

The sponge Oscarella lobularis (Porifera, Homoscleromorpha) as a suitable biomonitor of metallic contamination in Mediterranean coastal ecosystems

Kassandra de Pao Mendonca, Bernard Angeletti, Aurélie Dufour, Carole Borchiellini, Lars-Eric Heimbürger-Boavida, Emmanuelle Renard, Julien

Issartel

► To cite this version:

Kassandra de Pao Mendonca, Bernard Angeletti, Aurélie Dufour, Carole Borchiellini, Lars-Eric Heimbürger-Boavida, et al.. The sponge Oscarella lobularis (Porifera, Homoscleromorpha) as a suitable biomonitor of metallic contamination in Mediterranean coastal ecosystems. Marine Pollution Bulletin, 2023, 188, pp.114665. 10.1016/j.marpolbul.2023.114665. hal-03979892v2

HAL Id: hal-03979892 https://hal.science/hal-03979892v2

Submitted on 20 Sep 2023

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers. L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

1	
2	The sponge Oscarella lobularis (Porifera, Homoscleromorpha)
3	as a suitable biomonitor of metallic contamination in
4	Mediterranean coastal ecosystems
5	
6 7 8	Kassandra De Pao Mendonca ^{1,4} , Bernard Angeletti ^{3,5} , Aurélie Dufour ² , Carole Borchiellini ^{1°} , Lars-Eric Heimbürger-Boavida ^{2°} , Emmanuelle Renard ^{1,4,5°+} , Julien Issartel ^{1,5°+}
9	¹ Aix Marseille Univ, Avignon Univ, CNRS, IRD, IMBE, Marseille, France
10 11	² Aix Marseille Université, CNRS/INSU, Université de Toulon, IRD, Mediterranean Institute of Oceanography (MIO), Marseille, France
12	³ Aix Marseille Univ, CNRS, IRD, INRAE, Coll France, CEREGE, Aix-en-Provence
13	⁴ Aix Marseille Univ, CNRS, IBDM, Marseille, France
14	⁵ Aix Marseille Univ, CNRS, FR 3098 ECCOREV, F-13545 Aix-en-Provence, France
15	
16	° co-Pl
17	+ corresponding authors :
18 19	<u>emmanuelle.renard@imbe.fr</u> ; +33 6 76 94 79 38 ; Mediterranean Institute of marine and terrestrial Biodiversity and Ecology, 22 Chemin de la Batterie des Lions, 13007 Marseille, France
20 21	julien.issartel@imbe.fr ; +33 (0)4 13 94 93 51 ; Mediterranean Institute of marine and terrestrial Biodiversity and Ecology, Europôle de l'Arbois, Bâtiment Villemin, 13545, Aix-en-Provence, France

Abstract 22

23 The biomonitoring of metallic contamination in marine ecosystems is often focused on animal species of commercial interest and in lesser extent on non-model marine invertebrates. The 24 25 aim of the study was to compare the metal concentrations (Li, Al, Ti, Cr, Fe, Ni, Cu, Zn, As, Ag, 26 Cd, Hg, Pb) in seven marine sponges with a particular interest in the homoscleromorph sponge 27 Oscarella lobularis at different sites of the Bay of Marseille, France. Inter-species variabilities suggest that the seven sponge species studied accumulate metals differently. In O. lobularis, 28 a multi-site analysis shows different bioaccumulation between the eight sampled populations. 29 This inter-site differences may reflect differences in the hydrodynamic features and in past 30 and present industrial activities. Because Oscarella lobularis shows a homogeneous metal 31 accumulation pattern in comparison with the other tested species, it appears to be suitable 32 33 for metal contamination biomonitoring in Mediterranean coastal waters, in particular of the coralligenous communities. 34

35 Keywords: Sponge, bioconcentration factors, coralligenous, trace metals.

36 1.Introduction

37

The Mediterranean Sea is recognized as a major biodiversity hotspot, counting 7% of the 38 global marine species (Bianchi & Morri, 2000; European Environment Agency, 2006). During 39 the past century, this area has been exposed to intense anthropic pressure resulting from the 40 increasing population of the Mediterranean-bordering countries. Today, 450 million people 41 live in this area, without counting the important population increase occurring during summer 42 in a world leading tourist destination (Lanquar, 2011). As a consequence, agricultural, 43 domestic, commercial, and industrial land-based activities considerably intensified, increasing 44 45 contaminant inputs onto coastal marine environments via wastewaters, watersheds and 46 atmospheric inputs (Danovaro, 2003; Durrieu de Madron et al., 2011; Heimbürger et al., 2011; Heimbürger-Boavida et al., 2010). Metal contamination, known to impact aquatic 47 communities (size, distribution and density, species richness, composition of community) is a 48 major concern (Nogales et al., 2011; Zhou et al., 2008; Zhu et al., 2020). Marseille is the most 49 important city of the French Mediterranean coast with a dense urbanization and important 50 industrial, port and tourism activities. Thus, there is a need to monitor and to map the metallic 51 52 contamination and to provide reliable bioindicators in order to accurately evaluate the 53 abundance of hazardous metals that could affect coastal marine communities of this area.

54

Evaluation of metal contamination on marine organisms and ecosystems is often focused on species of commercial interest such as teleosts, decapods, bivalves and sea urchins, partly in order to estimate the sanitary risks for consumers or the economic risks of the loss of productivity for fisheries or farms (Liu et al., 2019; Mille et al., 2018; Penz et al., 2011; Schuhmacher et al., 1994; Zhang et al., 2012). Today, the most widely used bioindicators in

the marine environment are bivalves (Aly et al., 2013; Azizi et al., 2018; Beyer et al., 2017; 60 Gentric, 2016; Lafabrie et al., 2007; Pérez et al., 2004). Considering the ongoing major 61 biodiversity crisis (Ceballos et al., 2015, 2017; Eisenhauer et al., 2019), it is timely to extend 62 our knowledge concerning the impact of anthropic activities on ecosystem contamination by 63 studying a larger set of marine animals (Chifflet et al., 2022). To this purpose, several studies 64 65 investigated metal accumulation in sponges, as being filter feeders and sessile organisms, mainly in Demospongiae and Calcarea (Cebrian, 2007; Cebrian et al., 2003; Hansen et al., 1995; 66 Müller et al., 1998; Patel et al., 1985; Perez et al., 2005; Pérez et al., 2004). We propose to 67 evaluate for the first time the relevance of a Mediterranean sponge belonging to 68 Homoscleromorpha, Oscarella lobularis (Schmidt, 1862), as bioindicator for metal 69 70 contamination. Sponges (Porifera) are present in all marine habitats and gather about 9500 described species (de Voogd et al., 2022). In the Mediterranean Sea, sponges represent more 71 than 12% of marine fauna diversity (the same order as vertebrates) and are one of the main 72 organisms within benthic communities (Coll et al., 2010). The large amounts of water filtered 73 by sponges through their aquiferous system (Morganti et al., 2019; Weisz et al., 2008) yields 74 to the accumulation of metals present both in the dissolved and particulate phases (Caplat et 75 76 al., 2011; Orani et al., 2020; Rao et al., 2006). Several sponge species have already been used 77 as bioindicators of metal contamination (Keren et al., 2017; Khati et al., 2018; Mahaut et al., 78 2013; Pan et al., 2011). However, only few studies explored if the accumulation of metals 79 depends on sponge species (Orani, Barats, Vassileva, et al., 2018; Pérez et al., 2004; Roveta et al., 2020) thereby questioning the relevance of using only one species to monitor a large set 80 81 of environmental metal contaminants. The number of sponge species for which a convenient 82 set of biological knowledge and experimental tools are available is still limited, making it thus

far difficult to use sponges as model organisms for environmental assessment and
 ecotoxicology.

85

In this context, Oscarella lobularis (Schmidt, 1862) provides several advantages: i) being 86 abundant in shallow rocky substrates of the north-western Mediterranean Sea, including at 87 the vicinity of the Marseille Metropole and the French Riviera (Ereskovsky et al., 2009; Renard 88 89 et al., 2021). Oscarella lobularis is part of the coralligenous biocenosis which represents a 90 biodiversity hotspot in Mediterranean ecosystems (Bertolino et al., 2013; Martin et al., 2014); ii) the taxonomic identification of the species is clear (Gazave et al., 2012, 2013) and the life 91 cycle, development and morphoanatomical organization were deeply studied (Ereskovsky et 92 93 al., 2009, 2013; Fierro-Constain et al., 2017); iii) the high regeneration capability (Ereskovsky et al., 2015) and the high number of buds obtained in vitro by asexual reproduction(Renard et 94 95 al., 2021; Rocher et al., 2020) provide useful replicates for experimental ecotoxicological 96 studies; iv) in addition, both genomic and transcriptomic resources (Belahbib et al., 2018; Schenkelaars et al., 2015; Vernale et al., 2021) and molecular tools are available for this 97 species (Borchiellini et al., 2021; Fierro-Constain et al., 2017; Fierro-Constaín et al., 2021; 98 Renard et al., 2021; Rocher et al., 2020). This is convenient and suitable to perform 99 100 ecotoxicological experiments at different complementary levels of the biological scale (from 101 morphological, to physiological, cellular, and molecular levels).

The aim of this study was to evaluate the suitability of *O. lobularis* as a biomonitor of metallic contamination in the Mediterranean Sea. For this purpose, (i) the concentrations of thirteen metals of environmental concern (Li, Al, Ti, Cr, Fe, Ni, Cu, Zn, As, Ag, Cd, Hg, Pb) were measured in individuals sampled at seven locations of the Bay of Marseille, and (ii) the metal

concentrations in *O. lobularis* were compared with six other sponge species, commonly found
 in sympatry to assess potential interspecific variations. The study sites are characterized by
 contrasted anthropogenic activities and were used to assess possible links between metal
 accumulation by these organisms to past and present anthropogenic activities.

- 110 2. Materials and Methods
- 111

112 2.1. Sampling strategy

According to the distribution of the homoscleromorph sponge *Oscarella lobularis* populations,
seven sites in the Bay of Marseille (Mejean, Frioul, Prado, Maire, Perez cave, Caramassaigne,
Riou) and one site out of the bay (Fourmigues) were selected (Figure 1).

116 **Figure 1**

At each site, 3-14 adult individuals and surface sediment were both collected manually by 117 scientific divers at the same depth (Supplementary Data Table 1). Because Maïre Island is a 118 known study site in our laboratory for its large quantities and diversity of sponges species, 2 119 120 or 3 individuals of six other sponge species were sampled at this site for comparative 121 purposes: three Desmospongiae (Aplysina cavernicola (Vacelet, 1959), Agelas oroides (Schmidt, 1864), Axinella damicornis (Esper, 1794)), one Calcarea (Clathrina clathrus (Schmidt, 122 1864)), one homoscleromorph of the Plakinidae family (Corticium candelabrum (Schmidt, 123 1862)) and one homoscleromorph of the Oscarellidae family, the sister species of Oscarella 124 125 *lobularis, Oscarella uberculate* (Schmidt, 1868). All samples were placed in situ in clean (never 126 used) plastic resealable-zip storage bags and brought back to the laboratory in a cool box 127 within 1 hour.

129 **2.2. Sample preparation and metal measurements**

Sponge epibionts were removed with ceramic forceps under a magnifier, organisms were briefly rinsed with distilled water to limit salt crystals, dried on paper towel and then stored at -20°C. For the sediments, after the residual sea water was removed, samples were stored at -20°C. The frozen sponges and sediments were freeze-dried (Heto PowerDry PL3000 freeze dryer, Thermo Scientific). Sediments were sieved to recover the fraction less than 2 mm and sponges were homogenized in an agate mortar.

