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

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Abstract

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 aim of the study was to compare the metal concentrations (Li, Al, Ti, Cr, Fe, Ni, Cu, Zn, As, Ag, Cd, Hg, Pb) in seven marine sponges with a particular interest in the homoscleromorph sponge *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*, a multi-site analysis shows different bioaccumulation between the eight sampled populations. This inter-site differences may reflect differences in the hydrodynamic features and in past and present industrial activities. Because *Oscarella lobularis* shows a homogeneous metal accumulation pattern in comparison with the other tested species, it appears to be suitable for metal contamination biomonitoring in Mediterranean coastal waters, in particular of the coralligenous communities.

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

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1.Introduction

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

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

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the marine environment are bivalves (Aly et al., 2013; Azizi et al., 2018; Beyer et al., 2017; Gentric, 2016; Lafabrie et al., 2007; Pérez et al., 2004). Considering the ongoing major biodiversity crisis (Ceballos et al., 2015, 2017; Eisenhauer et al., 2019), it is timely to extend our knowledge concerning the impact of anthropic activities on ecosystem contamination by studying a larger set of marine animals (Chifflet et al., 2022). To this purpose, several studies 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; Müller et al., 1998; Patel et al., 1985; Perez et al., 2005; Pérez et al., 2004). We propose to evaluate for the first time the relevance of a Mediterranean sponge belonging to Homoscleromorpha, *Oscarella lobularis* (Schmidt, 1862), as bioindicator for metal 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 than 12% of marine fauna diversity (the same order as vertebrates) and are one of the main organisms within benthic communities (Coll et al., 2010). The large amounts of water filtered by sponges through their aquiferous system (Morganti et al., 2019; Weisz et al., 2008) yields to the accumulation of metals present both in the dissolved and particulate phases (Caplat et al., 2011; Orani et al., 2020; Rao et al., 2006). Several sponge species have already been used as bioindicators of metal contamination (Keren et al., 2017; Khati et al., 2018; Mahaut et al., 2013; Pan et al., 2011). However, only few studies explored if the accumulation of metals 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 of environmental metal contaminants. The number of sponge species for which a convenient set of biological knowledge and experimental tools are available is still limited, making it thus

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

In this context, *Oscarella lobularis* (Schmidt, 1862) provides several advantages: i) being abundant in shallow rocky substrates of the north-western Mediterranean Sea, including at the vicinity of the Marseille Metropole and the French Riviera (Ereskovsky et al., 2009; Renard et al., 2021). *Oscarella lobularis* is part of the coralligenous biocenosis which represents a 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 cycle, development and morphoanatomical organization were deeply studied (Ereskovsky et 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 al., 2021; Rocher et al., 2020) provide useful replicates for experimental ecotoxicological 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 species (Borchiellini et al., 2021; Fierro-Constain et al., 2017; Fierro-Constaín et al., 2021; Renard et al., 2021; Rocher et al., 2020). This is convenient and suitable to perform ecotoxicological experiments at different complementary levels of the biological scale (from 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

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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.

2. Materials and Methods

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).

Figure 1

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

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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 used to quantify Li, Al, Ti, Cr, Fe, Ni, Cu, Zn, As, Ag, Cd, and Pb (Supplementary Data Table 2 and 3). Before measurements, 300 mg (dry weight) of each sponge sample were digested (1.5 mL HCl 37% + 1.5 mL HNO_3 69% + 0.5 mL H_20_2 30%, quality TMA (Trace Metal Analysis)) and 50 mg (dry weight) of sediment were digested (1.5 mL HCl 37% + 1.5mL HNO_3 69% + 0.5 mL HF 40%, quality TMA (Trace Metal Analysis)). The samples were then mineralized with a 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 by ICP-MS. Procedural blank and a standard sample (corn powder V463, INRAE) were analyzed for each series of mineralization (Fehlauer et al., 2022).

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).

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153 **2.3. Data analyses**

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

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

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3. Results

3.1. Difference in metal contents between Porifera species

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.

3.1.2. Interspecific variability.

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

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186 strongly concentrated in the homoscleromorph *Oscarella uberculate* (1564 µg/g, 48.5 µg/g,
187 4394 µg/g, respectively.

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

193 The bioconcentration factor (BCF) (Supplementary Data Table 6) determines the accumulation
194 capacity of organisms in their tissues (Pan et al., 2011).

195 The comparison of the bioconcentration factors between the seven sponges shows that the
196 bioaccumulated metals are often similar (e.g., Cu and As show BCF>1 in all species; Ag and Cd
197 show BCF>1 in all except one species, see Supplementary Data Table 6). In agreement with
198 PCA analysis (Figure 2), figure 3 shows that some metals are much more highly accumulated
199 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 µg/g dw), Al (834 µg/g dw), Ni (3277 µg/g dw) and Cd (120 µg/g
207 dw), 11 times higher for Zn than those sampled at Riou, in which the sampled specimens

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present the lower Zn content. Sponges from the Prado reefs display the highest Fe body content with 2460 µg/g (4 times higher than sponges from Fourmigues) and the Maïre site individuals show the highest As concentration (61.5 µg/g dw), 3 times higher than in the ones from Fourmigues (Figure 4A). For the other metals, the variations between sites are slighter.

Figure 4

A PCA was performed to analyze inter-site variations. The first two axes of the PCA explain 60.7% of the total variability of the data (Figure 5). The PCA plot shows two distinct clusters: 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 Ni in the tissues of *Oscarella lobularis*. In addition, body contents in Zn, Ni and Cd are 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 mainly driven by the concentrations of Al, Ti, Fe, Hg and Pb. Sponges from the Prado reef site 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 concentrations (2460 ± 912 µg/g and 1740 ± 197 µg /g respectively) (Supplementary Data Table 5). The Frioul site is at the center of the PCA plot, this means that individuals display more homogeneous metal contents than those of the other sites. The population *of O. lobularis* from the Perez cave, Caramassaigne and Riou sites, located at the bottom of the biplot, show lower concentrations of metals than those of the other sites located in front of the urban coast (Figure 5).

Figure 5

3.2.2. Bioconcentration factors in *Oscarella lobularis.*

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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.

Figure 6

4. Discussion

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 (characterized by Al_2O_3 and iron oxides) formerly exploited, and many layers of *terra rossa* (i.e., iron oxides). Even though these rocks naturally contain metals, because the concerned area is characterized by the dominance of calcareous sedimentary rocks, the basicity of these 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., 2013; Tchounwou et al., 2012; Urien et al., 2017). Anthropic rock mining can nevertheless increase ground leaching and bring natural metals to the sea by runoff, in addition to industrial activities commonly considered to be responsible for most metal contaminations in the area.

