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1 Interspecific and geographical variations of trace
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24 **ABSTRACT :** Cd, Cu and Zn were analysed in the organs and tissues of the three
25 scallop species from the Bay of Biscay, the variegated *Chlamys varia* L., the queen
26 scallop *Aequipecten opercularis* L. and the common scallop *Pecten maximus* L. for
27 interspecific comparisons. In *P. maximus* the greatest concentrations of Cd and Cu were
28 found in the digestive gland, whereas the two other species showed similar levels of Cd
29 between digestive gland and kidneys and higher renal Cu concentrations. However, the
30 digestive gland of all Pectinidae species contained from 75 to 93 % and 52 to 74 % of the
31 total body burdens of Cd and Cu, respectively. Whatever the species and the origin,
32 kidneys displayed the highest Zn concentrations, which therefore contained from 53 to 97
33 % of the total body burden of this metal. Also using reported results, ratios between the
34 concentrations in the digestive gland and that in the kidneys discriminated two groups of
35 Pectinidae : 1) the *Pecten* group (*P. maximus*, *P. jacobus* and *Adamussium colbecki*)
36 with a Cd ratio ≥ 4 , a Cu ratio ≥ 1 and a Zn ratio $> 20 \cdot 10^{-3}$ 2) the *Chlamys* group with a
37 Cd ratio ≤ 1 , a Cu ratio ≤ 1 and a Zn ratio $\leq 6 \cdot 10^{-3}$. However, no differences in the
38 detoxification processes in the digestive gland were found between groups in this study.
39 72 to 80 % of the total Cd was found to be soluble, probably bound to metalloproteins
40 such as metallothioneins, while Cu and Zn were mainly found associated to the insoluble
41 fraction of the digestive gland cells whatever the species. Queen scallops from the Faroe
42 Islands were also considered to examine variations of the metal concentrations due to the
43 geographical origin. In this northern area, queen scallops displayed Cd concentrations 2,
44 4 and 6 times higher in kidneys, muscle and digestive gland, respectively, compared to
45 those from the Bay of Biscay. Consequently, the Cd concentrations in the whole soft
46 parts of the queen scallops were more than 6 times higher in the Faroe Islands than in the

47 Bay of Biscay. In contrast to Cd, both Cu and Zn concentrations in the whole soft parts
48 were 2 times lower in the Faroe Islands, suggesting lower bioavailability of these
49 essential elements.

50

51 Key words : Heavy metals ; Bioaccumulation ; Body distribution ; Subcellular
52 distribution ; Scallop

53

54

INTRODUCTION

55

56 Among seashells, numerous scallop species represent an important target to commercial
57 fisheries. Many of them are actually cultured for direct commercial production or to
58 provide juveniles for reintroduction programs in areas over-exploited. On the French
59 Atlantic coasts and in the English Channel, scallops are relatively common, largely
60 distributed and belong to three species: the variegated scallop *Chlamys varia* on rocky
61 substrata, the queen scallop *Aequipecten opercularis* and the common scallop *Pecten*
62 *maximus* on sandy bottoms. In France, these three species are captured during winter time
63 and mobilises around one thousand fishing boats with various duration. The total of the
64 catch varies greatly from one year to another, ranging from 6 000 tons to 25 000 tons per
65 year (Quéro and Vayne, 1998). The three species are also captured for leisure activities,
66 directly on the shore or by snorkelling. Scallops represent therefore an elevated economic
67 values in fishing areas.

68

69 Bivalves are classically chosen for studies on trace element bioaccumulation (Bryan
70 1976). They are poorly capable to regulate ion concentrations in internal fluids and they
71 are highly tolerant, metal ions being sequestered in excess of metabolic requirement
72 (George, 1980). Therefore, bivalves often display high accumulation levels without
73 apparent toxic effect. Among bivalves, scallops have shown a particular ability to
74 concentrate high levels of pollutants such as trace elements, even in areas remote from
75 direct anthropogenic contamination like the Antarctic (Mauri et al., 1990, Berkman and
76 Nigro, 1992, Viarengo et al., 1993) or the Arctic Oceans (Stepnowski and Skwarzec,
77 2000). Therefore, scallops are generally considered as valuable bioindicators to monitor
78 marine pollution (Bryan, 1973, Bustamante et al., 2002a). However, large interspecific
79 variations seem to occur when compared trace element concentrations between different
80 scallop species (Mauri et al., 1990, Viarengo et al., 1993). Such variations are not well
81 characterized since only a single species is usually studied at the same place.
82 Furthermore, geographical variations for a single scallop species have been rarely
83 considered, different species from various latitudes being generally compared.

