



HAL
open science

Bacterial community assemblages in sediments under high anthropogenic pressure at Ichkeul Lake/Bizerte Lagoon hydrological system, Tunisia

Fida Ben Salem, Olfa Ben Saïd, Cristiana Cravo-Laureau, Ezzeddine Mahmoudi, Noëlle Bru, Mathilde Monperrus, Robert Duran

► To cite this version:

Fida Ben Salem, Olfa Ben Saïd, Cristiana Cravo-Laureau, Ezzeddine Mahmoudi, Noëlle Bru, et al.. Bacterial community assemblages in sediments under high anthropogenic pressure at Ichkeul Lake/Bizerte Lagoon hydrological system, Tunisia. *Environmental Pollution*, 2019, 252, pp.644-656. 10.1016/j.envpol.2019.05.146 . hal-02170012

HAL Id: hal-02170012

<https://univ-pau.hal.science/hal-02170012v1>

Submitted on 25 Oct 2021

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.



Distributed under a Creative Commons Attribution - NonCommercial 4.0 International License

1 **Bacterial community assemblages in sediments under high anthropogenic pressure at**
2 **Ichkeul Lake / Bizerte Lagoon hydrological system, Tunisia**

3

4 **Fida Ben Salem^{a,b,d}, Olfa Ben Said^{a,b,d}, Cristiana Cravo-Laureau^{b,d}, Ezzeddine**
5 **Mahmoudi^a, Noëlle Bru^{c,d}, Mathilde Monperrus^{b,d}, Robert Duran^{b,d*}**

6

7 ^aLaboratoire de Biosurveillance de l'Environnement, Faculté des Sciences de Bizerte, Tunisie.

8 ^bMELODY group, Université de Pau et des Pays de l'Adour, E2S-UPPA, IPREM UMR
9 CNRS 5254, BP 1155, Pau Cedex, 64013, France

10 ^cLaboratoire de Mathématiques et de leurs Applications, PAU UMR CNRS 5142 – Université
11 de Pau et des Pays de l'Adour, E2S-UPPA, France

12 ^dFédération de recherche MIRA, Université de Pau et des Pays de l'Adour, E2S-UPPA,
13 France

14

15

16 *Corresponding author present address:

17 Pr. Robert Duran

18 Université de Pau et des Pays de l'Adour

19 Bâtiment IBEAS

20 BP1155

21 64013 Pau Cedex, France

22

23 Tel: +33559407568; Fax: +33559407490

24 Email: Robert.duran@univ-pau.fr

25

26 **Highlights**

- 27 • Evidence of bacterial continuum in Ichkeul Lake / Bizerte Lagoon
- 28 • The lake and the lagoon have specific bacterial assemblages
- 29 • Each type of pollutant is associated with specific bacterial genera
- 30 • Uncontaminated sediments have also specific bacterial genera
- 31 • Potential to develop tools for managing bacterial resources

32

33 **Abstract**

34 Bacterial communities inhabiting sediments in coastal areas endure the effect of strong
35 anthropogenic pressure characterized by the presence of multiple contaminants.
36 Understanding the effect of pollutants on the organization of bacterial communities is of
37 paramount importance in order to unravel bacterial assemblages colonizing specific
38 ecological niches. Here, chemical and molecular approaches were combined to investigate the
39 bacterial communities inhabiting the sediments of the Ichkeul Lake / Bizerte Lagoon, a
40 hydrological system under anthropogenic pressure. Although the microbial community of the
41 Ichkeul Lake sediment was different to that of the Bizerte Lagoon, common bacterial genera
42 were identified suggesting a lake-lagoon continuum probably due to the hydrology of the
43 system exchanging waters according to the season. These genera represent bacterial
44 "generalists" maintaining probably general biogeochemical functions. Linear discriminant
45 analysis effect size (LEfSe) showed significant differential abundance distribution of bacterial
46 genera according to the habitat, the pollution type and level. Further, correlation analyses
47 identified specific bacterial genera which abundance was linked with pesticides
48 concentrations in the lake, while in the lagoon the abundance of specific bacterial genera was
49 found linked with the concentrations of PAHs (Polycyclic aromatic hydrocarbons) and
50 organic forms of Sn. As well, bacterial genera which abundance was not correlated with the
51 concentrations of pollutants were identified in both lake and lagoon. These findings represent
52 valuable information, pointing out specific bacterial genera associated with pollutants, which
53 represent assets for developing bacterial tools for the implementation, the management, and
54 monitoring of bioremediation processes to mitigate the effect of pollutants in aquatic
55 ecosystems.

56 **Keywords:** 16S rRNA sequencing, pollution, pesticide, PAH, network analysis

57 **1. Introduction**

58 Lagoons and lakes are highly productive ecosystems, providing important ecological services
59 such as breeding grounds and nurseries for many fish and bird species (Vasconcelos et al.,
60 2011). They are threatened by human activities, and therefore they need careful monitoring
61 because they are very vulnerable (Pérez-Domínguez et al., 2012). Their status may change
62 rapidly in response to natural- and human-driven events (Cravo-Laureau et al., 2017a; Duran
63 et al., 2015c). Sediments are complex habitats densely colonized by diverse microorganisms
64 (Brito et al., 2009; Chronopoulou et al., 2013; Duran et al., 2015a; Fourcans et al., 2008). In
65 sediments, microbial communities are involved in the decomposition of organic matter
66 (Schultz and Urban, 2008). Microorganisms play a key role in carbon, nitrogen and
67 phosphorus cycling (Andrews et al., 2011) as well as in determining the fate of pollutants
68 (Bordenave et al., 2004a; Bordenave et al., 2004b; Guermouche M'rassi et al., 2015; Pringault
69 et al., 2008; Stauffert et al., 2013). Polycyclic aromatic hydrocarbons (PAHs), polychlorinated
70 biphenyls (PCBs) and organochlorine pesticides (OCPs) are considered as persistent organic
71 pollutants (POPs), which are ubiquitous in aquatic ecosystems (Doong et al., 2008; Kjellerup
72 et al., 2014; Mwanamoki et al., 2014). POPs are mainly derived from industrial and
73 agricultural sources, as well as maritime traffic (Chang and Doong, 2006; Wen et al., 1994).
74 They enter aquatic environments through atmospheric deposition and run-off (Botta et al.,
75 2009; Duran and Cravo-Laureau, 2016). Their fate in the marine environment depends on
76 abiotic and biotic factors (for review, see Duran and Cravo-Laureau, 2016). The rapid
77 development of molecular methods for microbial ecology has provided detailed information
78 on microbial diversity and dynamics in marine sediments (Bordenave et al., 2008; Stauffert et
79 al., 2014; Terrisse et al., 2017; Thiyagarajan et al., 2010). The molecular methods, particularly
80 high-throughput sequencing technologies have been proved to be useful tools for bio-
81 monitoring aquatic ecosystem (Cravo-Laureau and Duran, 2014; Cravo-Laureau et al.,

82 2017b). Their utilization in combination with chemical analyses might shed light on the
83 ecological role and the adaptation of bacteria to environmental factors. Correlation analyses
84 allowed the exploration of the interactions between microorganisms and their habitat
85 (Barberan et al., 2012; Fuhrman and Steele, 2008; Llado et al., 2015; Williams et al., 2014;
86 Yergeau et al., 2012). Particularly, such analyses have identified possible pollutant degraders
87 (Llado et al., 2015; Yergeau et al., 2012), thereby obtaining valuable information for guiding
88 the management of microbial communities to mitigate the effect of pollutants.

89 The Ichkeul Lake / Bizerte Lagoon hydrological system (Tunisia) is located in an area under
90 high anthropic pressure. The Ichkeul Lake, Ramsar protected area, is important for migrating
91 birds (Ramdani et al., 2001). The contaminants entering into the Ichkeul Lake / Bizerte
92 Lagoon hydrological system (Stevenson and Battarbee, 1991; Trabelsi and Driss, 2005) have
93 been shown to affect aquatic organisms and migratory birds (Ben Said et al., 2009;
94 Casagrande and Boudouresque, 2010; Gharbi-Bouraoui et al., 2008). During the past decades
95 several projects have studied the PAHs in the sediment of Bizerte Lagoon and their impact on
96 the benthic microorganisms and organisms (Ben Said et al., 2009; Ben Said et al., 2008; Ben
97 Said et al., 2015; Louati et al., 2013a; Louati et al., 2013b; Trabelsi and Driss, 2005).
98 Recently, we characterized the contamination of the Ichkeul Lake / Bizerte Lagoon
99 hydrological system. Almost all stations exceeded the threshold effect level (TEL) of the
100 standards published by NOAA (Ben Salem et al., 2017). The Bizerte Lagoon has been shown
101 to contain industrial contaminants including hydrocarbon (PAHs, polycyclic aromatic
102 hydrocarbons) and metallic compounds, while the Ichkeul Lake was found to contain
103 agrochemicals contaminants including organochlorine pesticides such as endrin and dieldrin
104 (Ben Salem et al., 2016a; Ben Salem et al., 2016b; Ben Salem et al., 2017). In a previous
105 study, based on 16S rRNA gene T-RFLP (terminal restriction fragment length polymorphism)
106 analysis investigating the bacterial communities in four contrasted stations of the Ichkeul

107 Lake / Bizerte Lagoon hydrological system (two located at the Ichkeul Lake, and two at the
108 Bizerte Lagoon), we demonstrated that bacterial community compositions were linked with
109 the type and level of pesticide contamination at the dry season (Ben Salem et al., 2016a). The
110 current study aims to characterize the bacterial communities from the sediments of the Ichkeul
111 Lake / Bizerte Lagoon hydrological system. We further characterize the bacterial
112 communities structure by 16S rRNA genes T-RFLP analysis, expanding the study by
113 investigating 20 stations around the Ichkeul Lake / Bizerte Lagoon hydrological system at two
114 contrasted seasons (April, dry season; September, wet season). Also, we further characterize
115 the bacterial composition of four contrasted stations by sequencing 16S rRNA gene PCR-
116 amplified fragments via 454-pyrosequencing. The effect of organic contaminants such as
117 PAHs, PCBs, OCPs on the bacterial assemblages were determined. Specific bacteria, which
118 abundance was linked with the presence of a pollutant, were identified.