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

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

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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 2; As and Hg for *C. candelabrum*; Cr, Ti, Li and Cu for *A. damicornis* and *C. clathrus*; Ni, Cd and Zn for *O.tuberculata*). In contrast *Oscarella lobularis* displays a more homogeneous metal accumulation pattern than the other sponge species studied. High inter-specific differences in metal bioaccumulation were already observed in previous studies performed on different sets of sponge species from different geographic areas (Batista et al., 2014; Genta-Jouve et al., 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., 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 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 al., 2012), by iii) different food preference (Maldonado, 2016; Perez et al., 2005), or by iv) differences in associated microorganisms (Keren et al., 2017; Lee et al., 2009; Selvin et al., 2009; Tian et al., 2014). In our case, the huge differences observed between *O.lobularis* and *O.tuberculata* BCFs (Supplementary Data Table 6) cannot be explained neither by molecular, 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.,

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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.

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

Although *Oscarella lobularis* accumulates metals more homogeneously than other sponge species, we note a variability of metal contents in tissues between the different sampling sites (Figure 5). These differences may be related to the past and present industrial activities in the bay of Marseille. The industrialization of Marseille increased in the middle of the 19th century with the creation of factories of several types (glass, petroleum, soda, tartaric acid, etc.). No obvious direct link has been made between metal contaminations in the bay and the factories, 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 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, was previously characterized (Lasalle, 2007), and high concentrations of Pb and As in soils (633 µg/g and 29.4 µg/g respectively), in marine sediments (99 to 1189 µg/g and 8.2 to 648 µg/g) and in marine organisms were highlighted. For example, in mussels, Pb and As are respectively up to 4 and 2 times higher in comparison with mussels from other sites of the Mediterranean sea (Andral et al., 2004; Santos-Echeandía et al., 2021), with highest Pb and As body amounts ranging from 4.5 to 5.3 µg/g dw and from 25.4 to 26.2 µg/g dw, respectively. Interestingly, *O.*

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lobularis specimens sampled in the Maïre site, located in the vicinity of this former industrial site, show the highest concentrations of Pb (8.76 ± 0.87 µg/g dw) and As (61.5 ± 10.3 µg/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 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, Cu, Pb, Cr, Hg and Fe at the Cortiou Cove during dry conditions (Millet et al., 2018; Oursel, 2013; Tumolo et al., 2020) and into the autonomous harbour ("*Port autonome*") of Marseille, the old harbour ("*vieux port*") and at the level of the Prado site, without prior passage through the treatment plant During rainy conditions. Finally, the small coastal rivers such as the 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 contaminants toward the bay (i.e., Hg up to 62 ng/L in the Huveaune and Aygalades) (Jany & 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 on certain sites in the Bay of Marseille. In accordance with previous studies on sponges of the Marseille region (Perez et al., 2005), our results indicate for example that the populations present in the bay (i.e., Prado) have significant quantities of Fe. These large quantities of Fe 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 with the North Current toward the bay (Figure 7). Fe isotope studies would confirm or refute

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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.

Figure 7

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 and Cd (Figure 5). The Toulon Bay, located at about 10 km from the Fourmigues sampling site, is characterized by important industrial, touristic, and military nautical activities, and is submitted to significant inputs of metals. In a previous study, high Zn concentrations with a range of 24.3 – 1340 µg/g dw were found in sediments in the large bay of Toulon (Tessier et 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 nautical activity of this area (Jones & Turner, 2010). In addition, the bay of Toulon was also reported to present high Cd concentration in seawater (between 0.007 and 0.14 µg/L) (Rossi & 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 maritime activities of the port (Augier, 1980). Therefore, our data at the Fourmigues site 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

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

4.3. *Oscarella lobularis* as a valuable bioindicator for metallic contamination

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 candidate as a bioindicator of metallic pollution in Mediterranean Sea. The choice of sentinel or bioindicator species should be guided by several criteria: i) preferring sessile species or species displaying limited dispersion (Azizi et al., 2018; Vieira et al., 2021), ii) opting for filter feeders (Batista et al., 2014; Milanese et al., 2003), iii) identifying, capturing and collecting easily a species with a sufficient population size to study spatial variations of ecosystems (Berthet, 2013). *Oscarella lobularis* fits all these criteria. Even if, several sponges have been proposed as bioindicator species (mostly Demospongiae but also one species of Calcarea *Paraleucilla magna*) (Batista et al., 2014), as far as we know, only one species (*Haliclona fulva* (Topsent, 1893)) has been showed as a generalist species in the Mediterranean Sea (Orani, Barats, Vassileva, et al., 2018), which appears to be a crucial criterion for choosing a sentinel organism.

Conclusions

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.

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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.

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ACCEPTED VERSION

References:

Aly, W., Williams, I. D., & Hudson, M. D. (2013). Metal contamination in water, sediment and biota from a semi-enclosed coastal area. *Environmental Monitoring and Assessment*, *185*(5), 3879-3895. https://doi.org/10.1007/s10661-012-2837-0

Andral, B., Stanisiere, J. Y., Sauzade, D., Damier, E., Thebault, H., Galgani, F., & Boissery, P. (2004). Monitoring chemical contamination levels in the Mediterranean based on the use of mussel caging. *Marine Pollution Bulletin*, *49*(9), 704-712. https://doi.org/10.1016/j.marpolbul.2004.05.008

Araújo, D. F., Ponzevera, E., Briant, N., Knoery, J., Bruzac, S., Sireau, T., & Brach-Papa, C. (2019). Copper, zinc and lead isotope signatures of sediments from a mediterranean coastal bay impacted by naval activities and urban sources. *Applied Geochemistry*, *111*, 104440-. https://doi.org/10.1016/j.apgeochem.2019.104440

Araújo, M., Conceição, A., Barbosa, T., Lopes, M., & Humanes, M. (2003). Elemental composition of marine sponges from the Berlengas Natural Park, western Portuguese coast. *X-Ray Spectrometry*, *32*, 428-433. https://doi.org/10.1002/xrs.660

Arnaud, A., Prudent, P., & Laffont-Schwob, I. (2021). Évaluation du risque sanitaire de sols pollués méditerranéens : Choix de variables et spatialisation. *VertigO - la revue électronique en sciences de l'environnement*, *Volume 21 numéro 1*, Art. Volume 21 numéro 1. 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-parcnational.fr/fr/rapports-scientifiques/le-port-de-porquerolles-iles-dhyeres-mediterranee-france-ii-contamination-par

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 Materials and Environmental Science*, *9*. https://doi.org/10.26872/jmes.2018.9.4.129

ACCEPTED VERSION

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
463 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,

ACCEPTED VERSION

Demospongiae, Oscarellidae). *Journal of Natural History*, *26*, 271-284. https://doi.org/10.1080/00222939200770131

Caplat, C., Basuyaux, O., & Mahaut, M.-L. (2011, juillet 3). *Water quality assessment using a sponge (Hymeniacidon perlevis) as bioindicator—First results of metallic and organic contaminations given by biotests*.