84

85 In this study, Cd, Cu and Zn, were analyzed in the organs and tissues of the three
86 common scallop species living along the Atlantic coast of France. Those metals were
87 determined by flame and graphite atomic absorption spectrophotometry in the variegated
88 scallop *C. varia*, in the queen scallop *A. opercularis* and in the common scallop *P.*
89 *maximus*, from the Charente-Maritime coasts, Bay of Biscay, France, in an attempt to
90 determine the interspecific variations in the metal bioaccumulation. Furthermore,
91 specimens of *A. opercularis*, a widely distributed species in the Northeastern Atlantic

92 from Morocco to the north of Norway, were collected off the Faroe Islands for
93 comparison. Elemental concentrations, tissular and subcellular distributions, and
94 interspecific and geographical differences were therefore examined.

95

96

MATERIAL AND METHODS

97

Sampling and sample preparation

98 In February 1996, 41 variegated scallops (*Chlamys varia*, Linné, 1758) were collected by
99 hand in the infralittoral zone in the coast of Charente-Maritime (Bay of Biscay, France).
100 After 48 hours depuration in clean sea water to eliminate faecal and pseudo-faecal
101 material, scallops were pooled by size classes.
102

103 The queen and common scallops (*Aequipecten opercularis*, Linné, 1758 and *Pecten*
104 *maximus*, Linné, 1758, respectively) have been opportunistically collected by bottom
105 trawl along the Charente-Maritime coasts between February and May 1997. Individuals
106 (n=11 for each species) were separated in individual plastic bags and frozen on board.

107 Queen scallops *A. opercularis* from the Faroe Islands have also been collected
108 opportunistically by bottom trawl in August 1996 and frozen on board as described above
109 (n=31).

110 For each scallop, adductor muscle, gonad, gills, kidneys, and digestive gland were
111 separated from the rest of the soft part. The remaining tissues including the mantle, the
112 foot, the intestine and the heart, were also analysed in order to calculate the whole trace
113 element contained of the soft parts.

114

115 *Subcellular fractionation*

116 The partitioning of trace elements between soluble and insoluble subcellular fractions of
117 the digestive gland was investigated in the three scallop species. Thus, 40 *Chlamys varia*,
118 23 *Pecten maximus*, and 10 *Aequipecten opercularis* were devoted to subcellular
119 investigations. Four pools were made with the digestive glands removed from the soft
120 parts of *C. varia* and *P. Maximus* freshly dissected. In the case of *A. opercularis*,
121 digestive glands were removed on board to be deep frozen in liquid nitrogen and
122 consequently were treated individually.

123 Both pooled and individual digestive glands were subsequently homogenised in a Dounce
124 potter on ice with 10 volumes of a 0.02 M TRIS-HCl buffer in 0.25 M sucrose with 1
125 mM PMSF (phenylmethylsulfonylfluoride, as protease inhibitor) and 5 mM DTT
126 (dithiothreitol, as reducing agent), at pH 8.6 (Bustamante et al. 2002b). The homogenates
127 were centrifuged at 100 000 G for 1 h at 4°C in a Beckman LE-70 ultracentrifuge.
128 Particle-free supernatants (cytosols) were separated from the pellet. Aliquots of the
129 homogenates, cytosols and pellets were analysed for trace elements.

130

131 *Trace element analyses*

132 Tissue samples were dried for several days at 80°C to constant weight. Whenever
133 possible, two aliquots of approx. 300 mg of each homogenised dry sample were digested
134 with 4 ml of 65% ultrapure HNO₃ and 0.3 ml of ultrapure 70 % HClO₄ at 80°C until the
135 solution was clear. Then acids were evaporated and residues were dissolved in 0.3N
136 ultrapure nitric acid.

137 Cadmium, copper and zinc were determined by flame and graphite furnace atomic
138 absorption spectrophotometry with a Varian spectrophotometer Vectra 250 Plus, with
139 Deuterium background correction. Certified reference materials, dogfish liver DOLT-2
140 (NRCC) and dogfish muscle DORM-2 (NRCC), were treated and analysed in the same
141 way as the samples. The results for standard reference materials were in good agreement
142 with certified values (Table 1). The detection limits calculated for 0.3 g of dry tissue were
143 ($\mu\text{g}\cdot\text{g}^{-1}$ dwt): 0.005 (Cd), 0.5 (Cu), and 3 (Zn). Metal concentrations in tissues are given
144 relatively to the dry weight ($\mu\text{g}\cdot\text{g}^{-1}$ dry wt) while the distribution percentages were
145 calculated for wet weight.