119

120 **2. Materials and Methods**

121 **2.1. Site description**

122 The Ichkeul Lake / Bizerte Lagoon hydrological system, North Tunisia, suffers the effects of
123 human activities because it is located in an area under high anthropic pressure. Bizerte
124 Lagoon has a central role because connected to the Mediterranean Sea and to the Ichkeul Lake
125 via the Tinja River; the hydrological system receives water via different rivers (Oueds) all
126 around (Fig. 1). The lake experiences large fluctuations in salinity each year, from freshwater
127 with average salinity around 4 PSU (Practical salinity unit) in winter (wet season) due to
128 rivers flood, to saline ecosystem with average salinity around 35 PSU in summer (dry season)
129 receiving seawater from the Bizerte Lagoon and because of evaporation. In a previous study,

130 we demonstrated that pollution levels fluctuated according to seasons (Ben Salem et al.,
131 2016a).

132 **2.2. Sampling**

133 Sediment samples were collected from 20 stations (ST) located all around the Ichkeul Lake /
134 Bizerte Lagoon hydrological system (Fig. 1): ST1 to ST8 in the lake and ST11 to ST20 in the
135 lagoon, ST9 on the channel connecting the lagoon to the sea, and ST10 in the Mediterranean
136 Sea. The lake and lagoon stations were located at the river mouths (13 stations), close to
137 industrial and agricultural areas (4 stations), and at the centers of the lake and the lagoon (2
138 stations). Two sampling campaigns were performed: September 2011 (wet season) and April
139 2012 (dry season). Surface sediments (0-10 cm) were collected in triplicates (3 independent
140 samples, i.e. biological replicates) at each station with a Van Veen grab as previously
141 described (Ben Salem et al., 2016a). Collected sediments were dispatched in cryotubes (3
142 technical replicates were taken), quickly frozen in liquid nitrogen for microbial analyses, and
143 in glass bottles kept at 4°C for chemical analyses. At lab, samples for microbial analyses were
144 stored at -80°C and those for chemical analyses at -20°C until use.

145 **2.3. Physical-chemical analyses**

146 Physical-chemical parameters, including temperature, dissolved oxygen, pH, salinity,
147 phosphate, ammonia, nitrite, and nitrate were determined as previously described (Ben Salem
148 et al., 2016a). Chemical analyses for contaminants followed the QuEChERS procedure
149 described in Ben Salem et al. (2016b). Chemical data and physical-chemical parameters are
150 summarized in supplementary material (Table S1). All chemical data and physical-chemical
151 parameters are presented in details in Ben Salem et al. (2017).

152 The contamination contents of the sediments were compared to sediment quality guidelines
153 (SQG) in order to determine their potential toxic effect. Sediments were classified according
154 the quality criteria developed by Bakke et al. (2010) for PCBs, OCPs, and PAHs. The toxic

155 potential of the sediments was evaluated by calculating the mean effect range medium (ERM)
156 quotient (m-ERM-Q) as previously described (Long et al., 1998).

157 **2.4. DNA extraction, PCR and T-RFLP analyses**

158 DNA extractions were performed using 250 mg of frozen sediment with the UltraClean soil
159 DNA isolation Kit (MoBio Laboratories, CA) as previously described (Precigou et al., 2004).
160 The bacterial universal primers 1387R 5'-GGGCGGWGTGTACAAGGC-3' and 63F 5'-
161 CAGGCCTAACACATGCAAGTC-3' (Marchesi et al., 1998) were used for 16S rRNA gene
162 amplifications. Primers were fluorescently labeled for T-RFLP analysis, which was performed
163 as previously described (Precigou et al., 2001) using 10 ng of extracted DNA. PCR mixture
164 and conditions were performed as previously described (Vercraene-Eairmal et al., 2010). PCR
165 amplicon purification, digestion and electrophoresis separation have been previously
166 described (Vitte et al., 2011). T-RFLP profiles were analyzed as previously described (Cravo-
167 Laureau et al., 2011).

168 **2.5. Sequencing and data analysis**

169 Among the 20 stations, 4 stations were selected according to the environmental parameters for
170 454 pyrosequencing analyses. The analysis was performed with the samples collected at the
171 dry season (April 2012) because the highest levels of contaminants (particularly PAHs and
172 pesticides) were recorded (supplementary material Table S1), and also because important
173 contrasted conditions were detected between Ichkeul Lake and Bizerte Lagoon (Table 1, and
174 supplementary material, Table S1). Two stations from the Ichkeul Lake (salinity 2.6 ± 0 , pH
175 8.29 ± 0.02 ; supplementary material Table S1) were chosen for contrasted contaminations:
176 ST5, showing multiple contaminations by PAHs and pesticides; and ST8, the most
177 contaminated by OCPs (Table 1; supplementary material, Table S1). Similarly, two stations
178 from the Bizerte Lagoon (salinity 29.5 ± 0.4 , pH 8.37 ± 0.03) were chosen for contrasted

179 contaminations: ST12, the most contaminated by PAHs; and ST17, the less contaminated of
180 the entire complex (Table 1; supplementary material, Table S1). Three replicates for each
181 station were sequenced at Molecular Research LP DNA (MR DNA sequencing service,
182 Shallowater, TX, USA) using Roche 454 FLX titanium as previously described (Dowd et al.,
183 2008; Duran et al., 2015a).

184 QIIME (Quantitative Insights Into Microbial Ecology; Caporaso et al., 2010) was used to
185 analyse the 16S rRNA gene sequence reads as described (Terrisse et al., 2017). Briefly, data
186 cleaning was performed according to Quince et al. (2009) by denoising of the data and by
187 eliminating chimera and sequences with length under 450 bp, with mistakes in primers
188 sequences or homopolymers. OTU picking was performed following the Usearch method
189 (Edgar, 2010) defining reference sequences by applying a 97% similarity threshold. The
190 taxonomic assignment was performed with BLASTn against May 2013 Greengenes database
191 (DeSantis et al., 2006; McDonald et al., 2012) to establish the OTU table. The OTU
192 abundances for each sample were rarefied to the minimum read number observed (12,977
193 sequences) for a sample using the R package vegan (Oksanen et al., 2013; Version: 2.0–10).
194 The complete dataset was deposited in the NCBI Sequence Read Archive (SRA) database
195 (SUB5043721). It is available under the Bioproject ID PRJNA515186.

196 **2.6. Statistical analysis**

197 Canonical Correspondence Analysis (CCA) correlating bacterial community structure (T-
198 RFLP patterns) with chemical data (PAH and pesticide concentrations) and physical-chemical
199 parameters, was performed with MVSP v3.22 software (Kovach Computing Service,
200 Anglesey Wales). T-RFLP profiles were normalized by calculating relative abundances of
201 each TRF, as previously described (Dominique et al., 2007). Data were square root
202 transformed to get a normal distribution of the data (Dollhopf et al., 2001).

203 All statistical analyses based on sequencing data were performed on the random subsample
204 of 12,977 bacterial sequences, corresponding to the smaller number of sequences per sample
205 in the dataset. Alpha diversity indices (Shannon-Wiener H' and richness R), based on
206 rarefied OTU table, were calculated using PRIMER v5.0 (Plymouth Routines in Multivariate
207 Ecological Research). The overall differences of these indices between the stations were
208 tested by ANOVA and Tukey HSD multiple comparisons (pairwise comparisons).
209 Significant difference was assumed when $p < 0.05$. The contribution of individual OTUs
210 explaining the differences between stations was determined by SIMPER (similarity
211 percentages) based on Bray-Curtis distance measure.

212 Linear discriminant analysis effect size (LEfSe; Segata et al., 2011) was performed to
213 determine biomarkers of contamination and habitat as previously described (Jeanbille et al.,
214 2016b). Briefly, the non-parametric Kruskal-Wallis sum-rank test ($\alpha = 0.05$) was performed
215 to detect taxa with significant differential abundance. The biological consistency was
216 investigated by performing a pairwise Wilcoxon test ($\alpha = 0.05$). A LDA threshold score of
217 2.0 and 1000 bootstrap iterations were applied. Heatmap and clustered correlation
218 (Weinstein et al., 1997) were performed with CIMminer
219 (<http://discover.nci.nih.gov/cimminer/>). These analyses were performed at the genus level
220 derived from the OTU abundance table in order to determine the correlations between
221 bacterial genera abundances and pollutant concentrations. Correlation analysis was based on
222 Pearson correlation. The hierarchical clustering of correlations was based on ward method.
223 Strong correlations ($>|0.5|$) were considered. The distribution of OTUs identified in the
224 bacterial patterns of the different stations was visualized with Venn diagrams constructed
225 with Venny using OTU occurrence table as previously described (Duran et al., 2015a). OTUs
226 present at least in two of the triplicates were retained for the analysis.