Ceballos, G., Ehrlich, P. R., Barnosky, A. D., García, A., Pringle, R. M., & Palmer, T. M. (2015). Accelerated modern human–induced species losses : Entering the sixth mass extinction. *Science Advances*, *1*(5), e1400253. https://doi.org/10.1126/sciadv.1400253

Ceballos, G., Ehrlich, P. R., & Dirzo, R. (2017). Biological annihilation via the ongoing sixth mass extinction signaled by vertebrate population losses and declines. *Proceedings of the National Academy of Sciences*, *114*(30), E6089-E6096. https://doi.org/10.1073/pnas.1704949114

Cebrian, E. (2007). *Sponges as biomonitors of heavy metals in spatial and temporal surveys in northwestern mediterranean : Multispecies comparison. - PubMed—NCBI*. https://www.ncbi.nlm.nih.gov/pubmed/17941749

Cebrian, E., Martí, R., Uriz, J. M., & Turon, X. (2003). Sublethal effects of contamination on the Mediterranean sponge Crambe crambe : Metal accumulation and biological responses. *Marine Pollution Bulletin*, *46*(10), 1273-1284. https://doi.org/10.1016/S0025-326X(03)00190-5

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

Chifflet, S., Briant, N., Tesán-Onrubia, J. A., Zaaboub, N., Amri, S., Radakovitch, O., Bănaru, D., & Tedetti, M. (2022). Distribution and accumulation of metals and metalloids in planktonic food webs of the Mediterranean Sea (MERITE-HIPPOCAMPE campaign). *Marine Pollution Bulletin*, *186*, 114384. https://doi.org/10.1016/j.marpolbul.2022.114384

ACCEPTED VERSION

Ciesielski, H. (2007). Effets du pH sur l'extraction des éléments traces métalliques dans les sols. *Etude et Gestion des Sols*, 24.

Coll, M., Piroddi, C., Steenbeek, J., Kaschner, K., Ben Rais Lasram, F., Aguzzi, J., Ballesteros, E., Bianchi, C. N., Corbera, J., Dailianis, T., Danovaro, R., Estrada, M., Froglia, C., Galil, B. S., Gasol, J. M., Gertwagen, R., Gil, J., Guilhaumon, F., Kesner-Reyes, K., … Voultsiadou, E. (2010). The Biodiversity of the Mediterranean Sea : Estimates, Patterns, and Threats. *PLoS ONE*, *5*(8), e11842. https://doi.org/10.1371/journal.pone.0011842

Danovaro, R. (2003). Pollution threats in the Mediterranean Sea : An overview. *Chemistry and Ecology*, *19*(1), 15-32. https://doi.org/10.1080/0275754031000081467

Daumalin, X., Laffont-Schwob, I., Barthélemy, C., Desrousseaux, M., Heckenroth, A., Masotti, V., Monsara, P., Kronenberger, S., Oursel, B., Prudent, P., Rabier, J., & Raveux, O. (2016). *Les calanques industrielles de Marseille et leurs pollutions—Une histoire au présent*. 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., Downey, R., Hajdu, E., Hooper, J. N. A., Kelly, M., Klautau, M., Manconi, R., Morrow, C. C., Pisera, A. B., Ríos, P., Rützler, K., Schönberg, C., Vacelet, J., & van Soest, R. W. M. (2022). *World Porifera Database*. https://doi.org/10.14284/359

Durrieu de Madron, X., Guieu, C., Sempéré, R., Conan, P., Cossa, D., D'Ortenzio, F., Estournel, C., Gazeau, F., Rabouille, C., Stemmann, L., Bonnet, S., Diaz, F., Koubbi, P., Radakovitch, O., Babin, M., Baklouti, M., Bancon-Montigny, C., Belviso, S., Bensoussan, N., … Verney, R. (2011). Marine ecosystems' responses to climatic and anthropogenic forcings in the Mediterranean. *Progress in Oceanography*, *91*(2), 97-166. 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

ACCEPTED VERSION

Ereskovsky, A. V., Borchiellini, C., Gazave, E., Ivanisevic, J., Lapébie, P., Perez, T., Renard, E., & Vacelet, J. (2009). The Homoscleromorph sponge Oscarella lobularis, a promising sponge model in evolutionary and developmental biology. *BioEssays*, *31*(1), 89-97. https://doi.org/10.1002/bies.080058

Ereskovsky, A. V., Borisenko, I. E., Lapébie, P., Gazave, E., Tokina, D. B., & Borchiellini, C. (2015). Oscarella lobularis (Homoscleromorpha, Porifera) Regeneration : Epithelial Morphogenesis and Metaplasia. *PloS One*, *10*(8), e0134566. https://doi.org/10.1371/journal.pone.0134566

Ereskovsky, A. V., Dubois, M., Ivanišević, J., Gazave, E., Lapebie, P., Tokina, D., & Pérez, T. (2013). Pluri-annual study of the reproduction of two Mediterranean Oscarella species (Porifera, Homoscleromorpha) : Cycle, sex-ratio, reproductive effort and phenology. *Marine Biology*, *160*(2), 423-438. https://doi.org/10.1007/s00227-012-2100-9

European Environment Agency (Éd.). (2006). *Priority issues in the Mediterranean environment*. European Environmental Agency.

Fehlauer, T., Collin, B., Angeletti, B., Santaella, C., Dentant, C., Chaurand, P., Levard, C., Gonneau, C., Borschneck, D., & Rose, J. (2022). Uptake patterns of critical metals in alpine plant species growing in an unimpaired natural site. *Chemosphere*, *287*(4), 132315. https://doi.org/10.1016/j.chemosphere.2021.132315

Fierro-Constaín, L., Rocher, C., Marschal, F., Schenkelaars, Q., Séjourné, N., Borchiellini, C., & Renard, E. (2021). In Situ Hybridization Techniques in the Homoscleromorph Sponge Oscarella lobularis. *Methods in Molecular Biology (Clifton, N.J.)*, *2219*, 181-194. https://doi.org/10.1007/978-1-0716-0974-3_11

Fierro-Constain, L., Schenkelaars, Q., Gazave, E., Haguenauer, A., Rocher, C., Ereskovsky, A., Borchiellini, C., & Renard, E. (2017). The Conservation of the Germline Multipotency Program, from Sponges to Vertebrates : A Stepping Stone to Understanding the Somatic and Germline Origins. *Genome Biology and Evolution*, *22*(3), 474-488. https://doi.org/10.1093/gbe/evw289

ACCEPTED VERSION

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

ACCEPTED VERSION

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
589 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
593 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/
595 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

ACCEPTED VERSION

598 Jourda, C., Santini, S., Rocher, C., Le Bivic, A., & Claverie, J.-M. (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, J.-L. (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-littoral--cellule-
616 interregionale-d-epidemiologie-sud-institut-de-veille-sanitaire
617 Lee, O. O., Wong, Y. H., & Qian, P.-Y. (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

ACCEPTED VERSION

624 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
643 Milanese, M., Chelossi, E., Manconi, R., Sarà, A., Sidri, M., & Pronzato, R. (2003). The marine sponge
644 Chondrilla nucula Schmidt, 1862 as an elective candidate for bioremediation in integrated
645 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