146

147 *Data analyses*

148 Comparison of metal concentrations between species were tested by one-way ANOVA
149 (after log transformation of data when necessary) followed by the Tukey's multiple
150 comparison test in the MINITAB 13.1 software. Changes in metal distribution among
151 scallop tissues were tested for significance by the G procedure (adapted from the log-
152 likelihood ratio test) for 2 x k contingency tables (Zar, 1996). The significance level for
153 statistical analyses was always set at $\alpha = 0.05$.

154

155

RESULTS

156

157 Concentrations of Cd, Cu and Zn in the tissues of 3 scallop species from the Bay of
158 Biscay and 1 species from the Faroe Islands are presented in Table 2. Concentrations in
159 the whole soft parts were calculated using the concentrations determined in each

160 compartments. The body distribution of the trace elements of the scallops is presented in
161 Table 3.

162

163 *Cadmium*

164 Among tissues, the digestive gland and the kidneys displayed the highest Cd
165 concentrations for all species and in both areas. Interestingly, the Cd values measured in
166 the different species were in the same range of concentrations in both organs with the
167 exception of *P. maximus*. In this species, Cd concentrations in this species were around 4
168 times higher in the digestive gland than in the kidneys, i.e. $264 \pm 33 \mu\text{g}\cdot\text{g}^{-1}$ dwt vs $70.6 \pm$
169 $12.6 \mu\text{g}\cdot\text{g}^{-1}$ dwt, respectively (Table 2). The digestive gland contained most of the total
170 body burden of Cd in all species, probably because of the significance in weight of this
171 organ (Table 3). For Cd, a G-test did not indicate any significant ($p>0.05$) difference
172 among metal body distribution between species and areas.

173 Among the scallops from the Bay of Biscay, *P. maximus* showed a strong accumulation
174 of Cd in its tissues, especially in kidneys, muscle and digestive gland compared to the
175 other species. Thus, these tissues displayed Cd concentrations 2, 4, and 6 times higher,
176 respectively, than those measured in *C. varia* and in *A. opercularis* (Table 2).
177 Consequently, the whole soft parts of *P. maximus* contained Cd concentrations more than
178 4 times higher than in the other two scallop species (Table 2).

179 When compared Faroe Islands and Bay of Biscay, Cd concentrations in *A. opercularis*
180 were in the same range in the gonad, gills, and muscles for both geographic areas ($F=2.31$
181 $p=0.140$). Nevertheless, *A. opercularis* from the Faroe Islands displayed more than 4
182 times Cd concentrations in the digestive gland and 7 times higher in the kidneys than in

183 queen scallops from the Bay of Biscay (Table 2). Consequently, the digestive gland of *A.*
184 *opercularis* from the Faroe Island contained even higher percentage to the total body
185 burdens of Cd, i.e. 90% vs 75% (Table 3). Whichever the origin was, in the digestive
186 gland of the three species more than 70 % of the total Cd was found in the particle free
187 supernatants (Table 4).

188

189 *Copper*

190 Similarly to Cd, the digestive gland and the kidneys displayed the highest Cu
191 concentrations for all species and in both areas. However, this metal displayed large
192 variations according to the species and to the origin of the scallop.

193 In the scallops from the Bay of Biscay, Cu clearly showed the highest concentrations in
194 kidneys except for *P. maximus*, whose concentrations were 3 times higher in the digestive
195 gland (Table 2). Specifically in this species, the gonad exhibited relatively elevated Cu
196 concentrations ($19.7 \pm 16.1 \mu\text{g.g}^{-1}$ dwt), which were in the same order of magnitude than
197 in kidneys ($13.5 \pm 2.0 \mu\text{g.g}^{-1}$ dwt). Despite such variations in the Cu concentrations, the
198 digestive gland of the different scallop species contained most of the whole body burden
199 of Cu (Table 3). Therefore, tissue distribution of Cu did not varied significantly (G-test,
200 $p > 0.05$) between all the different scallop species.