An Inductively Coupled Plasma Mass Spectrometry (ICP-MS Perkin Elmer NexION® 300X) was 136 used to quantify Li, Al, Ti, Cr, Fe, Ni, Cu, Zn, As, Ag, Cd, and Pb (Supplementary Data Table 2 137 and 3). Before measurements, 300 mg (dry weight) of each sponge sample were digested (1.5 138 mL HCl 37% + 1.5 mL HNO₃ 69% + 0.5 mL H₂O₂ 30%, quality TMA (Trace Metal Analysis)) and 139 50 mg (dry weight) of sediment were digested (1.5 mL HCl 37% + 1.5mL HNO₃ 69% + 0.5 mL 140 141 HF 40%, quality TMA (Trace Metal Analysis)). The samples were then mineralized with a 142 microwave (UltraWave, Milestone) (20-130°C in 15min – 130°C during 10 min – 130°C to 210°C in 10 min – 210°C during 10 min). Samples were diluted by a factor of 10-50 before analysis 143 144 by ICP-MS. Procedural blank and a standard sample (corn powder V463, INRAE) were analyzed for each series of mineralization (Fehlauer et al., 2022). 145

Total Hg measurements were performed on aliquots of about 50 mg dry weight (Supplementary Data Table 3) using atomic absorption spectrometry (AAS, Advanced Mercury Analyzer-254, Altec) equipped with a low Hg optical cell. The method detection limit was 2 pg, calculated as 3 times the standard deviation of the blank samples. The measured values were always within ± 5% of the recommended values of the certified reference materials MESS-4 (marine sediment, National Council of Canada) and DORM-4 (fish protein, National Council of Canada).

153 **2.3. Data analyses**

Principal Components Analysis (PCA) was performed to highlight the different trends in sponge metal content between sites and species. Because the Shapiro-Wilk and the Bartlett tests (performed in Rstudio) showed that the data do not fit the normality and homoscedasticity criteria, the significance of metal variations was assessed using a nonparametric Kruskal-Wallis analysis, followed by a Dunn post hoc test (p value < 0.05 considered significant). All PCA and statistical analyses were performed using Rstudio open-source edition package (https://www.rstudio.com/).

- 161 To calculate the bioconcentration factor (BCF), the metal concentrations found in sponge
- tissues were compared to the metal concentrations found in sediments sampled at the same
- 163 sites (Pan et al., 2011).

164 3. Results

165

166 **3.1. Difference in metal contents between Porifera species**

167 3.1.1. Intraspecific variability.

Variability between different individuals of the same species is calculated from the relative standard deviation (RSD) expressed in %. The highest mean variability is found for Cr (49.8%), Pb (30.7%) and Ag (30.3%). *Oscarella uberculate* has the highest variability in Cr (RSD = 88.7%) and Ag (RSD = 68.5%) while *Agelas oroides* has the highest RSD for Pb (60.4%) (Supplementary Data Table 4). In contrast, the lowest variations are observed for As (mean RSD = 13.6%) and Zn (mean RSD = 14.3%). The lowest variability for these two metals is found in *Corticium candelabrum* with 1.4% for Zn and 3.3% for As.

175

176 3.1.2. Interspecific variability.

A principal component analysis (PCA) of metal concentrations in the seven marine sponge 177 species collected in the Maïre site (for more information see Supplementary Data Table 5) was 178 179 performed (Figure 2). The first two axes of the PCA explain 60.2% of the total variability in the 180 data. Axis 1 of the PCA shows that the two demosponges, Aplysina cavernicola and Agelas 181 oroides, and the homoscleromorph Corticium candelabrum, display high concentrations of As $(211 \mu g/g, 200 \mu g/g, 387 \mu g/g respectively)$ and Hg (207 ng/g, 460 ng/g, 197 ng/g respectively)182 183 comparatively to the other sponge species. Axinella damicornis has high concentrations of Cr 184 (5.92 µg/g), Cu (52.2 µg/g), and Clathrina clathrus (Calcarea) has the highest concentrations 185 of Ti (32.7 μ g/g) and Li (2.67 μ g/g). Moreover, Ni, Cd, Zn, representing the second axis, are strongly concentrated in the homoscleromorph Oscarella uberculate (1564 μg/g, 48.5 μg/g,

187 4394 μ g/g, respectively.

188 In contrast, *Oscarella lobularis*, is localized at the center of the PCA. These finding highlights

that in contrast to the other species sampled at the Maïre site, O. lobularis displays a more

190 homogenous metal bioaccumulation profile than the other sponges studied here (Figure 2).

191 Figure 2

192 3.1.3. Bioconcentration factors in other marine sponges.

193 The bioconcentration factor (BCF) (Supplementary Data Table 6) determines the accumulation

194 capacity of organisms in their tissues (Pan et al., 2011).

The comparison of the bioconcentration factors between the seven sponges shows that the bioaccumulated metals are often similar (e.g., Cu and As show BCF>1 in all species; Ag and Cd show BCF>1 in all except one species, see Supplementary Data Table 6). In agreement with PCA analysis (Figure 2), figure 3 shows that some metals are much more highly accumulated in some species than in others (for example Ni and Cd in *Oscarella uberculate*).

200 Figure 3

201 3.2. O. lobularis' metal concentrations in eight sites of Marseille

202 3.2.1. Inter-site variations.

203 Metal concentrations measured in *Oscarella lobularis* at the different sites are presented in 204 figure 4. Al, Fe, Ni and Zn are the metals with the highest concentrations (Figure 4B) with 205 notable differences between sites. The Fourmigues individuals show the highest 206 concentrations of Zn (4920 μ g/g dw), Al (834 μ g/g dw), Ni (3277 μ g/g dw) and Cd (120 μ g/g 207 dw), 11 times higher for Zn than those sampled at Riou, in which the sampled specimens 208 present the lower Zn content. Sponges from the Prado reefs display the highest Fe body 209 content with 2460 μ g/g (4 times higher than sponges from Fourmigues) and the Maïre site 210 individuals show the highest As concentration (61.5 μ g/g dw), 3 times higher than in the ones 211 from Fourmigues (Figure 4A). For the other metals, the variations between sites are slighter.

212 Figure 4

A PCA was performed to analyze inter-site variations. The first two axes of the PCA explain 213 214 60.7% of the total variability of the data (Figure 5). The PCA plot shows two distinct clusters: 215 an upper left one characterized by Cr, Fe, Cu, As, Hg, Pb and an upper right one influenced by Li, Al, Ti, Ni, Zn, Ag, Cd. The first axis is mainly explained by the concentrations of Zn, Cd and 216 Ni in the tissues of Oscarella lobularis. In addition, body contents in Zn, Ni and Cd are 217 218 significantly higher in sponges from Fourmigues and in a lesser extent from Caramassaigne and Frioul (p < 0.05) (Figure 5 and Supplementary Data Table 5). The second axis of the PCA is 219 220 mainly driven by the concentrations of Al, Ti, Fe, Hg and Pb. Sponges from the Prado reef site 221 are characterized by high concentrations of Fe, Hg and Pb (Figure 5). Concerning Fe, Prado's reef and in a lesser extent Mejean populations display the significantly (p <0.05) highest 222 concentrations (2460 \pm 912 µg/g and 1740 \pm 197 µg/g respectively) (Supplementary Data 223 Table 5). The Frioul site is at the center of the PCA plot, this means that individuals display 224 more homogeneous metal contents than those of the other sites. The population of O. 225 226 lobularis from the Perez cave, Caramassaigne and Riou sites, located at the bottom of the 227 biplot, show lower concentrations of metals than those of the other sites located in front of the urban coast (Figure 5). 228

229 Figure 5

230 3.2.2. Bioconcentration factors in Oscarella lobularis.

As for other sponges, bioconcentration factor (BCF) for *Oscarella lobularis* was calculated
(Supplementary Data Table 6 for BCF).

BCFs are higher than 1 for Ag, As, Cd, Cu, Ni, Zn, and Hg, meaning that concentrations in Oscarella lobularis are higher than those in the sediments whatever the sampling sites (Supplementary Data Table 6). The highest BCF values are observed on 3 sites and for 3 metals (Figure 6): Ni at Frioul, Caramassaigne and Fourmigues (BCF ranging from 522 to 850), Cd at Caramassaigne (BCF = 584) and Zn at Fourmigues (BCF = 259). In contrast, Al, Cr, Fe, Li, Pb and Ti, show bioconcentration factors lower than 1, meaning that the concentrations in Oscarella *lobularis* tissues are lower than those measured in the surrounding sediment.

240 Figure 6

241 4. Discussion

242

243 Metals are naturally present in the environment in the form of geochemical backgrounds. The geological formations of the Marseille basin are composed of many deposits of bauxite 244 (characterized by Al₂O₃ and iron oxides) formerly exploited, and many layers of terra rossa 245 (i.e., iron oxides). Even though these rocks naturally contain metals, because the concerned 246 247 area is characterized by the dominance of calcareous sedimentary rocks, the basicity of these 248 rocks makes metals hardly mobile and available (Ciesielski, 2007). Thereby natural origin of metals in the sea is usually considered neglectable in this geographic area (Chandesris et al., 249 2013; Tchounwou et al., 2012; Urien et al., 2017). Anthropic rock mining can nevertheless 250 251 increase ground leaching and bring natural metals to the sea by runoff, in addition to industrial 252 activities commonly considered to be responsible for most metal contaminations in the area.

253 The analysis of metals on the surface sediment in the bay of Marseille shows higher concentrations for Ni, Cd, Cu, Zn (with a mean of 4.46 µg/g, 0.172 µg/g, 4.31 µg/g and 51.0 254 μ g/g respectively) and lower concentrations for Cr and Pb (with a mean of 10.6 μ g/g and 21.4 255 μ g/g respectively) (Supplementary Data Table 5) compared to the sediments in the Gulf of 256 Lions (Roussiez et al., 2006). Moreover, in the Ligurian Sea, the concentrations are higher for 257 258 Cr, Cu, Li, Ni, Zn and are lower for Al, Fe, Hg compared to surface sediment in the bay (Heimbürger et al., 2012). Concerning the bay of Toulon, the metal concentrations are much 259 higher than in the bay of Marseille, this is in agreement of a higher anthropic contamination 260 of this area (Tessier et al., 2011). However, the surface sediment represents the first 261 centimetres and thus supposedly the most recent particle deposits in the study area at a given 262 time (Martín et al., 2009; Tessier et al., 2011), making it a partial indicator of the quality of the 263 environment. In contrast, sponges, which are sessile organisms and active filter feeders, 264 represent a relevant proxy to monitor and evaluate metal contamination in addition to 265 sediment metal contents (Cebrian, 2007; Pan et al., 2011). 266

267 **4.1. Species-dependent and metal-dependent concentrations in sponge tissues.**

268 According to our results for a given site, metal concentrations are both sponge species-269 dependent and metal-dependent. Among the different species sampled in our study, 270 Oscarella lobularis and Agelas oroides show the highest mean variabilities (26.8% and 33.1% 271 respectively), and concerning metals, As, Zn and Cu show the lowest mean variabilities (13.6%, 272 14.3% and 15.3% respectively) (Supplementary Data Table 4), like in the study by Orani et al., 273 2018. Whatever the sponge species considered, Ag, As, Cd, Cu, Ni, Zn and Hg are present in 274 higher concentrations in sponges than in sediments (i.e., 2- to 37-fold higher than in sediment) 275 (Figure 3), as observed for other sponge species from the Mediterranean Sea (M. Araújo et al., 276 2003; Cebrian, 2007; Orani, Barats, Vassileva, et al., 2018) or from the Pacific Ocean (Padovan

et al., 2012). This consensual finding could give an indication of the bioavailability in Ag, As,
Cd, Cu, Ni, Zn and Hg. High metal concentrations in tissues are generally considered to present
a risk for organisms health, but bioaccumulation of some metals within organism tissues can
sometimes be explained by their biological role. For example, Cu is involved in sponge
metabolism as well as various enzymatic processes (Tian et al., 2014) while As can play a
protective role against oxidative stress (Orani, Barats, Zitte, et al., 2018; Yamaoka et al., 2001).