ACCEPTED VERSION

Biscay (North-East Atlantic Ocean). *Marine Pollution Bulletin*, *135*, 9-16. 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 Lagrangian transport of fine particles by Northern Current intrusions in the bays of Marseille (France). *PLOS ONE*, *13*, e0195257. https://doi.org/10.1371/journal.pone.0195257

Morganti, T. M., Ribes, M., Yahel, G., & Coma, R. (2019). Size Is the Major Determinant of Pumping Rates in Marine Sponges. *Frontiers in Physiology*, *10*, 1474. https://doi.org/10.3389/fphys.2019.01474

Müller, W. E. G., Batel, R., Lacorn, M., Steinhart, H., Simat, T., Lauenroth, S., Hassanein, H., & Schröder, H. C. (1998). Accumulation of cadmium and zinc in the marine sponge Suberites domuncula and its potential consequences on single-strand breaks and on expression of heat-shock protein : A natural field study. *Marine Ecology Progress Series*, *167*, 127-135. https://doi.org/10.3354/meps167127

Nogales, B., Lanfranconi, M. P., Piña-Villalonga, J. M., & Bosch, R. (2011). Anthropogenic perturbations in marine microbial communities. *FEMS Microbiology Reviews*, *35*(2), 275-298. 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 Environmental Contamination and Toxicology*, *52*(5), 722-728. https://doi.org/10.1007/BF00195494

Orani, A. M., Barats, A., Vassileva, E., & Thomas, O. P. (2018). Marine sponges as a powerful tool for trace elements biomonitoring studies in coastal environment. *Marine Pollution Bulletin*, *131*, 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

ACCEPTED VERSION

northwestern Mediterranean sponges. *Chemosphere*, *201*, 826-839. https://doi.org/10.1016/j.chemosphere.2018.03.078

Orani, A. M., Vassileva, E., Azemard, S., & Thomas, O. P. (2020). Comparative study on Hg bioaccumulation and biotransformation in Mediterranean and Atlantic sponge species. *Chemosphere*, *260*, 127515. https://doi.org/10.1016/j.chemosphere.2020.127515

Oursel, B. (2013). *Transferts et dynamique des contaminants métalliques en zone côtière. : Impact d'une grande agglomération méditerranéenne* (Numéro 2013TOUL0019) [Theses, Université de Toulon]. https://tel.archives-ouvertes.fr/tel-01132521

Padovan, A., Munksgaard, N., Alvarez, B., McGuinness, K., Parry, D., & Gibb, K. (2012). Trace metal concentrations in the tropical sponge Spheciospongia vagabunda at a sewage outfall : Synchrotron X-ray imaging reveals the micron-scale distribution of accumulated metals. *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 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 Environment*, *41*(2), 143-152. https://doi.org/10.1016/0048-9697(85)90184-6

Penz, C., Simonsen, T., & Devries, P. (2011). A new Orobrassolis butterfly (Nymphalidae, Brassolini) : A casualty of habitat destruction? *Zootaxa*, *2740*, 35-43. https://doi.org/10.11646/zootaxa.2740.1.3

Perez, T., Longet, D., Schembri, T., Rebouillon, P., & Vacelet, J. (2005). Effects of 12 years' operation of a sewage treatment plant on trace metal occurrence within a Mediterranean commercial sponge (Spongia officinalis, Demospongiae). *Marine Pollution Bulletin*, *50*(3), 301-309. https://doi.org/10.1016/j.marpolbul.2004.11.001

ACCEPTED VERSION

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. https://doi.org/10.1016/j.chemosphere.2006.01.072

Renard, E., Rocher, C., Ereskovsky, A., & Borchiellini, C. (2021). The Homoscleromorph Sponge, Oscarella lobularis. In *Handbook of Marine Model Organisms in Experimental Biology* (p. 79-100). CRC Press. https://doi.org/10.1201/9781003217503-5

Rocher, C., Vernale, A., Fierro-Constaín, L., Séjourné, N., Chenesseau, S., Marschal, C., Le Goff, E., Dutilleul, M., Matthews, C., Marschal, F., Brouilly, N., Massey-Harroche, D., Ereskovsky, A., Le Bivic, A., Borchiellini, C., & Renard, E. (2020). The buds of Oscarella lobularis (Porifera) : A new convenient model for sponge cell and developmental biology. *BioRxiv*, 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 compartments and suspended particulate matter in two coupled Mediterranean coastal ecosystems (Toulon Bay, France). *Marine Pollution Bulletin*, *56*(11), 1862-1870. https://doi.org/10.1016/j.marpolbul.2008.07.018

Roussiez, V., Ludwig, W., Monaco, A., Probst, J.-L., Bouloubassi, J., Buscail, R., & Saragoni, G. (2006). 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. (2020). Hg Levels in Marine Porifera of Montecristo and Giglio Islands (Tuscan Archipelago, Italy). *Applied Sciences*, *10*, 4342. https://doi.org/10.3390/app10124342

Santos-Echeandía, J., Campillo, J. A., Egea, J. A., Guitart, C., González, C. J., Martínez-Gómez, C., León, V. M., Rodríguez-Puente, C., & Benedicto, J. (2021). The influence of natural vs

ACCEPTED VERSION

anthropogenic factors on trace metal(loid) levels in the Mussel Watch programme : Two decades of monitoring in the Spanish Mediterranean sea. *Marine Environmental Research*, *169*, 105382. https://doi.org/10.1016/j.marenvres.2021.105382

Schenkelaars, Q., Fierro-Constain, L., Renard, E., Hill, A. L., & Borchiellini, C. (2015). Insights into Frizzled evolution and new perspectives. *Evolution & Development*, *17*(2), 160-169. https://doi.org/10.1111/ede.12115

Schuhmacher, M., Batiste, J., Bosque, M. A., Domingo, J. L., & Corbella, J. (1994). Mercury concentrations in marine species from the coastal area of Tarragona Province, Spain. Dietary intake of mercury through fish and seafood consumption. *The Science of the Total Environment*, *156*(3), 269-273. https://doi.org/10.1016/0048-9697(94)90193-7

Selvin, J., Shanmugha Priya, S., Seghal Kiran, G., Thangavelu, T., & Sapna Bai, N. (2009). Sponge-associated marine bacteria as indicators of heavy metal pollution. *Microbiological Research*, *164*(3), 352-363. https://doi.org/10.1016/j.micres.2007.05.005

Tchounwou, P. B., Yedjou, C. G., Patlolla, A. K., & Sutton, D. J. (2012). Heavy Metal Toxicity and the Environment. In A. Luch (Éd.), *Molecular, Clinical and Environmental Toxicology : Volume 3 : Environmental Toxicology* (p. 133-164). Springer. https://doi.org/10.1007/978-3-7643-8340-4_6

Tessier, E., Garnier, C., Mullot, J.-U., Lenoble, V., Arnaud, M., Raynaud, M., & Mounier, S. (2011). Study of the spatial and historical distribution of sediment inorganic contamination in the Toulon bay (France). *Marine Pollution Bulletin*, *62*(10), 2075-2086. https://doi.org/10.1016/j.marpolbul.2011.07.022