227

228 **3. Results and discussions**

229 **3.1. Contamination level**

230 The chemical analyses of the sediments collected during September 2011 and April 2012
231 sampling campaigns (supplementary material, Table S1) have been presented in detail in Ben
232 Salem et al. (2017). Table 1 summarizes the chemical data showing the sum of the different
233 types of pollutants obtained in April 2012 for four representative and contrasted stations: 2
234 from the lake (ST5 and ST8) and 2 from the lagoon (ST12 and ST17). The Bizerte Lagoon
235 was characterized by PAH concentrations ranging from 137 ± 26 to $17988 \pm 1183 \text{ ng g}^{-1}$,
236 corresponding to class I (ST12, insignificantly polluted) and class IV (ST17, severely
237 polluted), respectively according to the classification developed by Bakke et al. (2010). Both
238 stations of the Ichkeul Lake (ST5 and ST8) were classified as class II with PAH
239 concentrations of 1362 ± 560 and $444 \pm 24 \text{ ng g}^{-1}$, respectively. Considering the
240 contamination by organochlorine pesticides (OCPs), the Ichkeul Lake stations were classified
241 as class IV (severely polluted) while the Bizerte Lagoon stations were classified as class III
242 (markedly polluted) according to the Bakke et al. (2010) classification. The OCPs, including
243 endrin, dieldrin and lindane, and their derivatives, were detected in the Ichkeul Lake at
244 concentrations ranging from 160 ± 14 to $2021 \pm 85 \text{ ng g}^{-1}$. Additionally, the chemical
245 analyses revealed low PCB contamination of the sediments (ranging from 2.48 ± 0.8 to 5.58
246 $\pm 1 \text{ ng g}^{-1}$). The sediments were classified as class I (insignificantly polluted) except for the
247 station ST8 that was classified as class II (moderately polluted). In order to take into account
248 the multiple contaminations, the mean effect range medium quotients (m-ERM-Q) were
249 calculated (Table 1; supplementary material, Table S1). The m-ERM-Q showed that the toxic
250 potential of the sediments was higher (p value <0.01) in Ichkeul Lake stations than in Bizerte
251 Lagoon stations at both seasons (supplementary material, Table S1). The station were
252 classified according to increasing m-ERM-Q potential toxicity of sediments as follow ST17

253 < ST12 < ST5 < ST8 (Table 1). Station ST17 was considered as lowly contaminated, while
254 stations ST5, ST8 and ST12 were considered as highly contaminated.

255 The chemical data and physical-chemical parameters (supplementary material, Table S1)
256 presented in detail in Ben Salem et al. (2017) were used in the present study to characterize
257 the bacterial community structure according to environmental parameters.

258 **3.2. Bacterial community structure**

259 Correspondence canonical analysis (CCA) using T-RFLP patterns and chemical data
260 separated bacterial communities of the two ecosystems, lake and lagoon irrespective of the
261 season (Fig. 2a). Regarding the environmental parameters, the salinity, the temperature, and
262 the concentrations of nitrite and ammonia were the main factors influencing the bacterial
263 community composition (based on T-RF abundances) of the lagoon, while the bacterial
264 community composition of the lake correlated with dissolved oxygen (DO) and phosphate
265 concentrations. This observation was consistent with previous studies showing that bacterial
266 communities are affected by salinity (Jeanbille et al., 2016a; Jeanbille et al., 2016b; Xi et al.,
267 2014) and nutrients (Bodoque et al., 2017; Liu et al., 2015; Yao et al., 2017). Particularly,
268 nitrite and ammonia concentrations may affect the bacterial populations involved in the
269 nitrogen cycle in lagoon sediments as shown for saline lake sediments (Lipsewers et al., 2016)
270 and marine contaminated sediments (Mahmoudi et al., 2015). The bacterial populations
271 involved in the nitrogen cycle have been shown to play an important role in hydrocarbon
272 removal in marine sediments (Mahmoudi et al., 2015; Stauffert et al., 2016). The CCA also
273 showed that the pH and the concentrations of nitrate and organo-tin compounds influenced
274 slightly the bacterial community compositions probably because they varied within a small
275 range in the studied sediments.

276 Regarding the contaminants, the CCA analysis showed that the bacterial community
277 composition was correlated with pesticide concentrations in the lake, while bacterial

278 community composition was linked with PAHs concentrations in the lagoon (Fig. 2a). These
279 observations confirmed those previously reported (Ben Said et al., 2009) that showed the
280 impact of PAHs on bacteria communities in Bizerte Lagoon sediments. Considering the lake,
281 the bacterial community composition was correlated with PAHs concentrations in April,
282 except for those of station ST8 that was correlated with PCBs and Pesticides concentrations,
283 while bacterial community composition was correlated with the concentration of organo-tin
284 compounds in September (Fig. 2b). This observation suggested seasonal fluctuations in the
285 input of contaminants. It is likely that the lake receives pesticide inputs from crop treatments
286 in April. Additionally, the hydrology of the ecosystem might also play a crucial role: during
287 the dry season (April) lagoon water contaminated by PAHs enters into the lake (Ben Salem et
288 al., 2017). Considering the lagoon, the bacterial community composition was correlated with
289 PAHs and PCBs concentrations in April (Fig. 2c), suggesting that the pollutants were
290 concentrated (concentrating effect) during the dry season. It is likely that the dry season
291 represent a critical season for the Ichkeul Lake / Bizerte Lagoon hydrological system because
292 the environmental conditions favor the concentrating effect of pollutants in both the lake and
293 the lagoon. It is thus of paramount importance to investigate how benthic bacterial
294 communities are affected by such inputs of pollutants. The sediments from the stations ST5
295 and ST8 (lake) and the stations ST12 and ST17 (lagoon) collected in April were retained for
296 further bacterial characterization because they presented different type of contamination while
297 slightly influenced by environmental parameters (Fig. 2b,c; Table 1).

298 **3.3. Bacterial community composition**

299 In order to further investigate the effect of pollutants on bacterial communities at the dry
300 season (April), the composition of bacterial communities was determined by high throughput
301 pyrosequencing. Four stations showing contrasted chemical characteristics were chosen
302 (Table 1): station ST17 (lagoon), the less contaminated; station ST12 (lagoon), the most

303 contaminated with PAHs; station ST8 (lake), the most contaminated with OCPs (pesticides);
304 and station ST5 (lake), contaminated with PAHs and OCPs mixture. Three replicates from
305 each station were analysed. After trimming, the 191,278 sequences (400 bp) retained (12,977
306 sequences per sample after resampling) were dispatched within 858 OTUs, which were
307 grouped into 44 different phyla (Fig. 3). Rarefaction curves (Supplementary data Fig. S1)
308 showed high sequencing coverage in all samples. The coverage (~ 94%) indicated that the
309 sequencing effort was sufficient to assess the bacterial community diversity (Table 2). The
310 sequence affiliation showed that Proteobacteria largely dominated the sediments, with
311 predominance of Deltaproteobacteria (24%), Gammaproteobacteria (23%) and
312 Alphaproteobacteria (9%); Epsilonproteobacteria and Betaproteobacteria accounting for 6%
313 and 5% respectively. The dominance of Proteobacteria, particularly Deltaproteobacteria and
314 Gammaproteobacteria, has been often reported in both lake and lagoon sediments (Borin et
315 al., 2009; Obi et al., 2016; Pavlouidi et al., 2016; Quero et al., 2015; Sun et al., 2013)
316 suggesting that they have a pivotal role in biogeochemical cycles in sediment environments.
317 Besides Proteobacteria, Bacteroidetes (9%), Chloroflexi (6%), Planctomycetes (2%) and
318 Nitrospirae (2%) were the major phyla detected in all four stations (Fig. 3), which was
319 consistent with previous observation in sediment environments (Behera et al., 2017; Fan et al.,
320 2016). These phyla have been detected in freshwater and marine sediments (Behera et al.,
321 2017; Fan et al., 2016; Jacquiod et al., 2018b; Paissé et al., 2012; Pavlouidi et al., 2016).
322 Bacteroidetes are well known for their capacity to degrade organic matter (Hunter et al., 2006;
323 Zeng et al., 2011). They were in high abundance in contaminated sediments (Paisse et al.,
324 2008; Quero et al., 2015; Sun et al., 2013). Chloroflexi have been found as main players in the
325 transformation of organic compounds including organo-chlorines such as PCBs and pesticides
326 (Fagervold et al., 2007; Hug et al., 2013; Maturro et al., 2016a). Planctomycetes, which have
327 the capability to oxidize ammonia aerobically and anaerobically (Junier et al., 2010), have

328 been widely detected in contaminated sediments (Dell'Anno et al., 2012; Quero et al., 2015).
329 Similarly, Nitrospirae members playing an important role in the nitrogen cycle (Daims, 2014)
330 have been detected in contaminated sediments by organic compounds including pesticides and
331 PAHs (Ding et al., 2017; Fernanda Sánchez-Soto Jiménez et al., 2018; Stauffert et al., 2013).
332 Alpha diversity indices were not significantly different between stations (Table 2), an
333 observation that was in contradiction with other studies showing that the presence of
334 pollutants either reduce (Haller et al., 2011; Johnston and Roberts, 2009) or increase (Duran et
335 al., 2015a; Jacquiod et al., 2018a) bacterial diversity. It was thus surprising to observe similar
336 bacterial diversity indexes in sediments presenting different pollutant types and levels. Similar
337 observations have been reported by comparing uncontaminated and contaminated sediments
338 (Paisse et al., 2008; Yin et al., 2015) and soils (Berg et al., 2012). The pollution history,
339 particularly the fluctuant and successive pollution loads resulting in legacy effects, may
340 explain the fact that bacterial diversity was maintained (Jurgburg et al., 2017; Nunes et al.,
341 2018). Also, the coalescence of bacterial communities, mixing bacterial communities from
342 different ecosystems (Mansour et al., 2018; Rillig et al., 2015), here lake and lagoon, may
343 contribute to attenuate the diversity differences between the ecosystem, as previously
344 described in river sediments (Yin et al., 2015). The Venn diagram, showing shared OTUs
345 between lake and lagoon stations (Fig. 4a) evidenced the ecological continuum between the
346 lake and the lagoon, which are closely linked via the Tinja River (Fig. 1). Although similar
347 diversity indexes were observed, the composition of the bacterial diversity was different.
348 Indeed, the Venn diagram revealed that each station was characterized by specific OTUs, only
349 211 OTUs being shared between the stations (Fig. 4a). Such observation was consistent with
350 previous reports on bacterial communities in coastal areas (Duran et al., 2015b; Ikenaga et al.,
351 2010; Misson et al., 2016), indicating that bacterial communities were structured according to
352 the environmental conditions, as revealed by the T-RFLP analyses (Fig. 2). Additionally,