201 Considering the whole soft parts, scallops from the Faroe Islands showed Cu
202 concentrations 2 times lower than those from the Bay of Biscay (Table 2). Globally, the
203 concentrations of Cu in the gonad, gills, muscle and remaining tissues were slightly lower
204 in scallop from the Faroe Islands. This difference was even more important for the
205 digestive gland and kidneys (Table 2). However, the digestive gland of the scallops from

206 the Faroe Islands also contained most of the whole body burden of Cu, i.e. $61 \pm 4\%$
207 (Table 3).

208 Whichever the origin was, in the digestive gland most of the Cu was found associated to
209 the pellet of centrifugation, only 25 to 43% of the total Cu being soluble (Table 4).

210

211 *Zinc*

212 In scallops, kidneys displayed the highest Zn concentrations for all species and in both
213 areas (Table 2). These concentrations were several thousand of μg (i.e. exceeding 30,000
214 $\mu\text{g Zn.g}^{-1}$ in *A. opercularis* from the Bay of Biscay).

215 In the Bay of Biscay, the concentrations of Zn were 7 times higher in *A. opercularis* than
216 in *P. maximus* but not significantly different from *C. varia*. In contrast, *P. maximus*
217 exhibited Zn concentrations in the digestive gland which were 4 times higher than in *C.*
218 *varia* and *A. opercularis*. Despite such differences in the tissue concentrations, kidneys of
219 the different scallop species contained from 72 to 91 % of the whole body burden of Zn
220 (Table 3). Moreover, a G-test did not indicate any significant ($p > 0.05$) difference among
221 metal body distribution between these species.

222 In the Faroe Islands, *A. opercularis* exhibited lower Zn concentrations in all their tissues
223 than in the Bay of Biscay (Table 2). This was particularly obvious for the kidneys which
224 concentrations were 7 times lower compared to *A. opercularis* from the Bay of Biscay
225 while such a difference was only a 2 or 3 fold for the other tissues. Consequently, *A.*
226 *opercularis* from the Faroe Islands only exhibited 53 % of the whole body burden of Zn
227 in their kidneys (Table 3).

228 Despite interspecific and geographical differences in the concentrations, the proportion of
229 soluble Zn within the digestive gland cells poorly varied, i.e. from 38 to 50% in the
230 different scallop species (Table 4).

231

232

DISCUSSION

233

Interspecific comparisons

235 Among bivalves, scallops accumulate trace elements at often high concentrations,
236 depending on the metal and the tissue. The kidney and the digestive gland have a major
237 importance in the bioaccumulation of Cd, Cu and Zn in Pectinidae. These organs
238 exhibited high metal concentrations and contained most of the total body burden of these
239 trace elements (Tables 1 & 2). The other tissues seems to have a minor importance in the
240 storage of Cd, Cu and Zn although they might have a major role in uptake and transfer of
241 trace elements. However, one of the goals of the present study was to provide background
242 levels against which to perform interspecific comparison within the Pectinidae family.
243 Results from this study allowed to discriminate two groups of scallops : on the one hand
244 *Chlamys varia* and *Aequipecten opercularis* (formerly called *Chlamys opercularis*) and
245 on the other side *Pecten maximus*. These two groups are revealed by the ratio between the
246 concentrations of trace elements in the digestive gland and that in kidneys : 1) the *Pecten*
247 group (*P. maximus*) with a Cd ratio ≥ 4 , a Cu ratio ≥ 1 and a Zn ratio $> 20 \cdot 10^{-3}$ and, 2) the
248 *Chlamys* group (*C. varia* and *A. opercularis*) with a Cd ratio ≤ 1 , a Cu ratio ≤ 1 and a Zn
249 ratio $\leq 6 \cdot 10^{-3}$. Even though metal concentrations have been studied in various scallop
250 species, little data from the current literature allowed to make such calculations, mainly

251 because of the lack of values in kidneys. Table 5 reports Cd, Cu and Zn concentrations in
252 the digestive gland and kidneys of different scallops species from various locations. Even
253 though the variation of the concentrations of Cd, Cu and Zn in both tissues is very large,
254 the ratio of the concentrations between the digestive gland and the kidneys were
255 consistent with our results. Thus, *P. maximus* from the English Channel and *P. jacobeus*
256 from the Mediterranean showed ratios around 4 for Cd, $> 20 \cdot 10^{-3}$ for Zn, and respectively
257 3 and 1 for Cu. These calculated ratio were even higher for the Antarctic scallop
258 *Adamussium colbecki* (i.e. 12 for Cd, 3 for Cu and $377 \cdot 10^{-3}$ for Zn) suggesting a
259 metabolism of metals close to the one of the *Pecten* group. In contrast, taking into
260 account the moisture of the tissues, a ratio around 1 for Cd suggests that *Placopecten*
261 *magellanicus* is closer to the *Chlamys* group.