Tian, R.-M., Wang, Y., Bougouffa, S., Gao, Z.-M., Cai, L., Zhang, W.-P., Bajic, V., & Qian, P.-Y. (2014). Effect of Copper Treatment on the Composition and Function of the Bacterial Community in the Sponge Haliclona cymaeformis. *MBio*, *5*(6), e01980-14. https://doi.org/10.1128/mBio.01980-14

ACCEPTED VERSION

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
766 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
771 Yamaoka, Y., Carmona, M. L., Oclarit, J. M., Jin, K., & Shibata, Y. (2001). Arsenic compounds in marine
772 sponge (Haliclona permolis, Halichondria japonica, Halichondria okadai andHaliclona sp.
773 White) from Seto Inland Sea, Japan. *Applied Organometallic Chemistry*, *15*(4), 261-265.
774 https://doi.org/10.1002/aoc.135

ACCEPTED VERSION

775 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
778 Zhou, Q., Zhang, J., Fu, J., Shi, J., & Jiang, G. (2008). Biomonitoring : An appealing tool for assessment
779 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
783 through multiple assessment approaches. *Environmental Pollution*, *264*, 114791.
784 https://doi.org/10.1016/j.envpol.2020.114791
785
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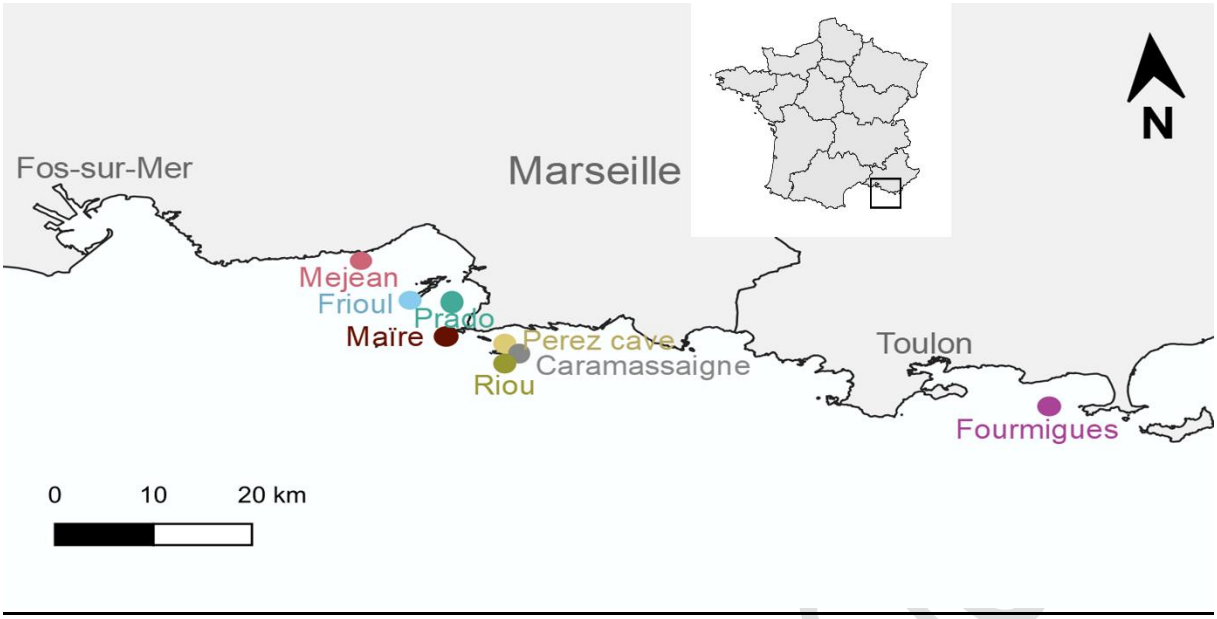


Figure 1: Location of the eight sampling sites (south of France; North-Western Mediterranean Sea).

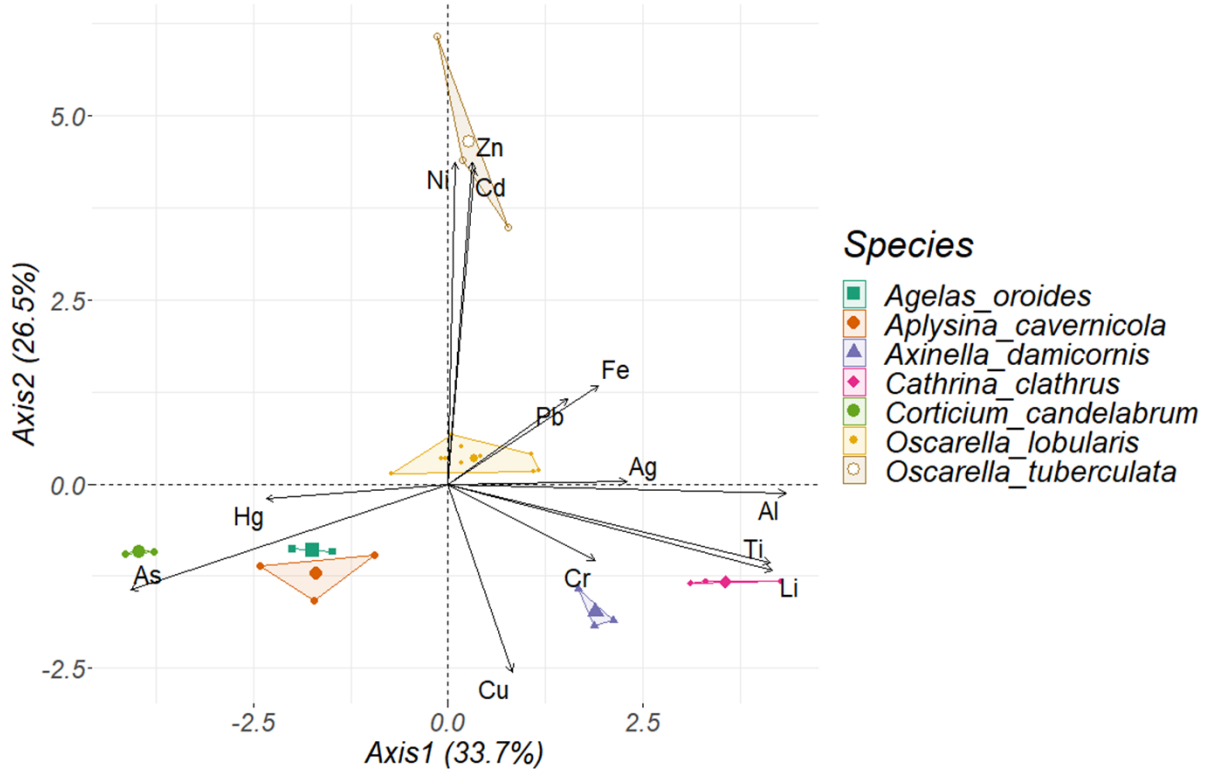


Figure 2: Principal component analysis (PCA) of metal concentrations in seven distinct marine sponge species collected in the Maïre site. Convex ellipses group heavy metal concentrations from the seven species.