353 linear discriminant analysis effect size (LEfSe) detected taxa differentially abundant
354 according to the ecosystems. LEfSe showed the habitat specificity of genera that were
355 specifically more abundant in either the lake or the lagoon (Fig. 4b). This observation was
356 consistent with previous reports showing habitat specificity for microbial taxa (Jeanbille et al.,
357 2016a; Jeanbille et al., 2016b; Xi et al., 2014). Similarly, LEfSe revealed the presence of
358 specific genera (*Sulfurimonas*, *Acidovorax*, *Pelobacter*, *Sediminibacter*, *Sinobacteraceae*,
359 *Desulfobulbus*, and *Sulfurovum*) in the contaminated stations (Fig. 4c). These observations
360 were further supported by clustering and SIMPER analyses. Clustering analysis, based on
361 Pearson correlation, showed that the lake stations had bacterial communities with different
362 composition to those of the lagoon stations (Fig. 5), which was corroborated by SIMPER
363 analysis (50%). The bacterial communities from the lake stations ST5 and ST8 showed more
364 similar composition (SIMPER: 93%) than those of the lagoon stations ST12 and ST17
365 (SIMPER: 71%). Consistent with LEfSe, the less contaminated station ST17 exhibited
366 specific dominant OTUs affiliated to *Rhodobacter*, *Planctomycetaceae*, *Rhizobium*,
367 *Rhodopirellula*, *Blastopirellula*, *Nitrosococcus* and *Sorangium* genera (Fig. 5), which related
368 members are typically detected in marine ecosystems (Ben Said et al., 2008; Brinkhoff et al.,
369 2012; Hernandez-Raquet et al., 2006; Jasmin et al., 2017; Klotz et al., 2006). Regarding the
370 contaminated stations, the clustering analysis allowed the identification of specific OTUs for
371 each station. The most PAH-contaminated station ST12 was dominated by specific OTUs
372 belonging to *Sediminibacter* and *Loktanella* genera and other genera affiliated to
373 *Rhodobacteraceae*, *Flavobacteriaceae* and *Saprospiraceae* families (Fig. 5), which related
374 members have been detected in marine sediments after oil spill (Acosta-Gonzalez et al., 2015;
375 Galazka et al., 2018; Jimenez et al., 2007). The lake stations ST5 and ST8 were dominated by
376 specific OTUs related to *Thiobacillus*, *Nitrospira* and *Acidovorax* genera, *Escherichia*-
377 *Shigella* group, and other genera affiliated to *Gemmatimonadaceae*, *Nitrospiraceae*,

378 *Caldilineaceae* and *Desulfarculaceae* families (Fig. 5), which was consistent with LEfSe (Fig.
379 4). Related members of these taxa have been found in sediments contaminated by
380 hydrocarbons (Jochum et al., 2018; Patel et al., 2015; Reyes-Sosa et al., 2018) and pesticides
381 (Douglass et al., 2015; Jiang et al., 2018).

382 Although LEfSe revealed that the abundance of some taxa were different between the lake
383 and lagoon ecosystems (Fig. 4), the clustering analysis showed that the four stations shared
384 dominant OTUs affiliated to *Acidithiobacillus*, *Desulfobulbus*, *Sinobacteraceae*, *Robiginitalea*,
385 *Anaerolineaceae* and *Nannocystineae* (Fig. 5). Members of the *Acidithiobacillus* genera have
386 been detected in metals contaminated ecosystems (Giloteaux et al., 2010), particularly in
387 sediments (Fonti et al., 2013). It is not surprising to detect metal-resistant bacteria in the
388 Ichkeul Lake / Bizerte Lagoon hydrological system since high levels of metal contents have
389 been detected in the sediments (Ben Said et al., 2009; Zaaboub et al., 2015). The detection of
390 members related to *Desulfobulbus*, *Robiginitalea*, *Sinobacteraceae*, *Anaerolineaceae* and
391 *Nannocystineae* encompassing marine species, further support the ecological continuum
392 between the Ichkeul Lake and Bizerte Lagoon. Among them some have been found in
393 contaminated sediments bearing the capacity to degrade hydrocarbons (Li et al., 2017; Yan et
394 al., 2018), and to resist and transform metals (Meng et al., 2019; Wang et al., 2018).

395 **3.4. Influence of contamination on bacterial assemblages**

396 In order to further decipher the associations between bacterial genera and environmental
397 parameters clustered correlation (ClusCor) analysis was performed at the genus level derived
398 from the OTU abundance table (Fig. 6). First, sequences related to 11 genera including
399 *Sulfurovum*, *Pseudomonas*, *Leptospirillum*, *Sulfurimonas*, *Acidovorax*, and *Nitrospira*
400 genera, and genera related to *Anaerolineaceae*, *Desulfarculaceae*, *Caldilineaceae*,
401 *Gemmatimonadaceae*, and *Nitrospiraceae* compose the Group1a. The abundance of the
402 Group1a genera showed strong ($0.6 > \text{Pearson correlation} < 0.8$) to very strong (Pearson

403 correlation > 0.8) positive correlations with the concentration of the different pesticides
404 (PCBs, Lindane, and other OCPs), and strong (-0.6 > Pearson correlation > -0.8) to very strong
405 (Pearson correlation < -0.8) negative correlation (exclusion) with PAHs concentrations. This
406 Group 1a was closely related with the Group 1b including *Desulfobulbus* and *Thiobacillus*
407 genera, and genera related to *Nannocystineae*, *Sinobacteraceae*, and *Escherichia-Shigella*,
408 which abundances showed strong (0.6 > Pearson correlation < 0.8) to very strong (Pearson
409 correlation > 0.8) positive correlations with the concentrations of pesticide compounds and
410 strong negative correlation (-0.6 > Pearson correlation > -0.8, i.e. exclusion) with the other
411 pollutants. Consistently, LEfSe identified all these genera (Group 1a and Group 1b)
412 specifically abundant for either the lake or the contamination (Fig. 4). Several *Pseudomonas*
413 and *Acidovorax* species have been isolated for their ability to degrade pesticides and other
414 organic compounds in diverse habitats (Marican and Duran-Lara, 2018; Song et al., 2000).
415 Interestingly, the presence of the sulfur-oxidizing bacteria related to the *Sulfurovum* and
416 *Sulfurimonas* genera suggested the potential anaerobic degradation of pesticides since these
417 genera have been described for their capacity to perform reductive dechlorination of PCB in
418 marine sediments (Maturro et al., 2016b), often associated with sulfate-reducing bacteria
419 (SRB). Also, both genera, *Sulfurovum* and *Sulfurimonas*, have been shown to be able to grow
420 under microaerophilic conditions using oxygen or nitrate as electron acceptor (Mitchell et al.,
421 2014; Xu et al., 2017). It is likely that these genera in association with the SRB
422 *Desulfarculaceae* related genera and *Desulfobulbus* in the Group 1 assemblage might play a
423 role in the sulfur cycle. They probably use the hydrogen sulfur (H₂S) produced by SRB and
424 oxygen for their growth playing a role in the degradation of pesticides. The genera related to
425 the *Nitrospiraceae* have been found dominant in contaminated aquatic ecosystems:
426 *Nitrospira* dominant in organic matter contaminated waters (Huang et al., 2017; Wang et al.,
427 2016), while *Leptospirillum* in the presence of metals and metalloids (Wang et al., 2013).

428 Also, the presence of *Nitrospira* together with *Thiobacillus*, *Gemmatimonadaceae*, and the
429 genera of the phylum Chloroflexi (*Anaerolineaceae* and *Caldilineaceae*), denitrifying genera,
430 suggested a contamination from wastewater treatment systems as previously reported
431 (Aanderud et al., 2018; Zhang et al., 2017), and further supported by the presence of
432 *Escherichia-Shigella* and *Sinobacteraceae* related genera (Zhang et al., 2018). Related
433 members of the *Nannocystineae* genera have been described harbouring the capacity to
434 degrade complex macromolecules (Garcia and Müller, 2013). They have been detected in
435 polluted sediments (Acosta-González et al., 2013), but their potential for pollutant
436 degradation still to be demonstrated.

437 The Group2 included the genera related to *Flavobacteriaceae*, *Flammeovirgaceae*,
438 *Saprospiraceae*, *Rhodobacteraceae* families, and the genera *Actibacter*, *Sediminibacter*,
439 *Robiginitalea*, *Loktanella*, *Pelobacter*, *Desulfuromonas*. The abundances of Group2 genera
440 showed very strong positive correlation with the concentrations of PAHs and organic-SN
441 compounds (Pearson correlation >0.8), and strong (-0.6 > Pearson correlation > -0.8) to very
442 strong (Pearson correlation < -0.8) negative correlations (exclusion) with the concentrations of
443 lindane and other OCPs. These genera, which abundance was correlated to either the lagoon
444 or the contamination level by LEfSe (Fig. 4), have been found in hydrocarbon contaminated
445 marine sediments (Acosta-Gonzalez et al., 2015; Chen See et al., 2018; Galazka et al., 2018)
446 and oil production facilities (Vigneron et al., 2016; Zapata-Peñasco et al., 2013). Particularly
447 interesting, the presence of members related to the *Desulfuromonadaceae* family (*Pelobacter*
448 and *Desulfuromonas*) in this Group2 assemblage that have been described in presence of
449 pesticide and for their capacity of reductive dechlorination (Greene, 2014; Sercu et al., 2013).
450 Such observation suggested that these genera might play a key role to cope with the multi-
451 contamination in lagoon sediments.