Metal concentrations vary across species with well-defined signatures for each species (Figure 283 2; As and Hg for C. candelabrum; Cr, Ti, Li and Cu for A. damicornis and C. clathrus; Ni, Cd and 284 Zn for O.tuberculata). In contrast Oscarella lobularis displays a more homogeneous metal 285 accumulation pattern than the other sponge species studied. High inter-specific differences in 286 metal bioaccumulation were already observed in previous studies performed on different sets 287 288 of sponge species from different geographic areas (Batista et al., 2014; Genta-Jouve et al., 289 2012; Patel et al., 1985). This finding may be partly explained by i) different filtration speed and efficiency between sponge species (Cebrian, 2007; Morganti et al., 2019; Weisz et al., 290 291 2008); ii) different molecular, cytological and histological features between species, as it had been shown that different Demospongiae species accumulate metals in various structures like 292 293 collagen, skeleton fibers, pinacoderm (i.e., external cell layer of sponge body) or even mesohylar cells (i.e., internal tissue of sponge) (Orani, Barats, Zitte, et al., 2018; Padovan et 294 al., 2012), by iii) different food preference (Maldonado, 2016; Perez et al., 2005), or by iv) 295 differences in associated microorganisms (Keren et al., 2017; Lee et al., 2009; Selvin et al., 296 297 2009; Tian et al., 2014). In our case, the huge differences observed between O.lobularis and 298 O.tuberculata BCFs (Supplementary Data Table 6) cannot be explained neither by molecular, 299 cytological nor histological differences. Indeed, the high similarity between these sister species has for long resulted in the absence of species discrimination (Boury-Esnault et al., 300

1992; Gazave et al., 2013). Such differences between species of a same genus has also been
observed in the genus *Mytilus* (Sussarellu et al., 2022). The microbial communities of *Oscarella lobularis* display a species-specific microbial diversity dominated by a Rhodobacteria (Gerçe
et al., 2011; Gloeckner et al., 2013; Jourda et al., 2015) but further studies would be needed
to decipher the relative contribution to metal accumulation of the sponge and its symbiotic
microorganisms.

307 4.2. Intersite differences in *Oscarella lobularis* and potential links with the industrial past and
 308 present anthropic activities.

Although Oscarella lobularis accumulates metals more homogeneously than other sponge 309 species, we note a variability of metal contents in tissues between the different sampling sites 310 (Figure 5). These differences may be related to the past and present industrial activities in the 311 bay of Marseille. The industrialization of Marseille increased in the middle of the 19th century 312 with the creation of factories of several types (glass, petroleum, soda, tartaric acid, etc.). No 313 obvious direct link has been made between metal contaminations in the bay and the factories, 314 315 except concerning the industrial sites of Montredon (1873-1884), Escalette (1851-1924) and Goudes (1854-1877). Located in the east of Marseille, these sites were specialized in Ag and 316 317 Pb production (Daumalin et al., 2016) (Figure 7). The metallic contamination in terrestrial and water compartments in the area of these former industries, located in front of the Maïre site, 318 was previously characterized (Lasalle, 2007), and high concentrations of Pb and As in soils (633 319 μ g/g and 29.4 μ g/g respectively), in marine sediments (99 to 1189 μ g/g and 8.2 to 648 μ g/g) 320 321 and in marine organisms were highlighted. For example, in mussels, Pb and As are respectively 322 up to 4 and 2 times higher in comparison with mussels from other sites of the Mediterranean 323 sea (Andral et al., 2004; Santos-Echeandía et al., 2021), with highest Pb and As body amounts ranging from 4.5 to 5.3 μ g/g dw and from 25.4 to 26.2 μ g/g dw, respectively. Interestingly, O. 324

lobularis specimens sampled in the Maïre site, located in the vicinity of this former industrial site, show the highest concentrations of Pb ($8.76 \pm 0.87 \mu g/g dw$) and As ($61.5 \pm 10.3 \mu g/g dw$) measured among the different studied *O. lobularis* populations. Our results are therefore in agreement with previous studies reporting that this area of the Bay of Marseille is still exposed to a significant metallic contamination despite that these factories have been closed since a century (Daumalin et al., 2016; Lasalle, 2007).

Marseille is the most important trade and cruise ship port of the France south littoral and has 331 332 the largest underground wastewater treatment plant in Europe in Calanques National Park located at 5 km from the city. The rejected treated water, not free of metals, discharges Ag, 333 Cu, Pb, Cr, Hg and Fe at the Cortiou Cove during dry conditions (Millet et al., 2018; Oursel, 334 2013; Tumolo et al., 2020) and into the autonomous harbour ("Port autonome") of Marseille, 335 the old harbour ("vieux port") and at the level of the Prado site, without prior passage through 336 the treatment plant During rainy conditions. Finally, the small coastal rivers such as the 337 338 Huveaune, the Jarret and the Aygalades that run through the town, as well as irregular big intrusion of the Rhône River (Fraysse et al., 2013), were already shown to collect and transport 339 contaminants toward the bay (i.e., Hg up to 62 ng/L in the Huveaune and Aygalades) (Jany & 340 341 Zebracki, 2012) (Figure 7). The hydrodynamics of the Gulf of Lion, with the southeast marine winds and the North Current, could explain the spread of contaminants and their localization 342 on certain sites in the Bay of Marseille. In accordance with previous studies on sponges of the 343 Marseille region (Perez et al., 2005), our results indicate for example that the populations 344 present in the bay (i.e., Prado) have significant quantities of Fe. These large quantities of Fe 345 346 could be due to the use of ferric chloride as a coagulation agent for the physico-chemical treatment of wastewater at the Cortiou plant and could be transported to the Prado reefs 347 348 with the North Current toward the bay (Figure 7). Fe isotope studies would confirm or refute

this hypothesis by specifically determining its source (natural or anthropogenic), as performed in previous studies for other metals in the Mediterranean Sea (D. F. Araújo et al., 2019; Orani, Barats, Vassileva, et al., 2018). Specimens sampled at the sites of Riou, Caramassaigne and Perez Cave are all present in the bottom of the PCA, below the second axis, reflecting lower metallic bioaccumulations in comparison to the other populations, that may be due to the hydrodynamic characteristics of these sites (Figure 7) and their respective distance from the urbanized area.

356 Figure 7

357 Sponges sampled in the Fourmigues site, located at the east of the Giens peninsula, are all grouped in the right side of the PCA and are characterized by high concentrations of Zn, Ni 358 and Cd (Figure 5). The Toulon Bay, located at about 10 km from the Fourmigues sampling site, 359 is characterized by important industrial, touristic, and military nautical activities, and is 360 361 submitted to significant inputs of metals. In a previous study, high Zn concentrations with a range of $24.3 - 1340 \,\mu$ g/g dw were found in sediments in the large bay of Toulon (Tessier et 362 363 al., 2011). High Zn content in sediments could be due to the intensive use of antifouling paints on boat hulls to prevent corrosion and fouling, which would be consistent with the intensive 364 nautical activity of this area (Jones & Turner, 2010). In addition, the bay of Toulon was also 365 reported to present high Cd concentration in seawater (between 0.007 and 0.14 µg/L) (Rossi 366 367 & Jamet, 2008) and, in the port of Porquerolles Island (located in the vicinity of Toulon), Zn, Ni and Cd contamination in water, sediment and marine organisms were linked to the 368 369 maritime activities of the port (Augier, 1980). Therefore, our data at the Fourmigues site 370 suggest that at least part of the contamination from the bay of Toulon and from Porquerolles Island may extend to the surrounding coastal waters. In particular, the North Current may be 371

372 responsible for the high concentrations of Ni (210-320 ng/L) previously measured in the
373 Ligurian sea located at the East of the Fourmigues site as suggested by (Migon et al., 2020).

374

4.3. Oscarella lobularis as a valuable bioindicator for metallic contamination

376 Finally, the inter-site variability does not question the relevance of Oscarella lobularis as valuable biomonitor: its homogeneous metal bioaccumulation profile makes it a good 377 candidate as a bioindicator of metallic pollution in Mediterranean Sea. The choice of sentinel 378 or bioindicator species should be guided by several criteria: i) preferring sessile species or 379 species displaying limited dispersion (Azizi et al., 2018; Vieira et al., 2021), ii) opting for filter 380 feeders (Batista et al., 2014; Milanese et al., 2003), iii) identifying, capturing and collecting 381 easily a species with a sufficient population size to study spatial variations of ecosystems 382 (Berthet, 2013). Oscarella lobularis fits all these criteria. Even if, several sponges have been 383 proposed as bioindicator species (mostly Demospongiae but also one species of Calcarea 384 Paraleucilla magna) (Batista et al., 2014), as far as we know, only one species (Haliclona fulva 385 386 (Topsent, 1893)) has been showed as a generalist species in the Mediterranean Sea (Orani, 387 Barats, Vassileva, et al., 2018), which appears to be a crucial criterion for choosing a sentinel organism. 388

389 Conclusions

390

This study is the first one i) reporting such a dataset concerning the bioaccumulation of metals in the bay of Marseille using a homoscleromorph sponge; ii) comparing different bioaccumulation profiles between sponge species at the Maïre site in order to objectively compare the relative relevance of sponge species for environmental assessment.

According to its numerous features fitting the ideal characteristics required for the use of a relevant model to biomonitor the metallic contamination in marine coastal waters, our work suggests *Oscarella lobularis* as a new bioindicator of contaminants in Mediterranean coastal environments. Consequently, *O.lobularis* and *Haliclona fulva* may be useful to implement a "Sponge Watch Program" for the western part of the Mediterranean Sea, as it was proposed in Denmark with the demosponge *Halichondria panicea* (Hansen et al., 1995; Olesen & Weeks, 1994).

The present results ar"also expected to be useful for future ecotoxicological studies. Indeed, *Oscarella lobularis* is now one of the best described Mediterranean sponge species at chemical, physiological, morphological, reproductive, cellular and molecular levels (Belahbib et al., 2018; Ereskovsky et al., 2015; Fierro-Constain et al., 2017; Renard et al., 2021; Rocher et al., 2020; Vernale et al., 2021), thereby enabling integrative approaches.

407 Acknowledgments

408

The authors acknowledge the CNRS for the funding of K. De Pao's PhD grant through the 409 80|Prime program. This work was also cofounded by the Eccorev research federation and by 410 the Labex DRIIHM, French program "Investissements d'Avenir" (ANR-1-LABX-0010) which is 411 managed by the French ANR. The authors also thank the diving staff of the OSU Pythéas (in 412 413 particular Laurent Vanbostal and Dorian Guillemain) and the divers of the IMBE lab (in particular Sandrine Chenesseau and Christian Marschal) as well as Sandrine Ruitton (MIO) and 414 Stephane Sartoretto (IFREMER) for sampling sponges and sediment. For experiments, the 415 416 authors thank the LA-ICP-MS platform: elemental chemistry at Aix-Marseille. Geneviève Dur 417 (CEREGE) and Caroline Rocher (IMBE) are warmly thanked for their help in the preparation of

- samples and providing efficient lab facilities. We acknowledge Pr Jean Borgomano for the
- 419 geological information he provided on the studied area.