262 The bioaccumulation of trace elements in the digestive gland and kidneys might be
263 related to the detoxification processes of metals. The reasons of high concentrations of
264 trace elements in the tissues of Pectinidae are typically attributed to the binding to
265 cytosolic proteins in the digestive gland and to precipitation on granules in kidneys
266 (George et al., 1980, Ballan-Dufrançais et al., 1985, Stone et al., 1986, Fowler and Gould,
267 1988, Mauri et al., 1990, Viarengo et al., 1993). Globally, information on the subcellular
268 distribution of metals in Pectinidae is scarce. The cytosolic proteins of the digestive gland
269 cells of the three scallop species seem to have a minor importance in the binding of Cu
270 and Zn. These two metals were generally found in large proportions into the insoluble
271 fraction, which represent the metals trapped within the organites, associated to
272 membranes or to granules. This might be due to their numerous biochemical functions
273 that imply they are involved in the structure of several proteins. In contrast to Cu and Zn,

274 from 72 to 80 % of the total Cd was found in the cytosolic fraction of the digestive gland
275 cells of the three scallop species. With the exception of the Antarctic scallop *Adamussium*
276 *colbecki*, for which only 30 % of the total Cd was soluble (Viarengo et al., 1993), 60 to
277 99 % of the Cd was found in the soluble fraction of the digestive gland of Pectinidae
278 from field or after experimental exposure to this metal (Evtushenko et al., 1986, 1990,
279 Stone et al., 1986, Lukyanova and Evtushenko, 1989). This strongly suggests the
280 occurrence of mechanisms of accumulation and detoxification of Cd involving proteins
281 such as metallothioneins (Evtushenko et al., 1986, 1990, Stone et al., 1986, Viarengo et
282 al., 1993, Bustamante, 1998). Similar detoxification strategies for Cd, Cu and Zn in the
283 digestive gland of the *Chlamys* and the *Pecten* groups suggest that differences between
284 the two groups are related to detoxification and excretion processes in the kidneys.
285 Indeed, metal-rich granules from the kidney of *P. maximus* are considered as excretory
286 concretions which can be eliminated in urine (George et al., 1980). Higher excretion rates
287 in the *Pecten* group compared to the *Chlamys* one might lead to overall lower Cd, Cu and
288 Zn concentrations in this organ. However, further studies in the *Chlamys* group are
289 needed to support this hypothesis.

290

291 *Geographical comparisons*

292 Scallops of different species within the same area and scallops of the same species from
293 different latitudes showed elevated variations in their ability to concentrate trace elements
294 (Table 5). Such variations are due to interspecific diversity but also to geographic
295 conditions. Accumulation of high metal concentrations, particularly Cd, even in
296 unpolluted areas might be related to different bioavailability of the elements, to different

297 feeding habits, as well as to physiological features. As regards to the concentrations
298 found in the gills, seawater appears to be an important source of metals for scallops.
299 However, trace element concentrations in the soft tissues and primarily in the digestive
300 gland might substantially be influenced by the metal content of their food. Indeed,
301 scallops are filter feeders using different types of food such as phytoplankton, detritus
302 and resuspended matter which are major components of the variation in food quality in
303 the field through variation in their carbon and nitrogen contents (Grant and Cranford,
304 1991, Alber and Valiela, 1996). It would therefore be of particular interest to characterise
305 the food composition and its metal concentrations of the different scallops.

306 As reported for other marine invertebrates (Cubadda et al., 2001), the use of scallops as
307 biomonitors on large geographical scales, i.e. between 44 to 46° N for the Bay of Biscay
308 and 61 to 63 ° N for the Faroe Islands, might be envisaged with caution. In this
309 investigation, the higher Cd concentrations found in *A. opercularis* from the Faroe
310 Islands compared to those from the Bay of Biscay highlighted that the contents of one or
311 more metals in marine organisms can be increased by specific geochemical or
312 hydrodynamic conditions. In the Faroe Islands, enrichment of metals due to industrial
313 contamination does not exist. Consequently, the high Cd concentrations in the tissues of
314 *A. opercularis* might be considered as natural in this sub-polar area. Reported Cd
315 enrichment throughout the Arctic remained globally unclear. Several authors have
316 proposed that a phenomenon of Cd abnormality occurs in polar and subpolar areas,
317 leading to very high Cd concentrations in marine biota (Petri and Zauke, 1993, Bargagli
318 et al., 1996, Bustamante et al., 1998a, Zauke et al., 1999). Several results suggest that
319 such an abnormality has also to be considered in the Faroe Islands (Caurant et al., 1994,