452 Behind these two major groups, the ClusCor analyses identified bacterial assemblages
453 (Group3) including genera, which abundances showed mainly strong ($-0.6 > \text{Pearson}$
454 $\text{correlation} > -0.8$) to very strong ($\text{Pearson correlation} < -0.8$) negative correlations (exclusion)
455 with pollutant contents (Fig. 6). The Group3a included genera which abundance showed
456 exclusion with PCBs and PAHs. The abundance of genera from Group3b showed exclusion
457 with the content of all pesticides except heptachlor and strong positive correlation
458 ($0.6 < \text{Pearson correlation} < 0.8$) with PAHs content (Fig.6). The Group3c correspond to genera
459 which abundance showed exclusion with PCBs content, while the abundance of genera from
460 Group3d showed exclusion with PCBs and lindane contents (Fig. 6). Noteworthy, most of the
461 genera included in Group3c (*Candidatus-Thiobios*, *Rhizobium*, *Desulfocapsa*,
462 *Planctomycetaceae*, *Rhodopirellula*, and *Blastopirellula*) showed exclusion with pollutants,
463 which was consistent with LEfSe highlighting that the abundance of these genera was
464 correlated with low contamination level (Fig. 4c). It is likely that these genera were sensitive
465 to the presence of pollutants suggesting that their presence might report on ecosystem quality.
466 However, further investigations are requested to understand their role in the ecosystem
467 functioning.

468 The co-occurrence analyses showed specific bacterial assemblages linked with contaminants
469 revealing possible metabolic capacities of bacterial genera. Our results are consistent with
470 previous reports that revealed specific populations linked with pollutants in marine
471 ecosystems (Duran et al., 2015a), in freshwater lakes (Warnecke et al., 2005), and in soils
472 (Llado et al., 2015). Such information provides new insights useful to guide future research to
473 answer important questions in the field of microbial ecology such as the role of specific
474 populations in the ecosystem functioning and pollutants degradation, and the role of
475 environmental parameters in driving bacterial assemblages.

476 **4. Conclusions**

477 The determination of bacterial composition in sediment of the Ichkeul Lake / Bizerte Lagoon
478 hydrological system showed that the lake bacterial communities were distinct from those of
479 the lagoon. Although the difference could be attributed to physical-chemical parameters, the
480 salinity and the presence of pollutants playing an important role in structuring the bacterial
481 communities, the study revealed that the lake and the lagoon shared some bacterial genera.
482 Such result suggested a lake-lagoon continuum due to the hydrology of the system
483 exchanging waters according to the season resulting in bacterial community coalescence. It is
484 likely that such bacterial genera ensure universal biogeochemical functions and, therefore
485 constitute "generalists" bacteria essential for the basic ecosystem functioning. The study also
486 revealed "specialists" bacteria either for the habitat (lake or lagoon) or for the pollutants.
487 Bacterial "specialists" able to cope with the presence of pesticides in the lake and bacterial
488 "specialists" for PAHs and organic forms of Sn in the Lagoon. These findings represent
489 valuable information, particularly the bacterial "specialists", which represent yet an
490 unexplored metabolic potential for the development of bioremediation processes to mitigate
491 the effect of pollutants in aquatic ecosystems. The study also revealed bacterial genera, which
492 abundance showed exclusion (negative correlation), with the pollutants concentrations. They
493 represent potential bacterial biomarkers reporting on ecosystem quality, which will be useful
494 to develop bacterial tools for monitoring the effect of countermeasures taken for mitigating
495 the pollution.

496 **Acknowledgements**

497 The authors would like to thank Mme Nabiha M'barek (National Agency of Environment
498 Protection Tunisia) and Mr. Taher Kaabi (Commissariat Regional Commission for
499 Agricultural Development of Bizerte) for their logistic contribution. We acknowledge the
500 Regional Platform for Environmental Microbiology PREMICE supported by the Aquitaine
501 Regional Government Council and the urban community of Pau-Pyrénées.

502

503 **Funding information**

504 This study was funded by the European programme ERANET-MED through the
505 AQUASALT (NMED-0003-01) project.

506

507 **Compliance with ethical standards**

508 **Conflict of interest**

509 The authors declare that they have no conflict of interest.

510 **Ethical statement**

511 This article does not contain any studies with human participants or animals performed by any
512 of the authors.

513

514 **References**

- 515
- 516 Aanderud, Z.T., Saurey, S., Ball, B.A., Wall, D.H., Barrett, J.E., Muscarella, M.E., Griffin,
517 N.A., Virginia, R.A., Adams, B.J., 2018. Stoichiometric Shifts in Soil C:N:P Promote
518 Bacterial Taxa Dominance, Maintain Biodiversity, and Deconstruct Community
519 Assemblages. *Front Microbiol* 9, 1401.
- 520 Acosta-Gonzalez, A., Martirani-von Abercron, S.M., Rossello-Mora, R., Wittich, R.M.,
521 Marques, S., 2015. The effect of oil spills on the bacterial diversity and catabolic function in
522 coastal sediments: a case study on the Prestige oil spill. *Environ Sci Pollut R* 22, 15200-
523 15214.
- 524 Acosta-González, A., Rosselló-Móra, R., Marqués, S., 2013. Characterization of the
525 anaerobic microbial community in oil-polluted subtidal sediments: aromatic biodegradation
526 potential after the Prestige oil spill. *Environmental Microbiology* 15, 77-92.
- 527 Andrews, D.M., Lin, H., Zhu, Q., Jin, L.X., Brantley, S.L., 2011. Hot Spots and Hot Moments
528 of Dissolved Organic Carbon Export and Soil Organic Carbon Storage in the Shale Hills
529 Catchment. *Vadose Zone J* 10, 943-954.
- 530 Bakke, T., Källqvist, T., Ruus, A., Breedveld, G.D., Hylland, K., 2010. Development of
531 sediment quality criteria in Norway. *Journal of Soils and Sediments* 10, 172-178.
- 532 Barberan, A., Bates, S.T., Casamayor, E.O., Fierer, N., 2012. Using network analysis to
533 explore co-occurrence patterns in soil microbial communities. *ISME J* 6, 343-351.
- 534 Behera, P., Mahapatra, S., Mohapatra, M., Kim, J.Y., Adhya, T.K., Raina, V., Suar, M.,
535 Pattnaik, A.K., Rastogi, G., 2017. Salinity and macrophyte drive the biogeography of the
536 sedimentary bacterial communities in a brackish water tropical coastal lagoon. *Science of the*
537 *Total Environment* 595, 472-485.
- 538 Ben Said, O., Goñi-Urriza, M., El Bour, M., Aissa, P., Duran, R., 2009. Bacterial Community
539 Structure of Sediments of the Bizerte Lagoon (Tunisia), a Southern Mediterranean Coastal
540 Anthropized Lagoon. *Microb Ecol* 59, 445-456.
- 541 Ben Said, O., Goni-Urriza, M.S., El Bour, M., Dellali, M., Aissa, P., Duran, R., 2008.
542 Characterization of aerobic polycyclic aromatic hydrocarbon-degrading bacteria from Bizerte
543 lagoon sediments, Tunisia. *J Appl Microbiol* 104, 987-997.
- 544 Ben Said, O., Louati, H., Soltani, A., Preud'homme, H., Cravo-Laureau, C., Got, P., Pringault,
545 O., Aissa, P., Duran, R., 2015. Changes of benthic bacteria and meiofauna assemblages
546 during bio-treatments of anthracene-contaminated sediments from Bizerta lagoon (Tunisia).
547 *Environ Sci Pollut R* 22, 15319-15331.
- 548 Ben Salem, F., Ben Said, O., Aissa, P., Mahmoudi, E., Monperrus, M., Grunberger, O.,
549 Duran, R., 2016a. Pesticides in Ichkeul Lake-Bizerta Lagoon Watershed in Tunisia: use,
550 occurrence, and effects on bacteria and free-living marine nematodes. *Environ Sci Pollut R*
551 23, 36-48.
- 552 Ben Salem, F., Ben Said, O., Duran, R., Monperrus, M., 2016b. Validation of an Adapted
553 QuEChERS Method for the Simultaneous Analysis of Polycyclic Aromatic Hydrocarbons,
554 Polychlorinated Biphenyls and Organochlorine Pesticides in Sediment by Gas
555 Chromatography-Mass Spectrometry. *Bull Environ Contam Toxicol* 96, 678-684.
- 556 Ben Salem, F., Ben Said, O., Mahmoudi, E., Duran, R., Monperrus, M., 2017. Distribution of
557 organic contamination of sediments from Ichkeul Lake and Bizerte Lagoon, Tunisia. *Mar*
558 *Pollut Bull* 123, 329-338.
- 559 Berg, J., Brandt, K.K., Al-Soud, W.A., Holm, P.E., Hansen, L.H., Sørensen, S.J., Nybroe, O.,
560 2012. Selection for Cu-tolerant bacterial communities with altered composition, but unaltered
561 richness, via long-term cu exposure. *Applied and Environmental Microbiology* 78, 7438-
562 7446.