420 **References:**

- 421 Aly, W., Williams, I. D., & Hudson, M. D. (2013). Metal contamination in water, sediment and biota
- 422 from a semi-enclosed coastal area. Environmental Monitoring and Assessment, 185(5),
- 423 3879-3895. https://doi.org/10.1007/s10661-012-2837-0
- 424 Andral, B., Stanisiere, J. Y., Sauzade, D., Damier, E., Thebault, H., Galgani, F., & Boissery, P. (2004).
- 425 Monitoring chemical contamination levels in the Mediterranean based on the use of mussel
- 426 caging. *Marine Pollution Bulletin*, 49(9), 704-712.
- 427 https://doi.org/10.1016/j.marpolbul.2004.05.008
- 428 Araújo, D. F., Ponzevera, E., Briant, N., Knoery, J., Bruzac, S., Sireau, T., & Brach-Papa, C. (2019).
- 429 Copper, zinc and lead isotope signatures of sediments from a mediterranean coastal bay
- 430 impacted by naval activities and urban sources. *Applied Geochemistry*, 111, 104440-.
- 431 https://doi.org/10.1016/j.apgeochem.2019.104440
- 432 Araújo, M., Conceição, A., Barbosa, T., Lopes, M., & Humanes, M. (2003). Elemental composition of
- 433 marine sponges from the Berlengas Natural Park, western Portuguese coast. *X-Ray*

434 *Spectrometry*, *32*, 428-433. https://doi.org/10.1002/xrs.660

- 435 Arnaud, A., Prudent, P., & Laffont-Schwob, I. (2021). Évaluation du risque sanitaire de sols pollués
- 436 méditerranéens : Choix de variables et spatialisation. VertigO la revue électronique en
- 437 sciences de l'environnement, Volume 21 numéro 1, Art. Volume 21 numéro 1.
- 438 https://doi.org/10.4000/vertigo.31309
- Augier. (1980). Le port de Porquerolles (Îles d'Hyères, Mediterranée, France) II. Contamination par les
 metaux lourds | Parc national de Port-Cros et Porquerolles. http://www.portcros-
- 441 parcnational.fr/fr/rapports-scientifiques/le-port-de-porquerolles-iles-dhyeres-mediterranee-
- 442 france-ii-contamination-par
- 443 Azizi, G., M., A., Baghour, M., Mostafa, L., & Moumen, A. (2018). The use of Mytilus spp. Mussels as
- bioindicators of heavy metal pollution in the coastal environment. A review. *Journal of*
- 445 Materials and Environmental Science, 9. https://doi.org/10.26872/jmes.2018.9.4.129

- 446 Batista, D., Muricy, G., Rocha, R. C., & Miekeley, N. F. (2014). Marine sponges with contrasting life
- 447 histories can be complementary biomonitors of heavy metal pollution in coastal ecosystems.
- 448 Environmental Science and Pollution Research International, 21(9), 5785-5794.
- 449 https://doi.org/10.1007/s11356-014-2530-7
- 450 Belahbib, H., Renard, E., Santini, S., Jourda, C., Claverie, J.-M., Borchiellini, C., & Le Bivic, A. (2018).
- 451 New genomic data and analyses challenge the traditional vision of animal epithelium
- 452 evolution. *BMC Genomics*, *19*(1), 393. https://doi.org/10.1186/s12864-018-4715-9
- 453 Berthet, B. (2013). Sentinel Species. In *Ecological Biomarkers* (p. 155-185).
- 454 https://doi.org/10.1201/b13036-8
- 455 Bertolino, M., Cerrano, C., Bavestrello, G., Carella, M., Pansini, M., & Calcinai, B. (2013). Diversity of
- 456 Porifera in the Mediterranean coralligenous accretions, with description of a new species.
- 457 *ZooKeys*, *336*, 1-37. https://doi.org/10.3897/zookeys.336.5139
- 458 Beyer, J., Green, N. W., Brooks, S., Allan, I. J., Ruus, A., Gomes, T., Bråte, I. L. N., & Schøyen, M.
- 459 (2017). Blue mussels (Mytilus edulis spp.) as sentinel organisms in coastal pollution
- 460 monitoring : A review. *Marine Environmental Research*, *130*, 338-365.
- 461 https://doi.org/10.1016/j.marenvres.2017.07.024
- 462 Bianchi, C. N., & Morri, C. (2000). Marine Biodiversity of the Mediterranean Sea : Situation, Problems
- and Prospects for Future Research. *Marine Pollution Bulletin*, 40(5), 367-376.
- 464 https://doi.org/10.1016/S0025-326X(00)00027-8
- 465 Borchiellini, C., Degnan, S., Goff, E., Rocher, C., Vernale, A., Baghdiguian, S., Séjourné, N., Marschal,
- 466 F., Le Bivic, A., Godefroy, N., Degnan, B., & Renard, E. (2021). Staining and Tracking Methods
- 467 for Studying Sponge Cell Dynamics. In *Methods in molecular biology (Clifton, N.J.)* (Vol. 2219,
- 468 p. 81-97). https://doi.org/10.1007/978-1-0716-0974-3_5
- 469 Boury-Esnault, N., Solé-Cava, A., & Thorpe, J. P. (1992). Genetic and cytological divergence between
- 470 colour morphs of the Mediterranean sponge Oscarella lobularis Schmidt (Porifera,

- 471 Demospongiae, Oscarellidae). *Journal of Natural History*, *26*, 271-284.
- 472 https://doi.org/10.1080/00222939200770131
- 473 Caplat, C., Basuyaux, O., & Mahaut, M.-L. (2011, juillet 3). Water quality assessment using a sponge
 474 (Hymeniacidon perlevis) as bioindicator—First results of metallic and organic contaminations
 475 given by biotests.
- 476 Ceballos, G., Ehrlich, P. R., Barnosky, A. D., García, A., Pringle, R. M., & Palmer, T. M. (2015).
- 477 Accelerated modern human–induced species losses : Entering the sixth mass extinction.
- 478 *Science Advances*, *1*(5), e1400253. https://doi.org/10.1126/sciadv.1400253
- 479 Ceballos, G., Ehrlich, P. R., & Dirzo, R. (2017). Biological annihilation via the ongoing sixth mass
- 480 extinction signaled by vertebrate population losses and declines. *Proceedings of the National*
- 481 Academy of Sciences, 114(30), E6089-E6096. https://doi.org/10.1073/pnas.1704949114
- 482 Cebrian, E. (2007). Sponges as biomonitors of heavy metals in spatial and temporal surveys in
- 483 northwestern mediterranean : Multispecies comparison. PubMed—NCBI.
- 484 https://www.ncbi.nlm.nih.gov/pubmed/17941749
- 485 Cebrian, E., Martí, R., Uriz, J. M., & Turon, X. (2003). Sublethal effects of contamination on the
- 486 Mediterranean sponge Crambe crambe : Metal accumulation and biological responses.
- 487 *Marine Pollution Bulletin*, 46(10), 1273-1284. https://doi.org/10.1016/S0025-326X(03)00190-
- 488

- Chandesris, A., Canal, J., Coquery, M., & Bougon, N. (2013). Détermination du fond géochimique pour *les métaux dissous dans les eaux de surface continentales. Rapport final* (p. 231) [Research
 Report]. irstea. https://hal.inrae.fr/hal-02599390
- 492 Chifflet, S., Briant, N., Tesán-Onrubia, J. A., Zaaboub, N., Amri, S., Radakovitch, O., Bǎnaru, D., &
- 493 Tedetti, M. (2022). Distribution and accumulation of metals and metalloids in planktonic food
- 494 webs of the Mediterranean Sea (MERITE-HIPPOCAMPE campaign). *Marine Pollution Bulletin*,
- 495 *186*, 114384. https://doi.org/10.1016/j.marpolbul.2022.114384

- 496 Ciesielski, H. (2007). Effets du pH sur l'extraction des éléments traces métalliques dans les sols. *Etude*497 *et Gestion des Sols*, 24.
- 498 Coll, M., Piroddi, C., Steenbeek, J., Kaschner, K., Ben Rais Lasram, F., Aguzzi, J., Ballesteros, E., Bianchi,
- 499 C. N., Corbera, J., Dailianis, T., Danovaro, R., Estrada, M., Froglia, C., Galil, B. S., Gasol, J. M.,
- 500 Gertwagen, R., Gil, J., Guilhaumon, F., Kesner-Reyes, K., ... Voultsiadou, E. (2010). The
- 501 Biodiversity of the Mediterranean Sea : Estimates, Patterns, and Threats. *PLoS ONE*, 5(8),
- 502 e11842. https://doi.org/10.1371/journal.pone.0011842
- 503 Danovaro, R. (2003). Pollution threats in the Mediterranean Sea : An overview. *Chemistry and* 504 *Ecology*, *19*(1), 15-32. https://doi.org/10.1080/0275754031000081467
- 505 Daumalin, X., Laffont-Schwob, I., Barthélemy, C., Desrousseaux, M., Heckenroth, A., Masotti, V.,
- 506 Monsara, P., Kronenberger, S., Oursel, B., Prudent, P., Rabier, J., & Raveux, O. (2016). Les
- 507 calanques industrielles de Marseille et leurs pollutions—Une histoire au présent.
- 508 https://hal.archives-ouvertes.fr/hal-01437604
- de Voogd, N., Alvarez, B., Boury-Esnault, N., Carballo, J. L., Cárdenas, P., Díaz, M.-C., Dohrmann, M.,
- 510 Downey, R., Hajdu, E., Hooper, J. N. A., Kelly, M., Klautau, M., Manconi, R., Morrow, C. C.,
- 511 Pisera, A. B., Ríos, P., Rützler, K., Schönberg, C., Vacelet, J., & van Soest, R. W. M. (2022).
- 512 World Porifera Database. https://doi.org/10.14284/359
- 513 Durrieu de Madron, X., Guieu, C., Sempéré, R., Conan, P., Cossa, D., D'Ortenzio, F., Estournel, C.,
- 514 Gazeau, F., Rabouille, C., Stemmann, L., Bonnet, S., Diaz, F., Koubbi, P., Radakovitch, O.,
- 515 Babin, M., Baklouti, M., Bancon-Montigny, C., Belviso, S., Bensoussan, N., ... Verney, R.
- 516 (2011). Marine ecosystems' responses to climatic and anthropogenic forcings in the
- 517 Mediterranean. *Progress in Oceanography*, *91*(2), 97-166.
- 518 https://doi.org/10.1016/j.pocean.2011.02.003
- Eisenhauer, N., Bonn, A., & A. Guerra, C. (2019). Recognizing the quiet extinction of invertebrates.
 Nature Communications, *10*(1), Art. 1. https://doi.org/10.1038/s41467-018-07916-1

- 521 Ereskovsky, A. V., Borchiellini, C., Gazave, E., Ivanisevic, J., Lapébie, P., Perez, T., Renard, E., &
- 522 Vacelet, J. (2009). The Homoscleromorph sponge Oscarella lobularis, a promising sponge
 523 model in evolutionary and developmental biology. *BioEssays*, *31*(1), 89-97.
- 524 https://doi.org/10.1002/bies.080058
- 525 Ereskovsky, A. V., Borisenko, I. E., Lapébie, P., Gazave, E., Tokina, D. B., & Borchiellini, C. (2015).
- 526 Oscarella lobularis (Homoscleromorpha, Porifera) Regeneration : Epithelial Morphogenesis
- 527 and Metaplasia. *PloS One, 10*(8), e0134566. https://doi.org/10.1371/journal.pone.0134566
- 528 Ereskovsky, A. V., Dubois, M., Ivanišević, J., Gazave, E., Lapebie, P., Tokina, D., & Pérez, T. (2013).
- 529 Pluri-annual study of the reproduction of two Mediterranean Oscarella species (Porifera,
- 530 Homoscleromorpha) : Cycle, sex-ratio, reproductive effort and phenology. *Marine Biology*,
- 531 *160*(2), 423-438. https://doi.org/10.1007/s00227-012-2100-9
- 532 European Environment Agency (Éd.). (2006). *Priority issues in the Mediterranean environment*.
- 533 European Environmental Agency.
- 534 Fehlauer, T., Collin, B., Angeletti, B., Santaella, C., Dentant, C., Chaurand, P., Levard, C., Gonneau, C.,
- 535 Borschneck, D., & Rose, J. (2022). Uptake patterns of critical metals in alpine plant species
- 536 growing in an unimpaired natural site. *Chemosphere*, *287*(4), 132315.
- 537 https://doi.org/10.1016/j.chemosphere.2021.132315
- 538 Fierro-Constaín, L., Rocher, C., Marschal, F., Schenkelaars, Q., Séjourné, N., Borchiellini, C., & Renard,
- 539 E. (2021). In Situ Hybridization Techniques in the Homoscleromorph Sponge Oscarella
- 540 lobularis. *Methods in Molecular Biology (Clifton, N.J.), 2219,* 181-194.
- 541 https://doi.org/10.1007/978-1-0716-0974-3_11
- 542 Fierro-Constain, L., Schenkelaars, Q., Gazave, E., Haguenauer, A., Rocher, C., Ereskovsky, A.,
- 543 Borchiellini, C., & Renard, E. (2017). The Conservation of the Germline Multipotency
- 544 Program, from Sponges to Vertebrates : A Stepping Stone to Understanding the Somatic and
- 545 Germline Origins. *Genome Biology and Evolution*, 22(3), 474-488.
- 546 https://doi.org/10.1093/gbe/evw289