320 Bustamante et al., 1998b, 2004). Indeed, similarly to polar regions, soluble trace element
321 concentrations in seawater appear to be relatively low around this area (Mart and
322 Nürnberg, 1984). Despite that, very high Cd concentrations have also been found in
323 several cephalopod species caught around the archipelago, i.e. from $2.1 \mu\text{g}\cdot\text{g}^{-1}$ dwt in the
324 Loliginid squid *Loligo forbesi*, to $46.8 \mu\text{g}\cdot\text{g}^{-1}$ dwt in the octopus *Eledone cirrhosa*
325 (Bustamante, 1998, Bustamante et al., 1998b). Moreover, coastal invertebrates such as
326 dog-whelks and limpets also displayed elevated Cd concentrations, i.e. $140 \mu\text{g}\cdot\text{g}^{-1}$ dwt
327 and $50 \mu\text{g}\cdot\text{g}^{-1}$ dwt, respectively (Dam, 2000). Therefore, the high Cd concentrations in the
328 tissues of *A. opercularis* might reflect a global enrichment by this metal in the Faroe
329 Islands biota. Nevertheless, the reasons of such an enrichment remain to be determined.

330 In contrast, Cu and Zn appeared to be concentrated in much lower extent in the scallops
331 from the Faroe Islands compared to those from the Bay of Biscay and from the English
332 Channel (Bryan, 1973). Lower concentrations of these essential elements could be due to
333 low bioavailability of these two metal in the Faroe Island waters. High Cd concentrations
334 associated with low Cu and Zn values are typically attributed to the substitution of Cd
335 ions for essential metals under deficient conditions (Zauke and Petri, 1993, Bustamante et
336 al., 1998a). This possibly occurred in *A. opercularis* from the Faroe Islands. Indeed, the
337 digestive gland showed a high Cd / Cu ratio (i.e. 8.8) compared to the Bay of Biscay (i.e.
338 0.6). In the Faroe scallops, this high ratio was due to both elevated Cd concentrations and
339 to low Cu concentrations. Cu concentrations in the digestive gland were lower than
340 concentrations found in the temperate scallop species but in the same range than the
341 concentrations reported for the Antarctic scallop *A. colbecki* (Table 5). Large amounts of
342 Cu are essential to marine mollusks, for which Cd and Cu are known to interfere on the

343 same type of metalloproteins (i.e. metallothioneins) in the digestive gland. A competition
344 or a substitution between the two elements may have occurred in the Faroe scallops.

345

346

CONCLUSION

347

348 The three different species examined in this study accumulated high trace element
349 concentrations in their tissues, particularly in the digestive gland and in the kidney. The
350 accumulation of Cd was even higher for Cd in the subpolar Faroe area. In contrast to Cd,
351 lower Cu and Zn concentrations suggests a poor bioavailability of these essential
352 elements in that area. Regardless of the species and the origin, Cd appeared to be
353 associated to cytosolic proteins involved in detoxification functions. The presence of Cu
354 and Zn on organites and membranes might be due to their essentiality.

355

356

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363

364

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486 Table 1. Comparison of trace elements concentrations ($\mu\text{g}\cdot\text{g}^{-1}$ dry wt) of dogfish liver
487 DOLT-2 (NRCC), and dogfish muscle (NRCC) determined in the present study with
488 certified values.

489

Metals	DOLT-2 (n=5)		DORM-2 (n=5)	
	Certified values	Present study	Certified values	Present study
Cd	20.8 ± 0.5	19.7 ± 0.6	0.043 ± 0.008	0.042 ± 0.014
Cu	25.8 ± 1.1	24.9 ± 1.2	2.34 ± 0.16	2.30 ± 0.22
Zn	85.8 ± 2.5	84.5 ± 2.8	25.6 ± 2.3	26.3 ± 1.8

490

491

492 Table 2. Mean \pm SD of Cd, Cu, and Zn concentrations ($\mu\text{g}\cdot\text{g}^{-1}$ dwt) in the organs and tissues of three scallop species from the Bay of
 493 Biscay and the Faroe Islands. N : number of pools (sum of the individuals)