563 Bodoque, J.M., Ladera, J., Yela, J.L., Alonso-Azcárate, J., Brito, D., Antigüedad, I., Duran,
564 R., Attard, E., Lauga, B., Sánchez-Pérez, J.M., 2017. Recovering hydromorphological
565 functionality to improve natural purification capacity of a highly human-modified wetland.
566 *Ecological Engineering* 103, 332-343.

567 Bordenave, S., Fourcans, A., Blanchard, S., Goni, M.S., Caumette, P., Duran, R., 2004a.
568 Structure and functional analyses of bacterial communities changes in microbial mats
569 following petroleum exposure. *Ophelia* 58, 195-203.

570 Bordenave, S., Goni-Urriza, M., Vilette, C., Blanchard, S., Caumette, P., Duran, R., 2008.
571 Diversity of ring-hydroxylating dioxygenases in pristine and oil contaminated microbial mats
572 at genomic and transcriptomic levels. *Environ Microbiol* 10, 3201-3211.

573 Bordenave, S., Jézéquel, R., Fourçans, A., Budzinski, H., Merlin, F.X., Fourel, T., Goñi-
574 Urriza, M., Guyoneaud, R., Grimaud, R., Caumette, P., Duran, R., 2004b. Degradation of
575 the "Erika" oil. *Aquat Living Resour* 17, 261-267.

576 Borin, S., Brusetti, L., Daffonchio, D., Delaney, E., Baldi, F., 2009. Biodiversity of
577 prokaryotic communities in sediments of different sub-basins of the Venice lagoon. *Research*
578 *in Microbiology* 160, 307-314.

579 Botta, F., Lavison, G., Couturier, G., Alliot, F., Moreau-Guigon, E., Fauchon, N., Guery, B.,
580 Chevreuil, M., Blanchoud, H., 2009. Transfer of glyphosate and its degradate AMPA to
581 surface waters through urban sewerage systems. *Chemosphere* 77, 133-139.

582 Brinkhoff, T., Fischer, D., Vollmers, J., Voget, S., Beardsley, C., Thole, S., Musmann, M.,
583 Kunze, B., Wagner-Dobler, I., Daniel, R., Simon, M., 2012. Biogeography and phylogenetic
584 diversity of a cluster of exclusively marine myxobacteria. *ISME J* 6, 1260-1272.

585 Brito, E.M., Duran, R., Guyoneaud, R., Goni-Urriza, M., Garcia de Oteyza, T., Crapez, M.A.,
586 Aleluia, I., Wasserman, J.C., 2009. A case study of in situ oil contamination in a mangrove
587 swamp (Rio De Janeiro, Brazil). *Mar Pollut Bull* 58, 418-423.

588 Caporaso, J.G., Kuczynski, J., Stombaugh, J., Bittinger, K., Bushman, F.D., Costello, E.K.,
589 Fierer, N., Pena, A.G., Goodrich, J.K., Gordon, J.I., Huttley, G.A., Kelley, S.T., Knights, D.,
590 Koenig, J.E., Ley, R.E., Lozupone, C.A., McDonald, D., Muegge, B.D., Pirrung, M., Reeder,
591 J., Sevinsky, J.R., Tumbaugh, P.J., Walters, W.A., Widmann, J., Yatsunenko, T., Zaneveld, J.,
592 Knight, R., 2010. QIIME allows analysis of high-throughput community sequencing data. *Nat*
593 *Methods* 7, 335-336.

594 Casagrande, C., Boudouresque, C.F., 2010. A first quantification of the overall biomass, from
595 phytoplankton to birds, of a Mediterranean brackish lagoon: Revisiting the ecosystem of Lake
596 Ichkeul (Tunisia). *Hydrobiologia* 637, 73-85.

597 Chang, S.M., Doong, R.A., 2006. Concentration and fate of persistent organochlorine
598 pesticides in estuarine sediments using headspace solid-phase microextraction. *Chemosphere*
599 62, 1869-1878.

600 Chen See, J.R., Ulrich, N., Nwanosike, H., McLimans, C.J., Tokarev, V., Wright, J.R.,
601 Campa, M.F., Grant, C.J., Hazen, T.C., Niles, J.M., Ressler, D., Lamendella, R., 2018.
602 Bacterial Biomarkers of Marcellus Shale Activity in Pennsylvania. *Front Microbiol* 9, 1697.

603 Chronopoulou, P.M., Fahy, A., Coulon, F., Paise, S., Goni-Urriza, M., Peperzak, L., Alvarez,
604 L.A., McKew, B.A., Lawson, T., Timmis, K.N., Duran, R., Underwood, G.J.C., McGenity,
605 T.J., 2013. Impact of a simulated oil spill on benthic phototrophs and nitrogen-fixing bacteria
606 in mudflat mesocosms. *Environ Microbiol* 15, 242-252.

607 Cravo-Laureau, C., Cagnon, C., Lauga, B., Duran, R., 2017a. Preface. *Microbial*
608 *Ecotoxicology*, v-vi.

609 Cravo-Laureau, C., Duran, R., 2014. Marine coastal sediments microbial hydrocarbon
610 degradation processes: contribution of experimental ecology in the omics'era. *Front Microbiol*
611 5, 39.

612 Cravo-Laureau, C., Hernandez-Raquet, G., Vitte, I., Jezequel, R., Bellet, V., Godon, J.J.,
613 Caumette, P., Balaguer, P., Duran, R., 2011. Role of environmental fluctuations and microbial
614 diversity in degradation of hydrocarbons in contaminated sludge. *Res Microbiol* 162, 888-
615 895.

616 Cravo-Laureau, C., Lauga, B., Cagnon, C., Duran, R., 2017b. Microbial responses to
617 pollution- ecotoxicology: Introducing the different biological levels, *Microbial*
618 *Ecotoxicology*, pp. 45-62.

619 Daims, H., 2014. The family nitrospiraceae, *The Prokaryotes: Other Major Lineages of*
620 *Bacteria and The Archaea*, pp. 733-749.

621 Dell'Anno, A., Beolchini, F., Rocchetti, L., Luna, G.M., Danovaro, R., 2012. High bacterial
622 biodiversity increases degradation performance of hydrocarbons during bioremediation of
623 contaminated harbor marine sediments. *Environmental Pollution* 167, 85-92.

624 DeSantis, T.Z., Hugenholtz, P., Larsen, N., Rojas, M., Brodie, E.L., Keller, K., Huber, T.,
625 Dalevi, D., Hu, P., Andersen, G.L., 2006. Greengenes, a chimera-checked 16S rRNA gene
626 database and workbench compatible with ARB. *Applied and Environmental Microbiology* 72,
627 5069-5072.

628 Ding, Q., Huang, X., Hu, H., Hong, M., Zhang, D., Wang, K., 2017. Impact of pyrene and
629 cadmium co-contamination on prokaryotic community in coastal sediment microcosms.
630 *Chemosphere* 188, 320-328.

631 Dollhopf, S.L., Hashsham, S.A., Tiedje, J.M., 2001. Interpreting 16S rDNA T-RFLP data:
632 Application of self-organizing maps and principal component analysis to describe community
633 dynamics and convergence. *Microbial Ecology* 42, 495-505.

634 Dominique, Y., Muresan, B., Duran, R., Richard, S., Boudou, A., 2007. Simulation of the
635 chemical fate and bioavailability of liquid elemental mercury drops from gold mining in
636 Amazonian freshwater systems. *Environmental Science and Technology* 41, 7322-7329.

637 Doong, R.A., Lee, S.H., Lee, C.C., Sun, Y.C., Wu, S.C., 2008. Characterization and
638 composition of heavy metals and persistent organic pollutants in water and estuarine
639 sediments from Gao-ping River, Taiwan. *Mar Pollut Bull* 57, 846-857.

640 Douglass, J.F., Radosevich, M., Tuovinen, O.H., 2015. Molecular analysis of atrazine-
641 degrading bacteria and catabolic genes in the water column and sediment of a created wetland
642 in an agricultural/urban watershed. *Ecol Eng* 83, 405-412.

643 Dowd, S.E., Callaway, T.R., Wolcott, R.D., Sun, Y., McKeenan, T., Hagevoort, R.G.,
644 Edrington, T.S., 2008. Evaluation of the bacterial diversity in the feces of cattle using 16S
645 rDNA bacterial tag-encoded FLX amplicon pyrosequencing (bTEFAP). *BMC Microbiol* 8,
646 125.

647 Duran, R., Bielen, A., Paradzik, T., Gassie, C., Pustijanac, E., Cagnon, C., Hamer, B.,
648 Vujaklija, D., 2015a. Exploring Actinobacteria assemblages in coastal marine sediments
649 under contrasted Human influences in the West Istria Sea, Croatia. *Environ Sci Pollut R* 22,
650 15215-15229.

651 Duran, R., Bonin, P., Jezequel, R., Dubosc, K., Gassie, C., Terrisse, F., Abella, J., Cagnon, C.,
652 Milton, C., Michotey, V., Gilbert, F., Cuny, P., Cravo-Laureau, C., 2015b. Effect of physical
653 sediments reworking on hydrocarbon degradation and bacterial community structure in
654 marine coastal sediments. *Environ Sci Pollut R* 22, 15248-15259.

655 Duran, R., Cravo-Laureau, C., 2016. Role of environmental factors and microorganisms in
656 determining the fate of polycyclic aromatic hydrocarbons in the marine environment. *FEMS*
657 *Microbiol Rev* 40, 814-830.

658 Duran, R., Cuny, P., Bonin, P., Cravo-Laureau, C., 2015c. Microbial ecology of hydrocarbon-
659 polluted coastal sediments. *Environ Sci Pollut R* 22, 15195-15199.