- 547 Fraysse, M., Pairaud, I., ROSS, O. N., FAURE, V., & PINAZO, C. (2014). Intrusion of Rhone River diluted
- 548 water into the Bay of Marseille : Generation processes and impacts on ecosystem
- 549 functioning. *Journal of Geophysical Research. Oceans*.
- 550 https://doi.org/10.1002/2014JC010022
- 551 Fraysse, M., PINAZO, C., Faure, V. M., Fuchs, R., Lazzari, P., Raimbault, P., & Pairaud, I. (2013).
- 552 Development of a 3D Coupled Physical-Biogeochemical Model for the Marseille Coastal Area
- 553 (NW Mediterranean Sea) : What Complexity Is Required in the Coastal Zone? PLoS ONE,
- 554 *8*(12), e80012. https://doi.org/10.1371/journal.pone.0080012
- 555 Gazave, E., Lapébie, P., Ereskovsky, A. V., Vacelet, J., Renard, E., Cárdenas, P., & Borchiellini, C.
- 556 (2012). No longer Demospongiae : Homoscleromorph sponges revisited by molecular
- 557 phylogeny, Linnaean classification and the PhyloCode. *Hydrobiologia*, 687(1), 3-10.
- 558 https://doi.org/10.1007/s10750-011-0842-x
- 559 Gazave, E., Lavrov, D. V., Cabrol, J., Renard, E., Rocher, C., Vacelet, J., Adamska, M., Borchiellini, C., &
- 560 Ereskovsky, A. V. (2013). Systematics and Molecular Phylogeny of the Family Oscarellidae
- 561 (Homoscleromorpha) with Description of Two New Oscarella Species. *PLOS ONE*, *8*(5),
- 562 e63976. https://doi.org/10.1371/journal.pone.0063976
- 563 Genta-Jouve, G., Cachet, N., Oberhänsli, F., Noyer, C., Teyssié, J.-L., Thomas, O. P., & Lacoue-
- 564 Labarthe, T. (2012). Comparative bioaccumulation kinetics of trace elements in
- 565 Mediterranean marine sponges. *Chemosphere*, *89*(3), 340-349.
- 566 https://doi.org/10.1016/j.chemosphere.2012.04.052
- 567 Gentric. (2016). *Bioaccumulation of metallic trace elements and organic pollutants in marine sponges*
- 568 from the South Brittany Coast, France. PubMed—NCBI.
- 569 https://www.ncbi.nlm.nih.gov/pubmed/26634290
- 570 Gerçe, B., Schwartz, T., Syldatk, C., & Hausmann, R. (2011). Differences Between Bacterial
- 571 Communities Associated with the Surface or Tissue of Mediterranean Sponge Species.
- 572 *Microbial Ecology*, *61*(4), 769-782. https://doi.org/10.1007/s00248-011-9802-2

- 573 Gloeckner, V., Hentschel, U., Ereskovsky, A. V., & Schmitt, S. (2013). Unique and species-specific
- 574 microbial communities in Oscarella lobularis and other Mediterranean Oscarella species

575 (Porifera : Homoscleromorpha). *Marine Biology*, *160*(4), 781-791.

- 576 https://doi.org/10.1007/s00227-012-2133-0
- 577 Hansen, I. V., Weeks, J. M., & Depledge, M. H. (1995). Accumulation of copper, zinc, cadmium and
- 578 chromium by the marine sponge Halichondria panicea Pallas and the implications for
- 579 biomonitoring. Marine Pollution Bulletin, 31(1), 133-138. https://doi.org/10.1016/0025-
- 580 326X(94)00228-2
- 581 Heimbürger, L.-E., Cossa, D., Thibodeau, B., Khripounoff, A., Mas, V., Chiffoleau, J.-F., Schmidt, S., &
- 582 Migon, C. (2012). Natural and anthropogenic trace metals in sediments of the Ligurian Sea
- 583 (Northwestern Mediterranean). *Chemical Geology*, 291, 141-151.
- 584 https://doi.org/10.1016/j.chemgeo.2011.10.011
- 585 Heimbürger, L.-E., Migon, C., & Cossa, D. (2011). Impact of atmospheric deposition of anthropogenic
- 586 and natural trace metals on Northwestern Mediterranean surface waters : A box model
- 587 assessment. *Environmental Pollution*, 159(6), 1629-1634.
- 588 https://doi.org/10.1016/j.envpol.2011.02.046
- Heimbürger-Boavida, L.-E., Migon, C., Dufour, A., Chiffoleau, J.-F., & Cossa, D. (2010). Trace metal
- 590 concentrations in the North-western Mediterranean atmospheric aerosol between 1986 and
- 591 2008 : Seasonal patterns and decadal trends. *Science of the Total Environment*, 408(13),

592 2629-2638. https://doi.org/10.1016/j.scitotenv.2010.02.042

- Jany, C., & Zebracki, M. (2012). *METROC : Évaluation des apports de contaminants chimiques de la*
- 594 *métropole marseillaise au milieu marin*. https://archimer.ifremer.fr/doc/00116/22692/
- Jones, D. E., & Turner, A. (2010). Bioaccessibility and mobilisation of copper and zinc in estuarine
- 596 sediment contaminated by antifouling paint particles. *Estuarine, Coastal and Shelf Science,*
- 597 87(3), 399-404. https://doi.org/10.1016/j.ecss.2010.01.018

598	Jourda, C., Santini, S., Rocher, C., Le Bivic, A., & Claverie, JM. (2015). Draft Genome Sequence of an
599	Alphaproteobacterium Associated with the Mediterranean Sponge Oscarella lobularis.
600	Genome Announcements, 3(5). https://doi.org/10.1128/genomeA.00977-15
601	Keren, R., Mayzel, B., Lavy, A., Polishchuk, I., Levy, D., Fakra, S. C., Pokroy, B., & Ilan, M. (2017).
602	Sponge-associated bacteria mineralize arsenic and barium on intracellular vesicles. Nature
603	Communications, 8(1), Art. 1. https://doi.org/10.1038/ncomms14393
604	Khati, W., Nabila, B., Ouali, K., & Derbal, F. (2018). Sponges (Porifera) as bioindicator species of
605	environmental stress from de Gulf of Annaba (Algeria). Biodiversity Journal, 9, 319-324.
606	https://doi.org/10.31396/Biodiv.Jour.2018.9.4.319.324
607	Lafabrie, C., Pergent, G., Kantin, R., Pergent-Martini, C., & Gonzalez, JL. (2007). Trace metals
608	assessment in water, sediment, mussel and seagrass species—Validation of the use of
609	Posidonia oceanica as a metal biomonitor. Chemosphere, 68(11), 2033-2039.
610	https://doi.org/10.1016/j.chemosphere.2007.02.039
611	Lanquar, R. (2011). Tourism in the Mediterranean : Scenarios Up to 2030. SSRN Electronic Journal.
612	https://doi.org/10.2139/ssrn.2000717
613	Lasalle, L. (2007). Présence de plomb et d'arsenic sur le littoral Cellule interrégionale
614	d'épidémiologie Sud—Institut de veille sanitaire. https://www.libraires-
615	ensemble.com/livre/1718193-presence-de-plomb-et-d-arsenic-sur-le-littoralcellule-
616	interregionale-d-epidemiologie-sud-institut-de-veille-sanitaire
617	Lee, O. O., Wong, Y. H., & Qian, PY. (2009). Inter- and Intraspecific Variations of Bacterial
618	Communities Associated with Marine Sponges from San Juan Island, Washington. Applied
619	and Environmental Microbiology, 75(11), 3513-3521. https://doi.org/10.1128/AEM.00002-09
620	Liu, Q., Xu, X., Zeng, J., Shi, X., Liao, Y., Du, P., Tang, Y., Huang, W., Chen, Q., & Shou, L. (2019). Heavy
621	metal concentrations in commercial marine organisms from Xiangshan Bay, China, and the
622	potential health risks. Marine Pollution Bulletin, 141, 215-226.

623 https://doi.org/10.1016/j.marpolbul.2019.02.058

- Mahaut, M.-L., Basuyaux, O., Baudinière, E., Chataignier, C., Pain, J., & Caplat, C. (2013). The porifera
- 625 Hymeniacidon perlevis (Montagu, 1818) as a bioindicator for water quality monitoring.

626 Environmental Science and Pollution Research International, 20(5), 2984-2992.

- 627 https://doi.org/10.1007/s11356-012-1211-7
- 628 Maldonado, M. (2016). Sponge waste that fuels marine oligotrophic food webs : A re-assessment of
- 629 its origin and nature. *Marine Ecology*, 37(3), 477-491. https://doi.org/10.1111/maec.12256
- 630 Martin, C. S., Giannoulaki, M., De Leo, F., Scardi, M., Salomidi, M., Knittweis, L., Pace, M. L., Garofalo,
- 631 G., Gristina, M., Ballesteros, E., Bavestrello, G., Belluscio, A., Cebrian, E., Gerakaris, V.,
- 632 Pergent, G., Pergent-Martini, C., Schembri, P. J., Terribile, K., Rizzo, L., ... Fraschetti, S. (2014).
- 633 Coralligenous and maërl habitats : Predictive modelling to identify their spatial distributions
- 634 across the Mediterranean Sea. *Scientific Reports*, *4*(1), 5073.
- 635 https://doi.org/10.1038/srep05073
- 636 Martín, J., Sanchez-Cabeza, J. A., Eriksson, M., Levy, I., & Miquel, J. C. (2009). Recent accumulation of
- 637 trace metals in sediments at the DYFAMED site (Northwestern Mediterranean Sea). *Marine*
- 638 *Pollution Bulletin, 59*(4-7), 146-153. https://doi.org/10.1016/j.marpolbul.2009.03.013
- 639 Migon, C., Heimbürger-Boavida, L.-E., Dufour, A., Chiffoleau, J.-F., & Cossa, D. (2020). Temporal
- 640 variability of dissolved trace metals at the DYFAMED time-series station, Northwestern
- 641 Mediterranean. *Marine Chemistry*, 225, 103846.
- 642 https://doi.org/10.1016/j.marchem.2020.103846
- Milanese, M., Chelossi, E., Manconi, R., Sarà, A., Sidri, M., & Pronzato, R. (2003). The marine sponge
 Chondrilla nucula Schmidt, 1862 as an elective candidate for bioremediation in integrated
 aquaculture. *Biomolecular Engineering*, 20(4), 363-368. https://doi.org/10.1016/S1389-
- 646 0344(03)00052-2
- 647 Mille, T., Cresson, P., Chouvelon, T., Bustamante, P., Brach-Papa, C., Bruzac, S., Rozuel, E., &
- 648 Bouchoucha, M. (2018). Trace metal concentrations in the muscle of seven marine species :
- 649 Comparison between the Gulf of Lions (North-West Mediterranean Sea) and the Bay of

- 650 Biscay (North-East Atlantic Ocean). *Marine Pollution Bulletin*, 135, 9-16.
- 651 https://doi.org/10.1016/j.marpolbul.2018.05.051
- Millet, B., Pinazo, C., Banaru, D., Pagès, R., Guiart, P., & Pairaud, I. (2018). Unexpected spatial impact
- of treatment plant discharges induced by episodic hydrodynamic events : Modelling
- 654 Lagrangian transport of fine particles by Northern Current intrusions in the bays of Marseille
- 655 (France). *PLOS ONE*, *13*, e0195257. https://doi.org/10.1371/journal.pone.0195257
- 656 Morganti, T. M., Ribes, M., Yahel, G., & Coma, R. (2019). Size Is the Major Determinant of Pumping
- 657 Rates in Marine Sponges. *Frontiers in Physiology*, *10*, 1474.
- 658 https://doi.org/10.3389/fphys.2019.01474
- 659 Müller, W. E. G., Batel, R., Lacorn, M., Steinhart, H., Simat, T., Lauenroth, S., Hassanein, H., &
- 660 Schröder, H. C. (1998). Accumulation of cadmium and zinc in the marine sponge Suberites
- 661 domuncula and its potential consequences on single-strand breaks and on expression of
- 662 heat-shock protein : A natural field study. *Marine Ecology Progress Series*, 167, 127-135.
- 663 https://doi.org/10.3354/meps167127
- 664 Nogales, B., Lanfranconi, M. P., Piña-Villalonga, J. M., & Bosch, R. (2011). Anthropogenic
- 665 perturbations in marine microbial communities. *FEMS Microbiology Reviews*, 35(2), 275-298.
- 666 https://doi.org/10.1111/j.1574-6976.2010.00248.x
- Olesen, T. M. E., & Weeks, J. M. (1994). Accumulation of Cd by the marine sponge Halichondria
 panicea pallas : Effects upon filtration rate and its relevance for biomonitoring. *Bulletin of*
- 669 Environmental Contamination and Toxicology, 52(5), 722-728.
- 670 https://doi.org/10.1007/BF00195494
- 671 Orani, A. M., Barats, A., Vassileva, E., & Thomas, O. P. (2018). Marine sponges as a powerful tool for
- 672 trace elements biomonitoring studies in coastal environment. *Marine Pollution Bulletin*, 131,
- 673 633-645. https://doi.org/10.1016/j.marpolbul.2018.04.073
- Orani, A. M., Barats, A., Zitte, W., Morrow, C., & Thomas, O. P. (2018). Comparative study on the
- bioaccumulation and biotransformation of arsenic by some northeastern Atlantic and