Element Species	Location	Digestive gland	Kidneys	Gonad	Gills	Muscle	Remaining tissues	Whole soft parts
Cd								
<i>Aequipecten</i>	Bay of Biscay	27.5 \pm 4.24	21.1 \pm 3.15	3.99 \pm 3.17	7.41 \pm 0.03	0.70 \pm 0.24	4.74 \pm 2.13	5.70 \pm 1.21
"	Faroe Islands	123 \pm 14	159 \pm 80	3.54 \pm 0.89	8.46 \pm 3.38	0.49 \pm 0.09	10.5 \pm 2.84	38.7 \pm 11.1
<i>Chlamys varia</i>	Bay of Biscay	40.0 \pm 4.43	38.8 \pm 11.1	2.11 \pm 0.27	4.12 \pm 0.39	0.54 \pm 0.08	2.06 \pm 0.32	7.69 \pm 0.77
<i>Pecten maximus</i>	"	264 \pm 33	70.6 \pm 12.6	5.28 \pm 5.44	4.68 \pm 2.26	2.01 \pm 1.07	3.58 \pm 0.60	29.7 \pm 12.0
Cu								
<i>Aequipecten</i>	Bay of Biscay	43.6 \pm 19.5	507 \pm 144	8.1 \pm 0.6	7.1 \pm 0.7	1.7 \pm 0.9	4.5 \pm 0.8	15.0 \pm 4.6
"	Faroe Islands	14.0 \pm 0.8	106 \pm 49	6.5 \pm 0.7	4.7 \pm 0.2	1.0 \pm 0.1	2.5 \pm 0.1	7.6 \pm 2.3
<i>Chlamys varia</i>	Bay of Biscay	88.1 \pm 13.8	142 \pm 20	29.6 \pm 6.0	8.1 \pm 0.8	1.8 \pm 0.2	5.5 \pm 0.4	19.2 \pm 1.7
<i>Pecten maximus</i>	"	40.7 \pm 7.8	13.5 \pm 2.0	19.7 \pm 16.1	5.3 \pm 2.4	1.2 \pm 0.3	3.4 \pm 0.3	6.5 \pm 1.6
Zn								
<i>Aequipecten</i>	Bay of Biscay	198 \pm 47	31053 \pm 8147	195 \pm 16	384 \pm 74	79 \pm 17	200 \pm 35	437 \pm 159
"	Faroe Islands	77 \pm 14	6632 \pm 3664	120 \pm 15	96 \pm 7	39 \pm 4	105 \pm 32	193 \pm 106
<i>Chlamys varia</i>	Bay of Biscay	149 \pm 27	24107 \pm 3465	239 \pm 45	232 \pm 99	101 \pm 13	181 \pm 61	781 \pm 120
<i>Pecten maximus</i>	"	804 \pm 92	7278 \pm 3041	254 \pm 105	143 \pm 4	64 \pm 7	87 \pm 6	235 \pm 110

494

495 Table 3. Body distribution of Cd, Cu, and Zn (% of the wet weight) in the organs and tissues of three scallop species from the Bay of
 496 Biscay and the Faroe Islands. N : number of pools (sum of the individuals)

497

Species	Location	Digestive gland	Kidneys	Gonad	Gills	Muscle	Remaining tissues
Cd							
<i>Aequipecten</i>	Bay of Biscay	75 ± 8	4 ± 3	5 ± 2	3 ± 1	8 ± 3	12 ± 2
"	Faroe Islands	90 ± 3	5 ± 3	1 ± 0	1 ± 0	< 1	3 ± 1
<i>Chlamys varia</i>	Bay of Biscay	80 ± 3	8 ± 2	< 1	2 ± 0	4 ± 1	5 ± 1
<i>Pecten maximus</i>	"	93 ± 1	2 ± 1	< 1	< 1	3 ± 1	1 ± 0
Cu							
<i>Aequipecten</i>	Bay of Biscay	52 ± 9	37 ± 6	5 ± 3	1 ± 1	8 ± 5	3 ± 1
"	Faroe Islands	61 ± 4	18 ± 10	7 ± 3	4 ± 1	5 ± 2	5 ± 2
<i>Chlamys varia</i>	Bay of Biscay	74 ± 7	12 ± 3	2 ± 0	1 ± 0	5 ± 10	6 ± 1
<i>Pecten maximus</i>	"	70 ± 6	2 ± 2	11 ± 8	3 ± 3	11 ± 3	3 ± 1
Zn							
<i>Aequipecten</i>	Bay of Biscay	1 ± 1	97 ± 1	1 ± 0	1 ± 0	< 1	1 ± 0
"	Faroe Islands	17 ± 6	53 ± 18	6 ± 4	5 ± 2	9 ± 4	9 ± 5
<i>Chlamys varia</i>	Bay of Biscay	5 ± 1	72 ± 4	1 ± 0	2 ± 1	14 ± 3	7 ± 2
<i>Pecten maximus</i>	"	14 ± 11	81 ± 14	3 ± 1	1 ± 0	1 ± 1	1 ± 0