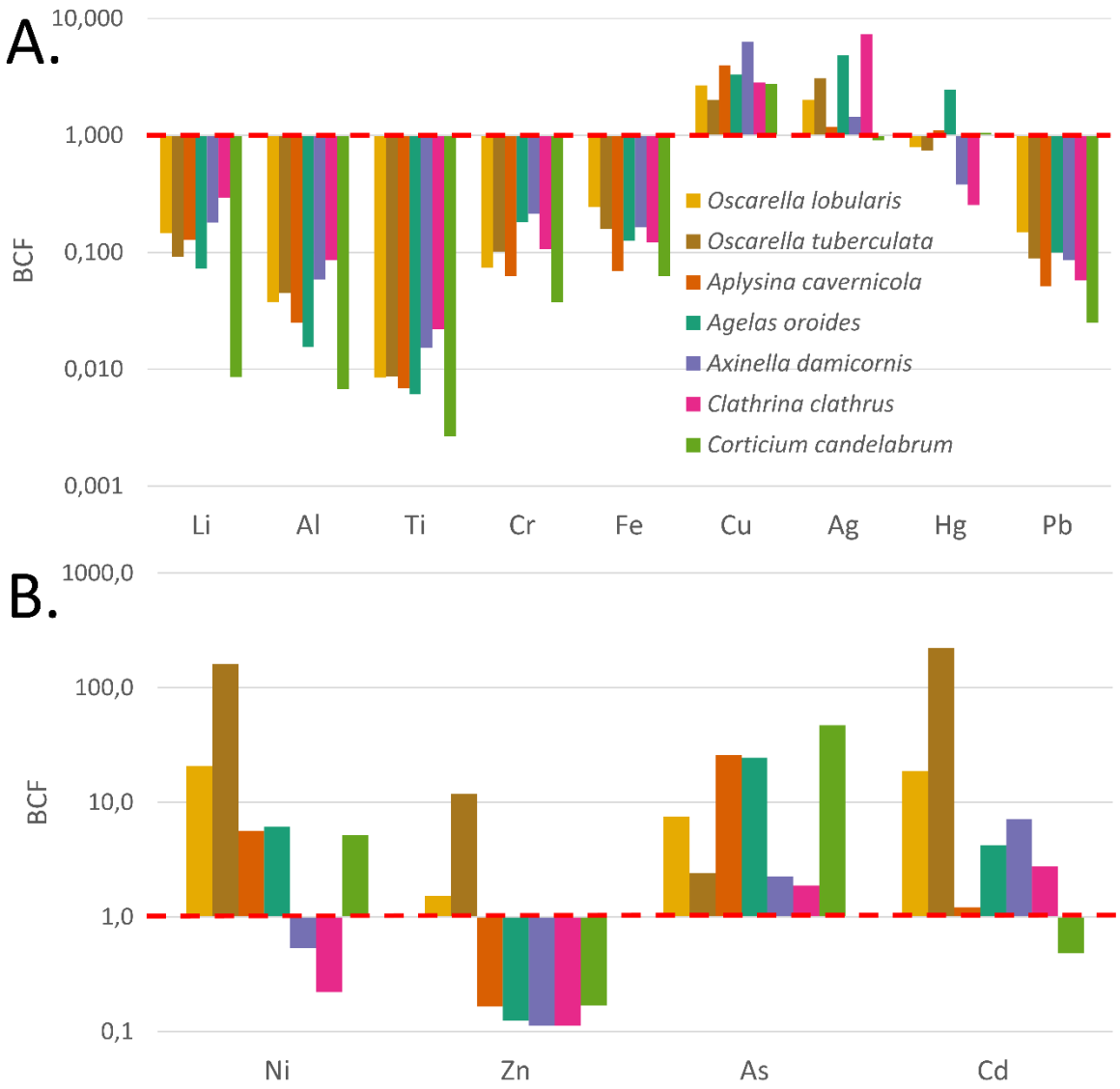
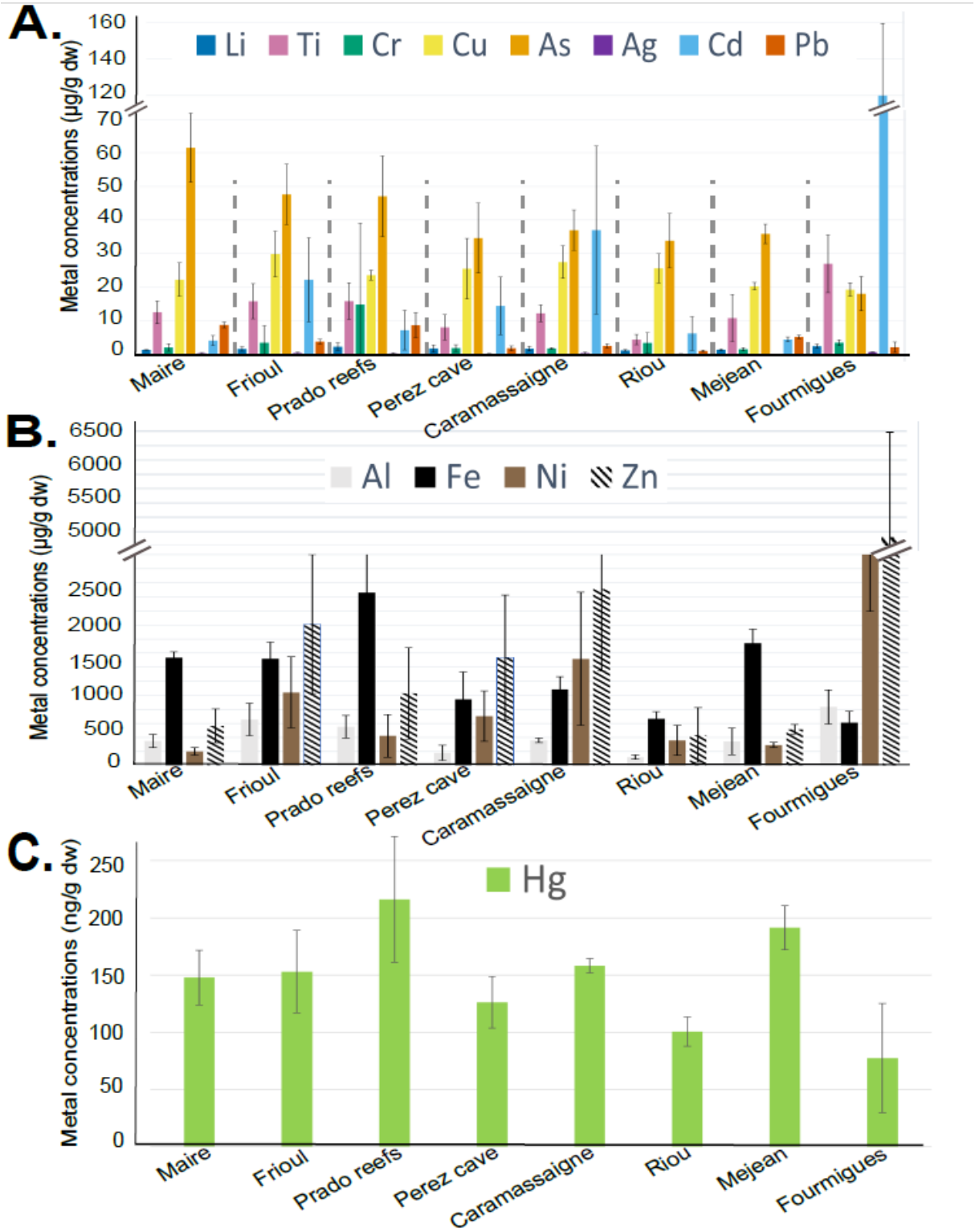