- 676 northwestern Mediterranean sponges. *Chemosphere*, 201, 826-839.
- 677 https://doi.org/10.1016/j.chemosphere.2018.03.078
- 678 Orani, A. M., Vassileva, E., Azemard, S., & Thomas, O. P. (2020). Comparative study on Hg
- bioaccumulation and biotransformation in Mediterranean and Atlantic sponge species.
- 680 *Chemosphere, 260,* 127515. https://doi.org/10.1016/j.chemosphere.2020.127515
- 681 Oursel, B. (2013). Transferts et dynamique des contaminants métalliques en zone côtière. : Impact
- 682 *d'une grande agglomération méditerranéenne* (Numéro 2013TOUL0019) [Theses, Université
- 683 de Toulon]. https://tel.archives-ouvertes.fr/tel-01132521
- Padovan, A., Munksgaard, N., Alvarez, B., McGuinness, K., Parry, D., & Gibb, K. (2012). Trace metal
- 685 concentrations in the tropical sponge Spheciospongia vagabunda at a sewage outfall :
- 686 Synchrotron X-ray imaging reveals the micron-scale distribution of accumulated metals.
- 687 *Hydrobiologia*, *687*(1), 275-288. https://doi.org/10.1007/s10750-011-0916-9
- Pan, K., Lee, O. O., Qian, P.-Y., & Wang, W.-X. (2011). Sponges and sediments as monitoring tools of
 metal contamination in the eastern coast of the Red Sea, Saudi Arabia. *Marine Pollution*
- 690 Bulletin, 62(5), 1140-1146. https://doi.org/10.1016/j.marpolbul.2011.02.043
- Patel, B., Balani, M. C., & Patel, S. (1985). Sponge 'sentinel' of heavy metals. *Science of The Total*
- 692 Environment, 41(2), 143-152. https://doi.org/10.1016/0048-9697(85)90184-6
- 693 Penz, C., Simonsen, T., & Devries, P. (2011). A new Orobrassolis butterfly (Nymphalidae, Brassolini) :
- 694 A casualty of habitat destruction? *Zootaxa*, 2740, 35-43.
- 695 https://doi.org/10.11646/zootaxa.2740.1.3
- 696 Perez, T., Longet, D., Schembri, T., Rebouillon, P., & Vacelet, J. (2005). Effects of 12 years' operation
- 697 of a sewage treatment plant on trace metal occurrence within a Mediterranean commercial
- 698 sponge (Spongia officinalis, Demospongiae). *Marine Pollution Bulletin*, *50*(3), 301-309.
- 699 https://doi.org/10.1016/j.marpolbul.2004.11.001

- 700 Pérez, T., Vacelet, J., & Rebouillon, P. (2004). In situ comparative study of several Mediterranean
- sponges as potential biomonitors of heavy metals. *Sponge Science in the New Millennium*,
 68, 517-525.
- Rao, J. V., Kavitha, P., Reddy, N. C., & Rao, T. G. (2006). Petrosia testudinaria as a biomarker for metal
- contamination at Gulf of Mannar, southeast coast of India. *Chemosphere*, 65(4), 634-638.
- 705 https://doi.org/10.1016/j.chemosphere.2006.01.072
- 706 Renard, E., Rocher, C., Ereskovsky, A., & Borchiellini, C. (2021). The Homoscleromorph Sponge,
- 707 Oscarella lobularis. In Handbook of Marine Model Organisms in Experimental Biology (p.
- 708 79-100). CRC Press. https://doi.org/10.1201/9781003217503-5
- 709 Rocher, C., Vernale, A., Fierro-Constaín, L., Séjourné, N., Chenesseau, S., Marschal, C., Le Goff, E.,
- 710 Dutilleul, M., Matthews, C., Marschal, F., Brouilly, N., Massey-Harroche, D., Ereskovsky, A., Le
- 711 Bivic, A., Borchiellini, C., & Renard, E. (2020). The buds of Oscarella lobularis (Porifera) : A
- 712 new convenient model for sponge cell and developmental biology. *BioRxiv*,
- 713 2020.06.23.167296. https://doi.org/10.1101/2020.06.23.167296
- Rossi, N., & Jamet, J.-L. (2008). In situ heavy metals (copper, lead and cadmium) in different plankton
- 715 compartments and suspended particulate matter in two coupled Mediterranean coastal
- r16 ecosystems (Toulon Bay, France). *Marine Pollution Bulletin*, *56*(11), 1862-1870.
- 717 https://doi.org/10.1016/j.marpolbul.2008.07.018
- 718 Roussiez, V., Ludwig, W., Monaco, A., Probst, J.-L., Bouloubassi, J., Buscail, R., & Saragoni, G. (2006).
- 719 Sources and sinks of sediment-bound contaminants in the Gulf of Lions (NW Mediterranean
- Sea) : A multi-tracer approach. *Continental Shelf Research*, *2*(16), Art. 16.
- Roveta, C., Pica, D., Calcinai, B., Girolametti, F., Truzzi, C., Illuminati, S., Annibaldi, A., & Puce, S.
- 722 (2020). Hg Levels in Marine Porifera of Montecristo and Giglio Islands (Tuscan Archipelago,
- 723 Italy). *Applied Sciences*, 10, 4342. https://doi.org/10.3390/app10124342
- 724 Santos-Echeandía, J., Campillo, J. A., Egea, J. A., Guitart, C., González, C. J., Martínez-Gómez, C., León,
- 725 V. M., Rodríguez-Puente, C., & Benedicto, J. (2021). The influence of natural vs

726	anthropogenic factors on trace metal(loid) levels in the Mussel Watch programme : Two
727	decades of monitoring in the Spanish Mediterranean sea. Marine Environmental Research,
728	169, 105382. https://doi.org/10.1016/j.marenvres.2021.105382
729	Schenkelaars, Q., Fierro-Constain, L., Renard, E., Hill, A. L., & Borchiellini, C. (2015). Insights into
730	Frizzled evolution and new perspectives. <i>Evolution & Development</i> , 17(2), 160-169.
731	https://doi.org/10.1111/ede.12115
732	Schuhmacher, M., Batiste, J., Bosque, M. A., Domingo, J. L., & Corbella, J. (1994). Mercury
733	concentrations in marine species from the coastal area of Tarragona Province, Spain. Dietary
734	intake of mercury through fish and seafood consumption. The Science of the Total
735	Environment, 156(3), 269-273. https://doi.org/10.1016/0048-9697(94)90193-7
736	Selvin, J., Shanmugha Priya, S., Seghal Kiran, G., Thangavelu, T., & Sapna Bai, N. (2009). Sponge-
737	associated marine bacteria as indicators of heavy metal pollution. Microbiological Research,
738	164(3), 352-363. https://doi.org/10.1016/j.micres.2007.05.005
739	Tchounwou, P. B., Yedjou, C. G., Patlolla, A. K., & Sutton, D. J. (2012). Heavy Metal Toxicity and the
740	Environment. In A. Luch (Éd.), Molecular, Clinical and Environmental Toxicology : Volume 3 :
741	Environmental Toxicology (p. 133-164). Springer. https://doi.org/10.1007/978-3-7643-8340-
742	4_6
743	Tessier, E., Garnier, C., Mullot, JU., Lenoble, V., Arnaud, M., Raynaud, M., & Mounier, S. (2011).
744	Study of the spatial and historical distribution of sediment inorganic contamination in the
745	Toulon bay (France). Marine Pollution Bulletin, 62(10), 2075-2086.
746	https://doi.org/10.1016/j.marpolbul.2011.07.022
747	Tian, RM., Wang, Y., Bougouffa, S., Gao, ZM., Cai, L., Zhang, WP., Bajic, V., & Qian, PY. (2014).
748	Effect of Copper Treatment on the Composition and Function of the Bacterial Community in
749	the Sponge Haliclona cymaeformis. <i>MBio</i> , 5(6), e01980-14.
750	https://doi.org/10.1128/mBio.01980-14

- 751 Tumolo, M., Ancona, V., De Paola, D., Losacco, D., Campanale, C., Massarelli, C., & Uricchio, V. F.
- 752 (2020). Chromium Pollution in European Water, Sources, Health Risk, and Remediation
- 753 Strategies : An Overview. International Journal of Environmental Research and Public Health,
- 754 *17*(15), Art. 15. https://doi.org/10.3390/ijerph17155438
- 755 Urien, N., Farfarana, A., Uher, E., Fechner, L. C., Chaumot, A., Geffard, O., & Lebrun, J. D. (2017).
- 756 Comparison in waterborne Cu, Ni and Pb bioaccumulation kinetics between different
- 757 gammarid species and populations : Natural variability and influence of metal exposure
- 758 history. *Aquatic Toxicology*, *193*, 245-255. https://doi.org/10.1016/j.aquatox.2017.10.016
- 759 Vernale, A., Prünster, M. M., Marchianò, F., Debost, H., Brouilly, N., Rocher, C., Massey-Harroche, D.,
- 760 Renard, E., Bivic, A. L., Habermann, B. H., & Borchiellini, C. (2021). *Evolution of mechanisms*
- 761 controlling epithelial morphogenesis across animals : New insights from dissociation -
- 762 reaggregation experiments in the sponge Oscarella lobularis (p. 2021.03.22.436370).
- 763 https://doi.org/10.1101/2021.03.22.436370
- 764 Vieira, K. S., Crapez, M. A. C., Lima, L. S., Delgado, J. F., Brito, E. B. C. C., Fonseca, E. M., Baptista Neto,
- 765 J. A., & Aguiar, V. M. C. (2021). Evaluation of bioavailability of trace metals through
- bioindicators in a urbanized estuarine system in southeast Brazil. *Environmental Monitoring*
- 767 *and Assessment*, 193(1), 18. https://doi.org/10.1007/s10661-020-08809-x
- 768 Weisz, J. B., Lindquist, N., & Martens, C. S. (2008). Do associated microbial abundances impact
- 769 marine demosponge pumping rates and tissue densities? *Oecologia*, *155*(2), 367-376.
- 770 https://doi.org/10.1007/s00442-007-0910-0
- Yamaoka, Y., Carmona, M. L., Oclarit, J. M., Jin, K., & Shibata, Y. (2001). Arsenic compounds in marine
 sponge (Haliclona permolis, Halichondria japonica, Halichondria okadai and Haliclona sp.
- 773 White) from Seto Inland Sea, Japan. *Applied Organometallic Chemistry*, 15(4), 261-265.
- 774 https://doi.org/10.1002/aoc.135

- Zhang, G., Liu, D., Wu, H., Chen, L., & Han, Q. (2012). Heavy metal contamination in the marine
- 776 organisms in Yantai coast, northern Yellow Sea of China. *Ecotoxicology*, *21*(6), 1726-1733.

777 https://doi.org/10.1007/s10646-012-0958-4

- Zhou, Q., Zhang, J., Fu, J., Shi, J., & Jiang, G. (2008). Biomonitoring : An appealing tool for assessment
- of metal pollution in the aquatic ecosystem. *Analytica Chimica Acta*, 606(2), 135-150.
- 780 https://doi.org/10.1016/j.aca.2007.11.018
- 781 Zhu, G., Noman, M. A., Narale, D. D., Feng, W., Pujari, L., & Sun, J. (2020). Evaluation of ecosystem
- 782 health and potential human health hazards in the Hangzhou Bay and Qiantang Estuary region
- through multiple assessment approaches. *Environmental Pollution*, *264*, 114791.
- 784 https://doi.org/10.1016/j.envpol.2020.114791
- 785
- 786

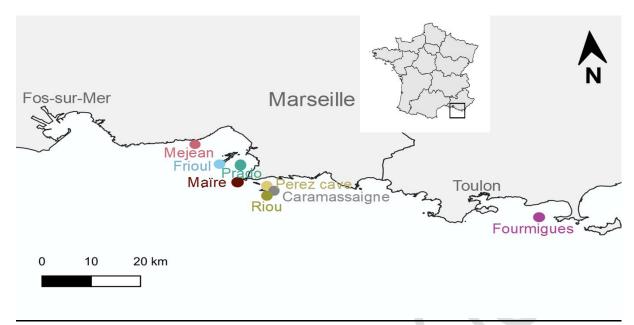




Figure 1: Location of the eight sampling sites (south of France; North-Western Mediterranean Sea).