660 Edgar, R.C., 2010. Search and clustering orders of magnitude faster than BLAST.
661 *Bioinformatics* 26, 2460-2461.

662 Fagervold, S.K., May, H.D., Sowers, K.R., 2007. Microbial reductive dechlorination of
663 Aroclor 1260 in Baltimore Harbor sediment microcosms is catalyzed by three phylotypes
664 within the Phylum chloroflexi. *Applied and Environmental Microbiology* 73, 3009-3018.

665 Fan, L., Barry, K., Hu, G., Meng, S., Song, C., Qiu, L., Zheng, Y., Wu, W., Qu, J., Chen, J.,
666 Xu, P., 2016. Characterizing bacterial communities in tilapia pond surface sediment and their
667 responses to pond differences and temporal variations. *World Journal of Microbiology and*
668 *Biotechnology* 33, 1.

669 Fernanda Sánchez-Soto Jiménez, M., Cerqueda-García, D., Montero-Muñoz, J.L., Leopoldina
670 Aguirre-Macedo, M., García-Maldonado, J.Q., 2018. Assessment of the bacterial community
671 structure in shallow and deep sediments of the Perdido Fold Belt region in the Gulf of
672 Mexico. *PeerJ* 2018.

673 Fonti, V., Dell'Anno, A., Beolchini, F., 2013. Influence of biogeochemical interactions on
674 metal bioleaching performance in contaminated marine sediment. *Water Res* 47, 5139-5152.

675 Fourcans, A., Ranchou-Peyruse, A., Caumette, P., Duran, R., 2008. Molecular analysis of the
676 spatio-temporal distribution of sulfate-reducing bacteria (SRB) in Camargue (France)
677 hypersaline microbial mat. *Microb Ecol* 56, 90-100.

678 Fuhrman, J.A., Steele, J.A., 2008. Community structure of marine bacterioplankton: patterns,
679 networks, and relationships to function. *Aquat Microb Ecol* 53, 69-81.

680 Galazka, A., Grzadziel, J., Galazka, R., Ukalska-Jaruga, A., Strzelecka, J., Smreczak, B.,
681 2018. Genetic and Functional Diversity of Bacterial Microbiome in Soils With Long Term
682 Impacts of Petroleum Hydrocarbons. *Front Microbiol* 9, 1923.

683 Garcia, R., Müller, R., 2013. The family nannocystaceae, *The Prokaryotes: Deltaproteobacteria*
684 *and Epsilonproteobacteria*, pp. 213-229.

685 Gharbi-Bouraoui, S., Gnassia-Barelli, M., Roméo, M., Dellali, M., Aïssa, P., 2008. Field
686 study of metal concentrations and biomarker responses in the neogastropod, *Murex trunculus*,
687 from Bizerta Lagoon (Tunisia). *Aquatic Living Resources* 21, 213-220.

688 Giloteaux, L., Goni-Urriza, M., Duran, R., 2010. Nested PCR and new primers for analysis of
689 sulfate-reducing bacteria in low-cell-biomass environments. *Appl Environ Microbiol* 76,
690 2856-2865.

691 Greene, A.C., 2014. The family desulfuromonadaceae, *The Prokaryotes: Deltaproteobacteria*
692 *and Epsilonproteobacteria*, pp. 143-155.

693 Guermouche M'rassi, A., Bensalah, F., Gury, J., Duran, R., 2015. Isolation and
694 characterization of different bacterial strains for bioremediation of n-alkanes and polycyclic
695 aromatic hydrocarbons. *Environ Sci Pollut R* 22, 15332-15346.

696 Haller, L., Tonolla, M., Zopfi, J., Peduzzi, R., Wildi, W., Pote, J., 2011. Composition of
697 bacterial and archaeal communities in freshwater sediments with different contamination
698 levels (Lake Geneva, Switzerland). *Water Res* 45, 1213-1228.

699 Hernandez-Raquet, G., Budzinski, H., Caumette, P., Dabert, P., Le Menach, K., Muyzer, G.,
700 Duran, R., 2006. Molecular diversity studies of bacterial communities of oil polluted
701 microbial mats from the Etang de Berre (France). *FEMS Microbiol Ecol* 58, 550-562.

702 Huang, W., Chen, X., Jiang, X., Zheng, B.H., 2017. Characterization of sediment bacterial
703 communities in plain lakes with different trophic statuses. *MicrobiologyOpen* 6.

704 Hug, L.A., Castelle, C.J., Wrighton, K.C., Thomas, B.C., Sharon, I., Frischkorn, K.R.,
705 Williams, K.H., Tringe, S.G., Banfield, J.F., 2013. Community genomic analyses constrain
706 the distribution of metabolic traits across the Chloroflexi phylum and indicate roles in
707 sediment carbon cycling. *Microbiome* 1.

708 Hunter, E.M., Mills, H.J., Kostka, J.E., 2006. Microbial community diversity associated with
709 carbon and nitrogen cycling in permeable shelf sediments. *Applied and Environmental*
710 *Microbiology* 72, 5689-5701.

711 Ikenaga, M., Guevara, R., Dean, A.L., Pisani, C., Boyer, J.N., 2010. Changes in Community
712 Structure of Sediment Bacteria Along the Florida Coastal Everglades Marsh-Mangrove-
713 Seagrass Salinity Gradient. *Microb Ecol* 59, 284-295.

714 Jacquiod, S., Cyriaque, V., Riber, L., Al-soud, W.A., Gillan, D.C., Wattiez, R., Sørensen, S.J.,
715 2018a. Long-term industrial metal contamination unexpectedly shaped diversity and activity
716 response of sediment microbiome. *Journal of Hazardous Materials* 344, 299-307.

717 Jacquiod, S., Nunes, I., Brejnrod, A., Hansen, M.A., Holm, P.E., Johansen, A., Brandt, K.K.,
718 Priemé, A., Sørensen, S.J., 2018b. Long-term soil metal exposure impaired temporal variation
719 in microbial metatranscriptomes and enriched active phages. *Microbiome* 6.

720 Jasmin, C., Anas, A., Tharakan, B., Jaleel, A., Puthiyaveetil, V., Narayanane, S., Lincy, J.,
721 Nair, S., 2017. Diversity of sediment-associated Planctomycetes in the Arabian Sea oxygen
722 minimum zone. *J Basic Microbiol* 57, 1010-1017.

723 Jeanbille, M., Gury, J., Duran, R., Tronczynski, J., Agogue, H., Ben Said, O., Ghiglione, J.F.,
724 Auguet, J.C., 2016a. Response of Core Microbial Consortia to Chronic Hydrocarbon
725 Contaminations in Coastal Sediment Habitats. *Front Microbiol* 7, 1637.

726 Jeanbille, M., Gury, J., Duran, R., Tronczynski, J., Ghiglione, J.F., Agogue, H., Said, O.B.,
727 Taib, N., Debros, D., Garnier, C., Auguet, J.C., 2016b. Chronic Polyaromatic Hydrocarbon
728 (PAH) Contamination Is a Marginal Driver for Community Diversity and Prokaryotic
729 Predicted Functioning in Coastal Sediments. *Front Microbiol* 7, 1303.

730 Jiang, L., Luo, C., Zhang, D., Song, M., Sun, Y., Zhang, G., 2018. Biphenyl-Metabolizing
731 Microbial Community and a Functional Operon Revealed in E-Waste-Contaminated Soil.
732 *Environ Sci Technol* 52, 8558-8567.

733 Jimenez, N., Vinas, M., Bayona, J.M., Albaiges, J., Solanas, A.M., 2007. The Prestige oil
734 spill: bacterial community dynamics during a field biostimulation assay. *Appl Microbiol*
735 *Biotechnol* 77, 935-945.

736 Jochum, L.M., Schreiber, L., Marshall, I.P.G., Jorgensen, B.B., Schramm, A., Kjeldsen, K.U.,
737 2018. Single-Cell Genomics Reveals a Diverse Metabolic Potential of Uncultivated
738 Desulfatiglans-Related Deltaproteobacteria Widely Distributed in Marine Sediment. *Front*
739 *Microbiol* 9.

740 Johnston, E.L., Roberts, D.A., 2009. Contaminants reduce the richness and evenness of
741 marine communities: a review and meta-analysis. *Environ Pollut* 157, 1745-1752.

742 Junier, P., Molina, V., Dorador, C., Hadas, O., Kim, O.S., Junier, T., Witzel, K.P., Imhoff,
743 J.F., 2010. Phylogenetic and functional marker genes to study ammonia-oxidizing
744 microorganisms (AOM) in the environment. *Applied Microbiology and Biotechnology* 85,
745 425-440.

746 Jurgburg, S.D., Nunes, I., Brejnrod, A., Jacquiod, S., Priemé, A., Sørensen, S.J., Van Elsas,
747 J.D., Salles, J.F., 2017. Legacy Effects on the Recovery of Soil Bacterial Communities from
748 Extreme Temperature Perturbation. *Frontiers in Microbiology* 8.

749 Kjellerup, B.V., Naff, C., Edwards, S.J., Ghosh, U., Baker, J.E., Sowers, K.R., 2014. Effects
750 of activated carbon on reductive dechlorination of PCBs by organohalide respiring bacteria
751 indigenous to sediments. *Water Res* 52, 1-10.

752 Klotz, M.G., Arp, D.J., Chain, P.S.G., El-Sheikh, A.F., Hauser, L.J., Hommes, N.G., Larimer,
753 F.W., Malfatti, S.A., Norton, J.M., Poret-Peterson, A.T., Vergez, L.M., Ward, B.B., 2006.
754 Complete Genome Sequence of the Marine, Chemolithoautotrophic, Ammonia-Oxidizing
755 Bacterium *Nitrosococcus oceani* ATCC 19707. *Appl Environ Microbiol* 72, 6299-6315.