Figure 3: Bioconcentration factors calculated for all metals in seven marine sponges. BCFs of (A) Li, Al, Ti, Cr, Fe, Cu, Ag, Hg, Pb and (B) Ni, Zn, As, Cd. Values are available in table A.6.

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

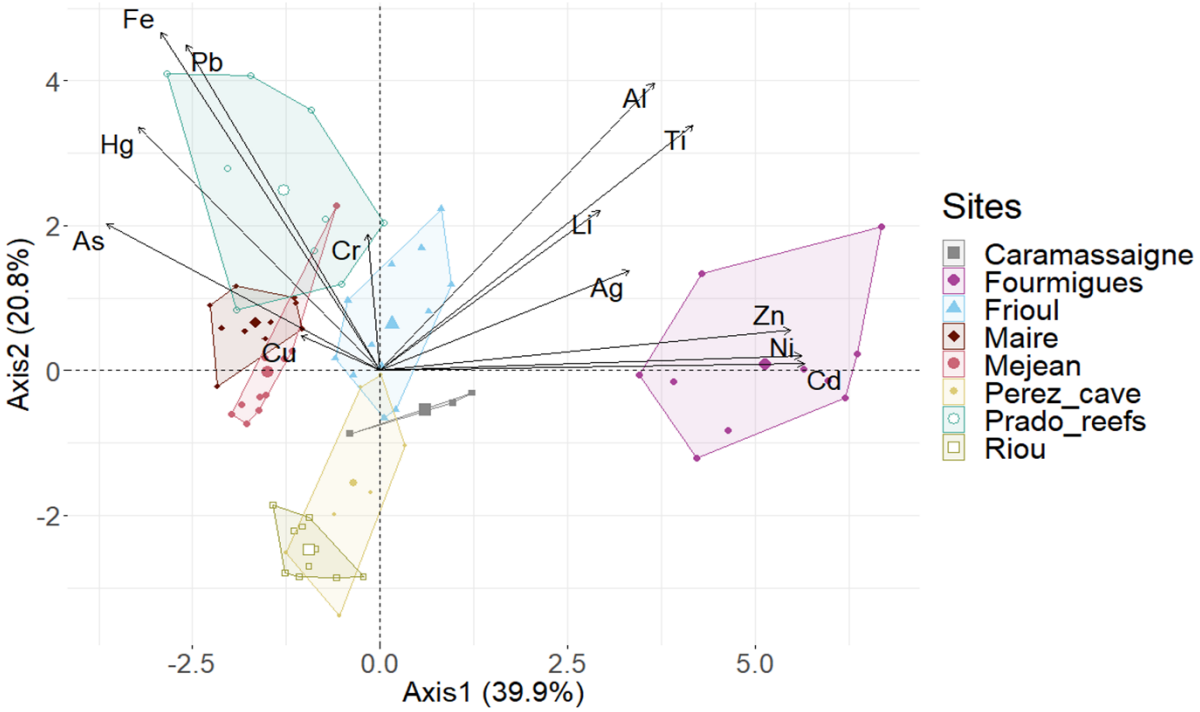


Figure 5: Principal component analysis (PCA) plot describing inter-site variation of metal contents present in *Oscarella lobularis* tissues.

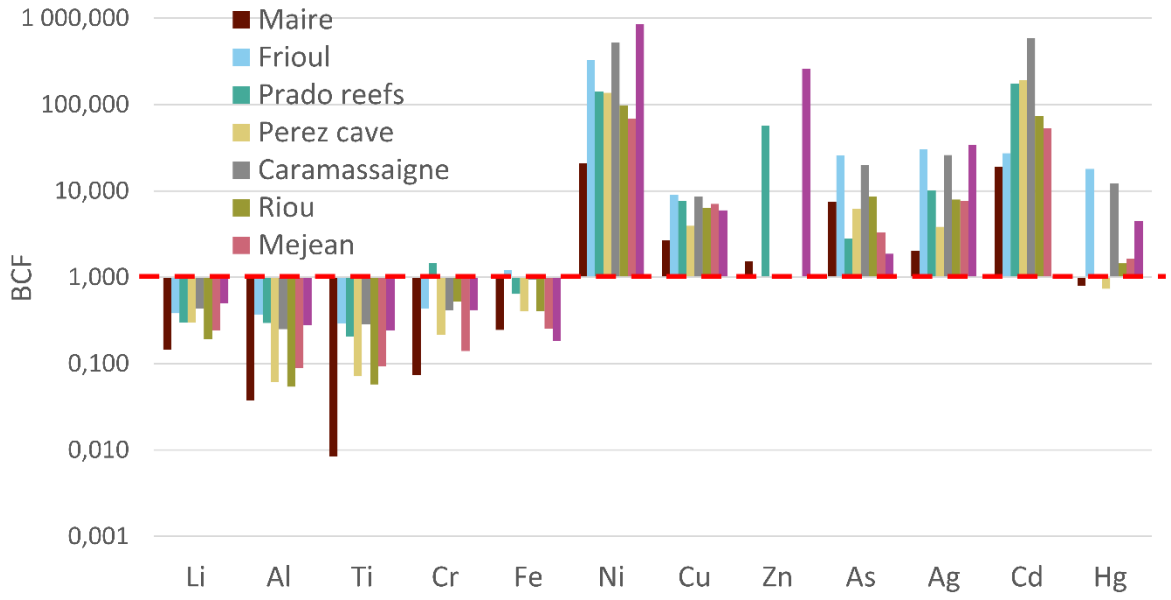


Figure 6: Bioconcentration factors (BCF) calculated for all metals in *Oscarella lobularis* individuals sampled at different sites (using metal concentrations measured in sediments as reference). Values are available in table A.6.