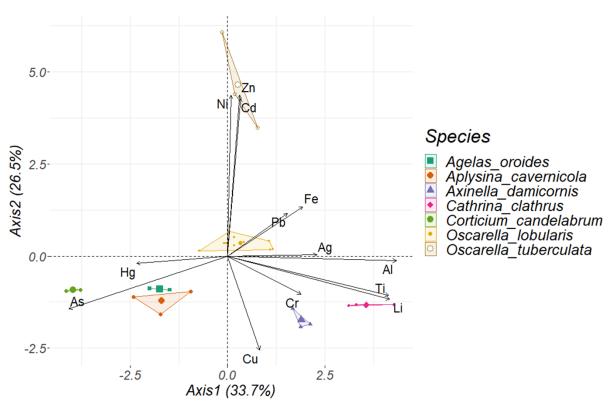
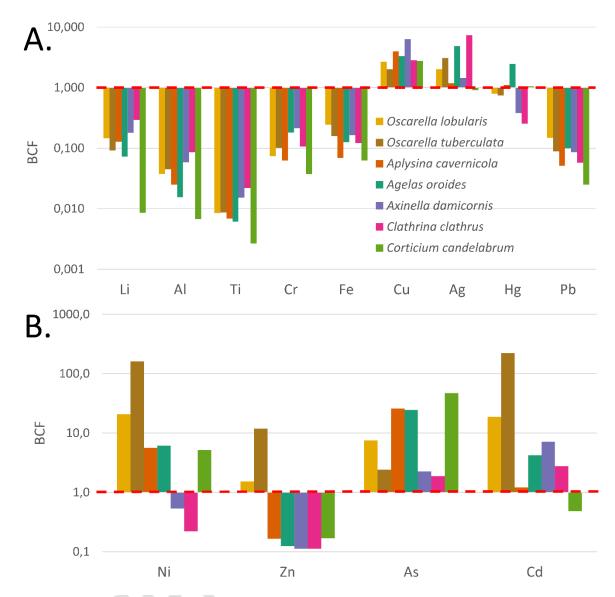




Figure 2: Principal component analysis (PCA) of metal concentrations in seven distinct marine sponge
 species collected in the Maïre site. Convex ellipses group heavy metal concentrations from the seven
 species.



- Figure 3: Bioconcentration factors calculated for all metals in seven marine sponges. BCFs of (A) Li, Al,
- Ti, Cr, Fe, Cu, Ag, Hg, Pb and (B) Ni, Zn, As, Cd. Values are available in table A.6.

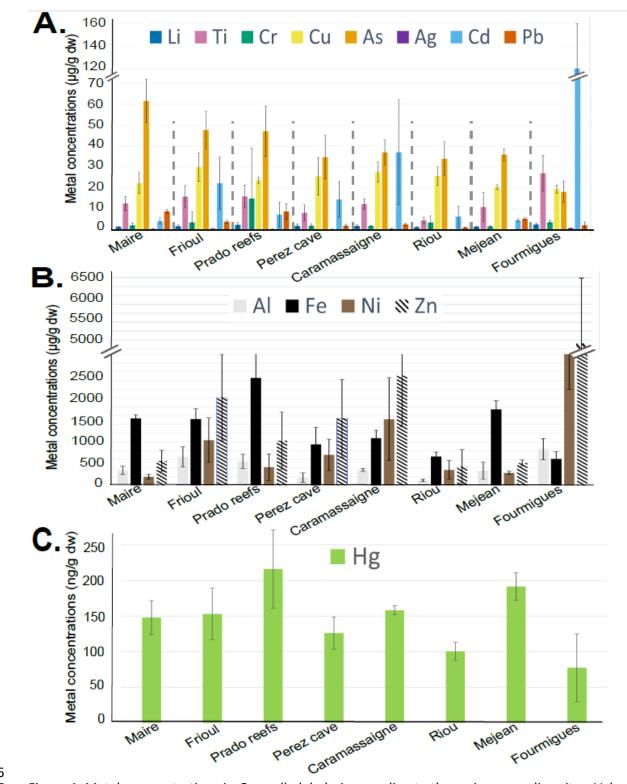




Figure 4: Metals concentrations in *Oscarella lobularis* according to the various sampling sites. Values
are means ± SD. Concentrations of (A) Ag, As, Cd, Cr, Cu, Li, Pb, Ti, (B) Al, Fe, Ni, Zn and (C) Hg were
measured in tissues.

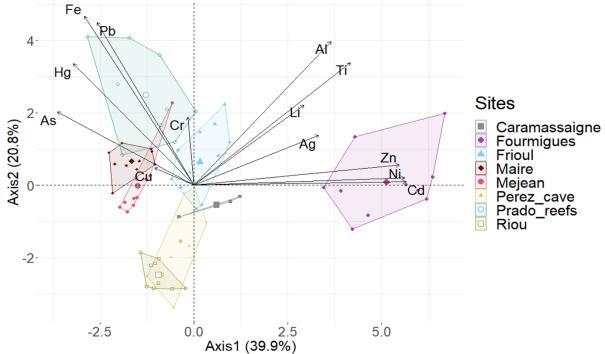
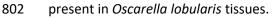




Figure 5: Principal component analysis (PCA) plot describing inter-site variation of metal contents



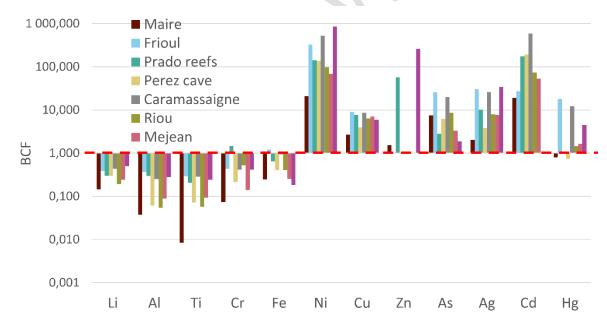


Figure 6: Bioconcentration factors (BCF) calculated for all metals in *Oscarella lobularis* individuals sampled at different sites (using metal concentrations measured in sediments as reference). Values are available in table A.6.

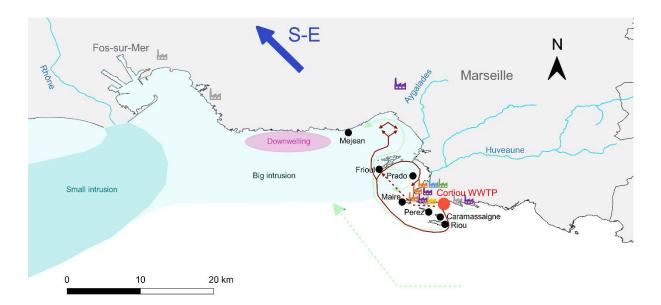


Figure 7: Hydrodynamics in the bay of Marseille in presence of south-east winds. Including the North Current intrusion (dotted green arrow for the surface current and green arrow for the subsurface), the downwelling formation and the intrusions of the Rhône River. Red arrows indicate the subsurface particles transported from the WWTP area (10-30 m depth) and red point indicates the WWTP outlet. The factory patterns represent the former lead factories established in the bay of Marseille in the 19th century. Figure adapted (Arnaud et al., 2021; Fraysse et al., 2014; Millet et al., 2018) with lead and silver factories in orange; glass factories in blue; oil factories in grey; tartaric acid factories in green; sulfur factories in yellow; and soda factories in purple.

- - -

Appendix A. Supplementary material

Sites	GPS coordinates	Depth (m)	Number of individuals	Date of sampling
Maïre Island	43°12.760'N, 5°19.800'E	12	12	02/2019 (Oscarellidae) ; 01/2021 (othe sponges phyla)
Prado's Reef	43°15.745'N, 5°20.360'E	27	10	04/2019 ; 01/2020
Frioul	43°15.703'N, 5°17.273'E	8	13	04/2019 ; 02/2020
Riou	43° 10.362'N, 5° 23.432'E	20	10	07/2020
Perez Cave	43°11.190'N, 5°23.470'E	13	8	01/2020 ; 05/2020
Caramassaigne	43°10.592'N, 5°23.621'E	13.5	3	01/2020 ; 02/2021
Mejean	43° 19.688'N, 5° 13.338'E	25-30	10	07/2020
Fourmigues	43°02.3630'N, 06°04.1796'E	30	14	01/2021

Supplemental Table 1. Number of sponge adult individuals collected at different sites and depths.

Supplemental Table 2. Limit of quantification (LOQ) in μ g/L for trace metal elements measured by ICP-MS and in ng for mercury measured with AMA-254.

						LOQ						
Li	Al	Ti	Cr	Fe	Ni	Cu	Zn	As	Ag	Cd	Hg	Pb
0,003	0,15	0,05	0,009	0,3	0,006	0,014	0,05	0,037	0,002	0,017	0,003	0,005
μg/L	μg/L	μg/L	μg/L	μg/L	μg/L	μg/L	μg/L	μg/L	μg/L	μg/L	ng	μg/L

837	7
-----	---

- **Supplemental Table 3.** Samples measured with AMA-254 and ICP-MS to determine the metal
- 844 concentration at the various sites. Each sample was replicated before measurement.

Type of sample	Site		ICP-MS		AMA-254
		No. Individuals	No. technical replicates	No. individuals	No. technical replicates
Oscarella lobularis Oscarella tuberculata Aplysina cavernicola Agelas oroides Axinella damicornis Cathrina clathrus	Maire	11	3	12	1-5
	Frioul	12	3	13	1-2
	Prado's Reef	9	3	10	2
Occarolla lobulario	Perez Cave	7	3	8	2
Oscurena iobularis	Caramassaigne	3	3	3	1-2
	Riou	10	3	10	2
	Mejean	10	3	10	2
	Fourmigues	10	3	10	1-2
Oscarella tuberculata	Maire	3	3	3	1
Aplysina cavernicola	Maire	3	3	3	1
Agelas oroides	Maire	2	3	2	1
Axinella damicornis	Maire	3	3	3	1
Cathrina clathrus	Maire	3	3	3	1
Corticium candelabrum	Maire	3	3	3	1
	Maire		3		1
	Frioul		3		3
	Prado's Reef		3		2
Sodimont	Perez Cave		3		3
seament	Caramassaigne		3		3
	Riou		3		2
	Mejean		3		2
	Fourmigues		3		3

Supplemental Table 4. Relative standard deviation (RSD) in seven sponges' species collected at the
 Maïre site. Values for each heavy metal are expressed in %.

