherland, 2010)	Recovery (%)
Cd	CSE	11.7 \pm 0.4	11.7 \pm 0.6	100
	MWSE	10.6 \pm 0.5		90.6
	USSE	10.8 \pm 0.2		92.3
Cr	CSE	111.2 \pm 3.5	272 \pm 12	41
	MWSE	114.4 \pm 1.7		42
	USSE	128.2 \pm 3.3		47
Cu	CSE	234.4 \pm 3.6	275 \pm 8	85.2
	MWSE	232.5 \pm 7.1		84.4
	USSE	232.1 \pm 11.2		84.0
Ni	CSE	76.24 \pm 2.30	103 \pm 3	74.0
	MWSE	75.43 \pm 1.52		73.2
	USSE	65.72 \pm 2.28		63.8
Pb	CSE	153.8 \pm 2.0	143 \pm 3	107.5
	MWSE	139.9 \pm 8.2		97.8
	USSE	142.7 \pm 13.8		99.8
Zn	CSE	424.4 \pm 14.4	454 \pm 12	93.5
	MWSE	399.5 \pm 6.5		88.0
	USSE	365.8 \pm 9.3		80.6

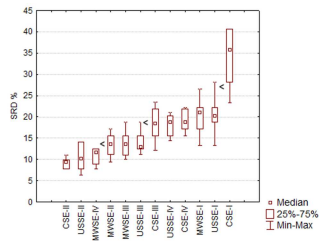


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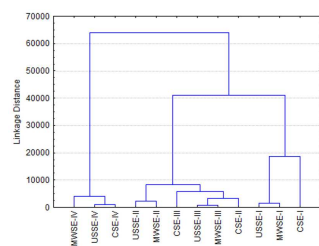




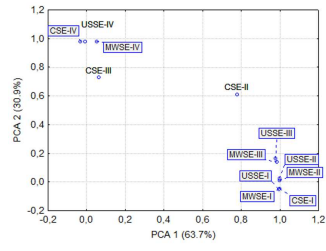
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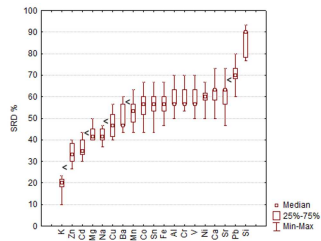
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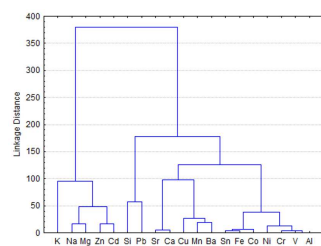
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Highlights

- Sum of ranking difference were applied for metals, samples, extractions, techniques.
- Similarity and dissimilarity of methods for metal extraction was revealed.
- Similarity and dissimilarity of metals regarding extraction efficiency was disclosed.
- Similarity and dissimilarity of methods regarding samples were achieved.
- Cluster and principal component analysis reveals similar grouping as applied method.