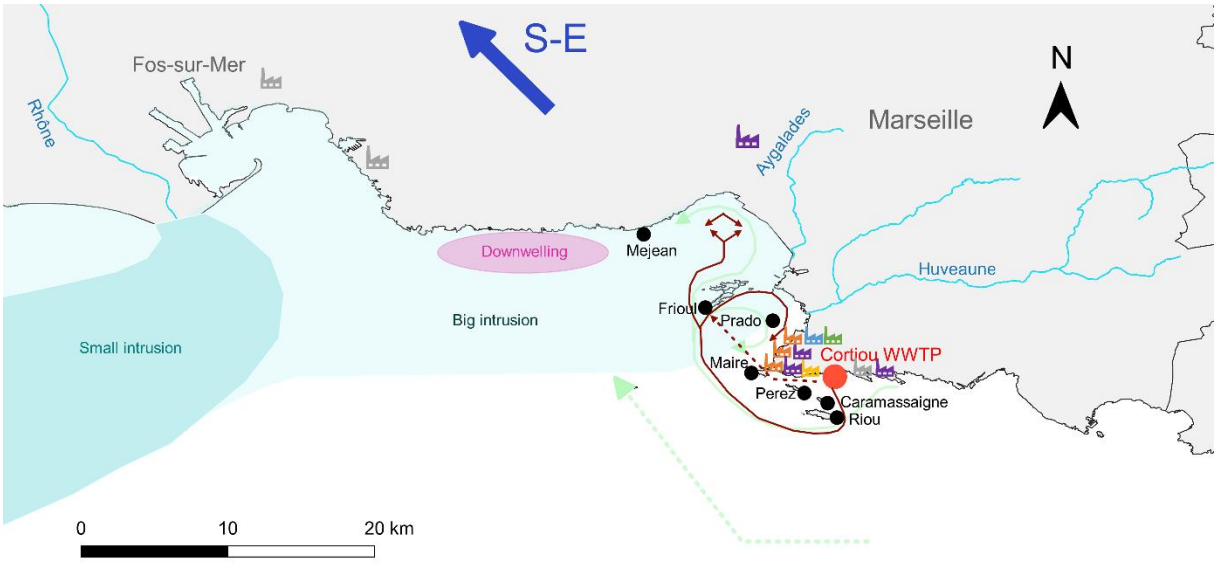


Figure 7: Hydrodynamics in the bay of Marseille in presence of south-east winds. Including the North Current intrusion (dotted green arrow for the surface current and green arrow for the subsurface), the downwelling formation and the intrusions of the Rhône River. Red arrows indicate the subsurface particles 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.

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Appendix A. Supplementary material

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

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 (other 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 2. Limit of quantification (LOQ) in µg/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 µg/L	0,15 µg/L	0,05 µg/L	0,009 µg/L	0,3 µg/L	0,006 µg/L	0,014 µg/L	0,05 µg/L	0,037 µg/L	0,002 µg/L	0,017 µg/L	0,003 ng	0,005 µg/L

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Supplemental Table 3. Samples measured with AMA-254 and ICP-MS to determine the metal 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	Maire	11	3	12	1-5
	Frioul	12	3	13	1-2
	Prado's Reef	9	3	10	2
	Perez Cave	7	3	8	2
	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
Sediment	Maire		3		1
	Frioul		3		3
	Prado's Reef		3		2
	Perez Cave		3		3
	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

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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 ± SD. Different letters indicate a significant difference between sites for *O. lobularis* samples (Kruskall-Wallis test followed by a Dunn post hoc test; $p < 0,05$)

Samples	Sites	Li	Al	Ti	Cr	Fe	Ni	Cu	Zn	As	Ag	Cd	Hg	Pb
Oscarella lobularis	Maire	1,32 ± 0,12 $^{(ab)}$	349 ± 93 $^{(ab)}$	12,5 ± 3,4 $^{(abc)}$	2,04 ± 1,12 $^{(a)}$	1536 ± 83 $^{(ab)}$	201 ± 53 $^{(a)}$	22,2 ± 5 $^{(ab)}$	564 ± 241 $^{(ab)}$	61,5 ± 10,3 $^{(a)}$	0,36 ± 0,2 $^{(ab)}$	4,1 ± 1,5 $^{(a)}$	0,148 ± 0,024 $^{(a)}$	8,76 ± 0,87 $^{(a)}$
Oscarella lobularis	Frioul	1,62 ± 0,69 $^{(ab)}$	655 ± 232 $^{(cd)}$	15,8 ± 5,2 $^{(a)}$	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 $^{(a)}$	3,79 ± 0,73 $^{(bc)}$
Oscarella lobularis	Prado reefs	2,3 ± 1,14 $^{(ac)}$	552 ± 162 $^{(acd)}$	15,8 ± 5,5 $^{(abcd)}$	14,8 ± 24,3 $^{(c)}$	2460 ± 912 $^{(c)}$	422 ± 305 $^{(ade)}$	23,6 ± 1,5 $^{(ac)}$	1026 ± 650 $^{(abcc)}$	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 $^{(ade)}$	25,5 ± 9 $^{(ac)}$	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 $^{(d)}$	358 ± 214 $^{(de)}$	25,6 ± 4,4 $^{(ac)}$	433 ± 389 $^{(b)}$	33,8 ± 8,1 $^{(d)}$	0,18 ± 0,04 $^{(b)}$	6,22 ± 5 $^{(de)}$	0,101 ± 0,013 $^{(c)}$	1,02 ± 0,28 $^{(d)}$
Oscarella lobularis	Mejean	1,38 ± 0,3 $^{(ab)}$	344 ± 189 $^{(ab)}$	10,8 ± 7,1 $^{(bc)}$	1,54 ± 0,47 $^{(a)}$	1740 ± 197 $^{(bc)}$	294 ± 37 $^{(de)}$	20,3 ± 1,1 $^{(b)}$	516 ± 68 $^{(b)}$	35,9 ± 2,9 $^{(d)}$	0,1 ± 0,01 $^{(c)}$	4,44 ± 0,74 $^{(be)}$	0,191 ± 0,019 $^{(b)}$	5,21 ± 0,51 $^{(ac)}$
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 ± 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%	0,815 ± 6,3%	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%	16,8 ± 1,5%	0,0278 ± 7,7%	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	5,58 ± 5,9%	0,0546 ± 3,9%	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%	0,0146 ± 1,3%	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%	0,0227 ± 2,6%	0,0852 ± 40%	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%	2,88 ± 4,7%	0	10,8 ± 4,1%	0,0131 ± 8,9%	0,0834 ± 0,9%	0,116 ± 21%	18 ± 2,3%
Sediment	Fourmigues	9,10 ± 13%	3008 ± 3,2%	111 ± 1,3%	8,32 ± 1,7%	3336 ± 1,4%	3,86 ± 11,1%	3,23 ± 3,6%	19 ± 4,5%	9,67 ± 1,5%	0,02 ± 2%	0	0,0172 ± 19,31%	11,3 ± 4,3%

Supplemental Table 6. Average bioconcentration factors (BCF) calculated for seven sponge species. 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	Al	Ti	Cr	Fe	Ni	Cu	Zn	As	Ag	Cd	Hg	Pb
O.lobularis	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
	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
	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

ACCEPTED VERSION

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ACCEPTED VERSION