498

499 Table 4. Proportion of Cd, Cu, and Zn in the cytosol of the digestive gland cells (% of the
500 wet weight) of three scallop species from the Bay of Biscay and the Faroe Islands. N :
501 number of individuals

Species	Location	N	Cd	Cu	Zn
<i>Pecten maximus</i>	Bay of Biscay	23	72 ± 5	43 ± 4	38 ± 7
<i>Chlamys varia</i>	Bay of Biscay	40	78 ± 5	25 ± 8	41 ± 3
<i>Aequipecten opercularis</i>	Faroe Islands	10	80 ± 7	35 ± 16	50 ± 11

502

503 Table 5. Mean \pm SD of Cd, Cu, and Zn concentrations ($\mu\text{g}\cdot\text{g}^{-1}$ dwt) in different scallop
 504 species from various locations. N: sample size.

505

Species	Location	N	Cd	Cu	Zn	Reference
Digestive gland						
<i>Amusium balloti</i>	Australia	10	111 ^a	-	-	Francesconi et al. 1993
<i>Adamusium colbecki</i>	Antarctic	30	142 \pm 57	12.6 \pm 3.3	74.9 \pm 25.4	Mauri et al. (1990)
"	"	6	26.51 \pm 6.09 ^a	3.52 \pm 0.49 ^a	17.42 \pm 2.91 ^a	Viarengo et al. (1993)
<i>Aequipecten opercularis</i>	Northeast Atlantic	31	123 \pm 14	14.0 \pm 0.8	77 \pm 14	Present study
"	English Channel	11	27	36.7 \pm 11.4	132 \pm 26	Bryan (1973)
"	Bay of Biscay	11	27.5 \pm 4.24	43.6 \pm 19.5	198 \pm 47	Present study
<i>Chlamys varia</i>	"	41	40.0 \pm 4.43	88.1 \pm 13.8	149 \pm 27	"
<i>Pecten jacobeus</i>	Mediterranean Sea	30	41.0 \pm 18.2	16.6 \pm 6.6	124 \pm 55	Mauri et al. (1990)
"	"	6	7.19 \pm 1.23 ^a	3.13 \pm 0.55 ^a	28.63 \pm 3.29 ^a	Viarengo et al. (1993)
<i>P. maximus</i>	English Channel	11	321	57.9 \pm 27.4	407 \pm 123	Bryan (1973)
"	Bay of Biscay	11	264 \pm 33	40.7 \pm 7.8	804 \pm 92	Present study
<i>Placopecten magellanicus</i>	Northwest Atlantic	6	94.68 \pm 8.42 ^a	-	-	Uthe & Chou (1987)
Kidney						
<i>Adamusium colbecki</i>	Antarctic	30	11.6 \pm 3.3	4.0 \pm 1.7	199 \pm 89	Mauri et al. (1990)
<i>Aequipecten opercularis</i>	Northeast Atlantic	31	159 \pm 80	106 \pm 49	6632 \pm 3664	Present study
"	English Channel	11	41	1285 \pm 591	40800 \pm 7290	Bryan (1973)
"	Bay of Biscay	11	21.1 \pm 3.15	507 \pm 144	31053 \pm 8147	Present study
<i>Chlamys varia</i>	"	41	38.8 \pm 11.1	142 \pm 20	24107 \pm 3465	"
<i>Pecten jacobeus</i>	Mediterranean Sea	30	10.7 \pm 4.0	17.5 \pm 10.9	2790 \pm 1340	Mauri et al. (1990)
<i>P. maximus</i>	English Channel	11	79	20.8 \pm 10.3	19300 \pm 4800	Bryan (1973)
"	Bay of Biscay	11	70.6 \pm 12.7	13.5 \pm 2.0	7278 \pm 3041	Present study
<i>Placopecten magellanicus</i>	Northwest Atlantic	6	62.61 \pm 25.33 ^a	-	-	Uthe & Chou (1987)

506

507 ^a: values expressed relatively to wet weight.