						RSD (%	%)						
Sponge species (Maïre site)	Li	Al	Ti	Cr	Fe	Ni	Cu	Zn	As	Ag	Cd	Hg	Pb
O.lobularis	8,9	26,8	27,2	54,8	5,4	26,1	22,5	42,7	16,8	68,5	36,5	16,1	9,9
O.tuberculata	8,5	10,7	27,8	88,7	12,3	35,0	14,1	20,9	9,5	40,1	13,0	10,6	18,1
A.cavernicola	21,4	39,9	36,8	58,4	19,6	12,8	28,8	9,9	10,3	8,7	8,8	4,5	47,0
A.oroides	11,0	7,1	24,4	61,6	41,3	24,4	13,6	18,6	15,0	46,3	15,8	76,3	60,4
A.damicornis	9,4	4,7	6,1	36,1	8,0	44,3	14,9	3,0	5,5	4,0	6,8	7,1	27,6
C.clathrus	3,1	16,6	11,7	11,3	11,0	16,4	12,1	3,6	34,9	32,2	19,7	5,8	27,3
C.candelabrum	62,2	30,1	23,6	37,4	14,8	11,9	1,4	1,4	3,3	12,0	19,0	9,5	24,8

- 854 Supplemental Table 5. Concentration of metals (µg/g of dry weight) in seven sponges' species and on
- surface sediment collected in the different sites. Values are means \pm SD. Different letters indicate a
- significant difference between sites for *O. lobularis* samples (Kruskall-Wallis test followed by a Dunn
- 857 post hoc test; p < 0,05)

Complex	Sites		A	ті	Cr	Fe	Ni	Cu	Zn	As	4.5	Cd	Hg	Pb
Samples		-									Ag		-	
Oscarella lobularis	Maire	1,32 ± 0,12 ^(sb)	349 ± 93 ^(ab)	12,5 ± 3,4 ^(abc)			201 ± 53 ^(a)	22,2 ± 5 ^(ab)	564 ± 241 ^(ab)	61,5 ± 10,3 ^(a)		4,1 ± 1,5 ^(*)	0,148 ± 0,024 ^(*)	8,76 ± 0,87 ^(*)
Oscarella lobularis	Frioul	1,62 ± 0,69 ^(ab)	655 ± 232 ^(cd)	15,8 ± 5,2 ^(*)	3,46 ± 5,09 ^(ab)	1518 ± 237 ^(ab)	1040 ± 505 ^(bc)	29,9 ± 6,8 ^(c)	2011 ± 987 ^(cd)	47,6 ± 9 ^(ab)	0,41 ± 0,3 ^(ac)	22,2 ± 12,5 ^(bc)	0,153 ± 0,036 ^(*)	3,79 ± 0,73 ^(bc)
Oscarella lobularis	Prado reefs	2,3 ± 1,14 ^(ac)	552 ± 162 ^(acd)	15,8 ± 5,5 ^(abd)	14,8 ± 24,3 ^(c)	2460 ± 912 ^(c)	422 ± 305 (ade)	23,6 ± 1,5 ^(ac)	1026 ± 650 (abc)	47,1 ± 12 ^(abc)	0,28 ± 0,2 ^(b)	7,23 ± 5,9 ^(ade)	0,216 ± 0,055 ^(b)	8,69 ± 3,75 ^(a)
Oscarella lobularis	Perez cave	1,76 ± 1,09 ^(ab)	182 ± 108 (be)	8,05 ± 3,76 ^(ce)	1,87 ± 1,02 ^(a)	943 ± 389 ^(d)	703 ± 358 ^(bde)	25,5±9 ^(*c)	1537 ± 888 ^(ac)	34,6 ± 10,4 ^(cd)	0,21 ± 0,1 ^(b)	14,4 ± 8,6 ^(bde)	0,126 ± 0,023 ^(ac)	1,82 ± 0,71 ^(bd)
Oscarella lobularis	Caramassaigne	1,73 ± 0,7 ^(abc)	359 ± 33 ^(abce)	12,2 ± 2,5 (abcd)	1,75 ± 0,29 ^(ab)	1085 ± 179 ^(ad)	1518 ± 946 ^(bcd)	27,5 ± 4,9 ^(ac)	2508 ± 1165 (cd)	37 ± 6 ^(bcde)	0,38 ± 0,3 ^(abc)	37 ± 25 ^(bcd)	0,158 ± 0,006 ^(ab)	2,51 ± 0,67 ^(bcd)
Oscarella lobularis	Riou	1,18 ± 0,26 ^(b)	121 ± 30 ^(e)	4,4 ± 1,53 ^(e)	3,41 ± 3,06 ^(a)	666 ± 101 ^(d)		25,6 ± 4,4 ^(ac)	433 ± 389 ^(b)	33,8 ± 8,1 ^(d)	0,18 ± 0,04 ^(b)	6,22 ± 5 ^(se)	0,101 ± 0,013 ^(c)	1,02 ± 0,28 ^(d)
Oscarella lobularis	Mejean	1,38 ± 0,3 ^(eb)	344 ± 189 ^(sb)	10,8 ± 7,1 ^(bc)	1,54 ± 0,47 ^(*)	1740 ± 197 ^(bc)	294 ± 37 ^(ee)	20,3 ± 1,1 ^(b)	516 ± 68 ^(b)	35,9 ± 2,9 ^(d)	0,1 ± 0,01 ^(d)	4,44 ± 0,74 ^(se)	0,191 ± 0,019 ^(b)	5,21 ± 0,51 ^(ec)
Oscarella lobularis	Fourmigues	2,45 ± 0,6 ^(c)	834 ± 243 ^(d)	27 ± 8,6 ^(d)	3,48 ± 0,92 ^(bc)	610 ± 160 ^(d)	3277 ± 1084 ^(c)	19,3 ± 2 ^(b)	4920 ± 1467 ^(d)	18,1 ± 5,1 ^(e)	0,68 ± 0,1 ^(c)	120 ± 39 ^(c)	0,0778 ± 0,0476 ^(c)	2,14 ± 1,55 ^(bd)
Oscarella tuberculata	Maire	0,833 ± 0,071	418 ± 45	12,8 ± 3,6	2,8 ± 2,48	996 ± 122	1564 ± 548	16,7 ± 2,4	4394 ± 918	19,9 ± 1,9	0,546 ± 0,219	48,5 ± 6,3	0,138 ± 0,015	5,23 ± 0,95
Aplysina cavernicola	Maire	1,16 ± 0,25	231 ± 92	10,1 ± 3,7	1,74 ± 1,01	433 ± 85	54,4 ± 6,9	32,8 ± 9,4	61,3 ± 6,1	211 ± 22	0,209 ± 0,018	0,264 ± 0,023	0,207 ± 0,009	3 ± 1,41
Agelas oroides	Maire	0,657 ± 0,073	143 ± 10	9,05 ± 2,21	5,01 ± 3,09	786 ± 325	59,5 ± 14,5	27,6 ± 3,8	46,5 ± 8,6	200 ± 30	0,86 ± 0,398	0,921 ± 0,146	0,46 ± 0,351	5,86 ± 3,54
Axinella damicornis	Maire	1,64 ± 0,15	541 ± 25	22,6 ± 1,4	5,92 ± 2,14	1032 ± 82	5,2 ± 2,31	52,2 ± 7,8	41,7 ± 1,2	18,5 ± 1	0,257 ± 0,01	1,55 ± 0,11	0,0707 ± 0,005	5,05 ± 1,39
Cathrina clathrus	Maire	2,67 ± 0,08	795 ± 132	32,7 ± 3,8	2,96 ± 0,33	759 ± 84	2,13 ± 0,35	23,4 ± 2,8	42 ± 1,5	15,5 ± 5,4	1,31 ± 0,4	0,602 ± 0,118	0,0473 ± 0,0028	3,39 ± 0,93
Corticium candelabrum	Maire	0,0783 ± 0,0485	63,2 ± 19	3,93 ± 0,93	1,04 ± 0,39	393 ± 58	50,3 ± 6	22,9 ± 0,3	62,4 ± 0,9	387 ± 13	0,162 ± 0,019	0,105 ± 0,02	0,197 ± 0,019	1,47 ± 0,36
Sediment	Maire	9,10 ± 4,6%	9275 ± 2%	1480 ± 2%	27,7±3,5%	6262 ± 2,6%	9.67 ± 3%	8,29 ± 9,6%	371 ± 8,3%	8,19 ± 26%	0 178 + 3.8%	0,217 ± 146,8%	0,18601539	58,8 ± 1,3%
Sediment	Frioul	$4.2 \pm 2.1\%$	1762 ± 5.6%	54.1 ± 5.1%	7.99 ± 0.9%	1250 ± 2.1%	3.18 ± 3%	3.33 ± 1%	0	1.86 ± 11%	0.0135 ± 7.8%		0.0085 ± 11%	19.1 ± 1.5%
Sediment	Prado reefs	7.67 ± 3%	1872 ± 5,7%	76.7 ± 2%	10.1 ± 3.4%	3838 ± 6,1%	3 ± 3.8%	3.07 ± 6%	18.1 ± 3.5%			0.0410 ± 7.1%	0.227 ± 10.59%	20.8 ± 5.8%
Sediment	Perez cave	5.92 ± 2.2%	2959 ± 3.2%	113 ± 19.2%	8.63 ± 0.7%	2347 ± 2.8%	5.15 ± 0.3%	6.47 ± 2.8%	0			0.0746 ± 11.7%	0.172 ± 14%	20.6 ± 0.8%
Sediment	Caramassaigne	3.97 ± 0%	1431 ± 8%	42.8 ± 2.4%	4.20 ± 4.3%	1156 ± 1%	2.91 ± 0.8%	3.18 ± 3%	0	1,86 ± 3,7%		0.0633 ± 17.2%	0.013 ± 18%	10.6 ± 1.1%
Sediment	Riou	6,21 ± 2%	2241 ± 8%	76,6 ± 17%	6.55 ± 1.9%	1640 ± 1,3%	3,66 ± 0,3%	4.02 ± 4%	0	3,96 ± 4,3%	0,0140 ± 1,5%		0,0697 ± 3%	12,3 ± 0,9%
Sediment	Mejean	5.66 ± 1.4%	3871 ± 3%	115 ± 4.3%	11.0 ± 1.4%	6866 ± 1.1%	4.30 ± 2.3%	4,02 ± 4/8 2.88 ± 4.7%	0	10.8 ± 4,3%		0.0832 ± 40%	0.116 ± 21%	18 ± 2.3%
Sediment	Fourmigues	9,10 ± 13%	3008 ± 3,2%	115 ± 4,5% 111 ± 1,3%	8,32 ± 1,7%	3336 ± 1,1%	4,50 ± 2,5% 3,86 ± 11,1%	2,00 ± 4,7% 3,23 ± 3,6%	19 ± 4,5%	10,8 ± 4,1% 9,67 ± 1,5%	0,0131 ± 8,9%	0,0854 ± 0,9%	0,0172 ± 19,31%	10 ± 2,5% 11,3 ± 4,3%
	-													

860 Supplemental Table 6. Average bioconcentration factors (BCF) calculated for seven sponge species.

- 861 Values were obtained for each metal in Oscarella lobularis collected at eight sites and in six other
- sponge species collected at the Maïre site.

					BCF									
Samples	Sites	Li	AI	Tì	Cr	Fe	Ni	Cu	Zn	As	Ag	Cd	Hg	Pb
	Maire	0,1	0,0	0,0	0,1	0,2	20,8	2,7	1,5	7,5	2,0	18,9	0,8	0,1
	Frioul	0,4	0,4	0,3	0,4	1,2	326,7	9,0		25,7	30,3	27,3	18,0	0,2
	Prado reefs	0,3	0,3	0,2	1,5	0,6	140,7	7,7	56,7	2,8	10,1	176,3	1,0	0,4
O.lobularis	Perez cave	0,3	0,1	0,1	0,2	0,4	136,6	3,9		6,2	3,8	193,3	0,7	0,1
0.10bulul is	Caramassaigne	0,4	0,3	0,3	0,4	0,9	522,0	8,6		19,9	26,0	584,1	12,1	0,2
	Riou	0,2	0,1	0,1	0,5	0,4	98,0	6,4		8,5	7,9	73,0	1,4	0,1
	Mejean	0,2	0,1	0,1	0,1	0,3	68,3	7,1		3,3	7,6	53,3	1,7	0,3
	Fourmigues	0,5	0,3	0,2	0,4	0,2	849,9	6,0	259,3	1,9	34,0		4,5	0,2
O.tuberculata	Maire	0,1	0,0	0,0	0,1	0,2	161,8	2,0	11,9	2,4	3,1	223,4	0,7	0,1
A.cavernicola	Maire	0,1	0,0	0,0	0,1	0,1	5,6	4,0	0,2	25,8	1,2	1,2	1,1	0,1
A.oroides	Maire	0,1	0,0	0,0	0,2	0,1	6,2	3,3	0,1	24,5	4,8	4,2	2,5	0,1
A.damicornis	Maire	0,2	0,1	0,0	0,2	0,2	0,5	6,3	0,1	2,3	1,4	7,2	0,4	0,1
C.clathrus	Maire	0,3	0,1	0,0	0,1	0,1	0,2	2,8	0,1	1,9	7,4	2,8	0,3	0,1
C. candelabrum	Maire		0,0	47,2	0,0	0,1	5,2	2,8	0,0		0,9	0,5	0,2	1,1