756 Li, J., Zhang, D., Song, M., Jiang, L., Wang, Y., Luo, C., Zhang, G., 2017. Novel bacteria
757 capable of degrading phenanthrene in activated sludge revealed by stable-isotope probing
758 coupled with high-throughput sequencing. *Biodegradation* 28, 423-436.

759 Lipsewers, Y.A., Hopmans, E.C., Meysman, F.J.R., Sinninghe Damsté, J.S., Villanueva, L.,
760 2016. Abundance and Diversity of Denitrifying and Anammox Bacteria in Seasonally
761 Hypoxic and Sulfidic Sediments of the Saline Lake Grevelingen. *Frontiers in Microbiology* 7.
762 Liu, S., Ren, H., Shen, L., Lou, L., Tian, G., Zheng, P., Hu, B., 2015. pH levels drive bacterial
763 community structure in sediments of the Qiantang River as determined by 454
764 pyrosequencing. *Frontiers in microbiology* 6, 285-285.
765 Llado, S., Covino, S., Solanas, A.M., Petruccioli, M., D'Annibale, A., Vinas, M., 2015.
766 Pyrosequencing reveals the effect of mobilizing agents and lignocellulosic substrate
767 amendment on microbial community composition in a real industrial PAH-polluted soil. *J*
768 *Hazard Mater* 283, 35-43.
769 Long, E.R., Field, L.J., MacDonald, D.D., 1998. Predicting toxicity in marine sediments with
770 numerical sediment quality guidelines. *Environmental Toxicology and Chemistry* 17, 714-
771 727.
772 Louati, H., Ben Said, O., Got, P., Soltani, A., Mahmoudi, E., Cravo-Laureau, C., Duran, R.,
773 Aissa, P., Pringault, O., 2013a. Microbial community responses to bioremediation treatments
774 for the mitigation of low-dose anthracene in marine coastal sediments of Bizerte lagoon
775 (Tunisia). *Environ Sci Pollut R* 20, 300-310.
776 Louati, H., Said, O.B., Soltani, A., Got, P., Mahmoudi, E., Cravo-Laureau, C., Duran, R.,
777 Aissa, P., Pringault, O., 2013b. The roles of biological interactions and pollutant
778 contamination in shaping microbial benthic community structure. *Chemosphere* 93, 2535-
779 2546.
780 Mahmoudi, N., Robeson, M.S., II, Castro, H.F., Fortney, J.L., Techtmann, S.M., Joyner, D.C.,
781 Paradis, C.J., Pfiffner, S.M., Hazen, T.C., 2015. Microbial community composition and
782 diversity in Caspian Sea sediments. *FEMS Microbiology Ecology* 91.
783 Mansour, I., Heppell, C.M., Ryo, M., Rillig, M.C., 2018. Application of the microbial
784 community coalescence concept to riverine networks. *Biological Reviews* 93, 1832-1845.
785 Marchesi, J.R., Sato, T., Weightman, A.J., Martin, T.A., Fry, J.C., Hiom, S.J., Wade, W.G.,
786 1998. Design and evaluation of useful bacterium-specific PCR primers that amplify genes
787 coding for bacterial 16S rRNA. *Appl Environ Microbiol* 64, 795-799.
788 Marican, A., Duran-Lara, E.F., 2018. A review on pesticide removal through different
789 processes. *Environ Sci Pollut R* 25, 2051-2064.
790 Matturro, B., Presta, E., Rossetti, S., 2016a. Reductive dechlorination of tetrachloroethene in
791 marine sediments: Biodiversity and dehalorespiring capabilities of the indigenous microbes.
792 *Science of the Total Environment* 545-546, 445-452.
793 Matturro, B., Ubaldi, C., Rossetti, S., 2016b. Microbiome Dynamics of a Polychlorobiphenyl
794 (PCB) Historically Contaminated Marine Sediment under Conditions Promoting Reductive
795 Dechlorination. *Front Microbiol* 7, 1502.
796 McDonald, D., Price, M.N., Goodrich, J., Nawrocki, E.P., Desantis, T.Z., Probst, A.,
797 Andersen, G.L., Knight, R., Hugenholtz, P., 2012. An improved Greengenes taxonomy with
798 explicit ranks for ecological and evolutionary analyses of bacteria and archaea. *ISME Journal*
799 6, 610-618.
800 Meng, D., Li, J., Liu, T., Liu, Y., Yan, M., Hu, J., Li, X., Liu, X., Liang, Y., Liu, H., Yin, H.,
801 2019. Effects of redox potential on soil cadmium solubility: Insight into microbial
802 community. *J Environ Sci (China)* 75, 224-232.
803 Misson, B., Garnier, C., Lauga, B., Dang, D.H., Ghiglione, J.F., Mullet, J.U., Duran, R.,
804 Pringault, O., 2016. Chemical multi-contamination drives benthic prokaryotic diversity in the
805 anthropized Toulon Bay. *Sci Total Environ* 556, 319-329.
806 Mitchell, H.M., Rocha, G.A., Kaakoush, N.O., O'Rourke, J.L., Queiroz, D.M.M., 2014. The
807 family helicobacteraceae, *The Prokaryotes: Deltaproteobacteria and Epsilonproteobacteria*,
808 pp. 337-392.

809 Mwanamoki, P.M., Devarajan, N., Thevenon, F., Birane, N., de Alencastro, L.F., Grandjean,
810 D., Mpiana, P.T., Prabakar, K., Mubedi, J.I., Kabele, C.G., Wildi, W., Pote, J., 2014. Trace
811 metals and persistent organic pollutants in sediments from river-reservoir systems in
812 Democratic Republic of Congo (DRC): Spatial distribution and potential ecotoxicological
813 effects. *Chemosphere* 111, 485-492.

814 Nunes, I., Jurgburg, S., Jacquiod, S., Brejnrod, A., Falcão Salles, J., Priemé, A., Sørensen, S.J.,
815 2018. Soil bacteria show different tolerance ranges to an unprecedented disturbance. *Biology
816 and Fertility of Soils* 54, 189-202.

817 Obi, C.C., Adebusoye, S.A., Ugoji, E.O., Ilori, M.O., Amund, O.O., Hickey, W.J., 2016.
818 *Microbial Communities in Sediments of Lagos Lagoon, Nigeria: Elucidation of Community
819 Structure and Potential Impacts of Contamination by Municipal and Industrial Wastes.*
820 *Frontiers in Microbiology* 7.

821 Oksanen, J., Blanchet, F., Kindt, R., Legendre, P., Minchin, P., O'Hara, R., Simpson, G.,
822 Solymos, P., Henry, M., Stevens, H., 2013. *Vegan: Community Ecology Package.* 2013. R-
823 package version 2.0-10. URL <http://CRAN.R-project.org/package=vegan>.

824 Paisse, S., Coulon, F., Goni-Urriza, M., Peperzak, L., McGenity, T.J., Duran, R., 2008.
825 Structure of bacterial communities along a hydrocarbon contamination gradient in a coastal
826 sediment. *FEMS Microbiol Ecol* 66, 295-305.

827 Paissé, S., Goñi-Urriza, M., Stadler, T., Budzinski, H., Duran, R., 2012. Ring-hydroxylating
828 dioxygenase (RHD) expression in a microbial community during the early response to oil
829 pollution. *FEMS Microbiology Ecology* 80, 77-86.

830 Patel, V., Munot, H., Shah, V., Shouche, Y.S., Madamwar, D., 2015. Taxonomic profiling of
831 bacterial community structure from coastal sediment of Alang-Sosiya shipbreaking yard near
832 Bhavnagar, India. *Mar Pollut Bull* 101, 736-745.

833 Pavloudi, C., Oulas, A., Vasileiadou, K., Sarropoulou, E., Kotoulas, G., Arvanitidis, C., 2016.
834 Salinity is the major factor influencing the sediment bacterial communities in a Mediterranean
835 lagoonal complex (Amvrakikos Gulf, Ionian Sea). *Marine Genomics* 28, 71-81.

836 Pérez-Domínguez, R., MacI, S., Courrat, A., Lepage, M., Borja, A., Uriarte, A., Neto, J.M.,
837 Cabral, H., St.raykov, V., Franco, A., Alvarez, M.C., Elliott, M., 2012. Current developments
838 on fish-based indices to assess ecological-quality status of estuaries and lagoons. *Ecological
839 Indicators* 23, 34-45.

840 Precigou, S., Goulas, P., Duran, R., 2001. Rapid and specific identification of nitrile hydratase
841 (NHase)-encoding genes in soil samples by polymerase chain reaction. *FEMS Microbiol Lett*
842 204, 155-161.

843 Precigou, S., Wieser, M., Pommars, P., Goulas, P., Duran, R., 2004. *Rhodococcus
844 pyridinovorans* MW3, a bacterium producing a nitrile hydratase. *Biotechnol Lett* 26, 1379-
845 1384.

846 Pringault, O., Duran, R., Jacquet, S., Torreton, J.P., 2008. Temporal variations of microbial
847 activity and diversity in marine tropical sediments (New Caledonia lagoon). *Microb Ecol* 55,
848 247-258.

849 Quero, G.M., Cassin, D., Botter, M., Perini, L., Luna, G.M., 2015. Patterns of benthic
850 bacterial diversity in coastal areas contaminated by heavy metals, polycyclic aromatic
851 hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs). *Frontiers in Microbiology* 6.

852 Quince, C., Lanzen, A., Curtis, T.P., Davenport, R.J., Hall, N., Head, I.M., Read, L.F., Sloan,
853 W.T., 2009. Accurate determination of microbial diversity from 454 pyrosequencing data. *Nat
854 Methods* 6, 639-641.

855 Ramdani, M., Flower, R.J., Elkhiaiti, N., Kraïem, M.M., Fathi, A.A., Birks, H.H., Patrick,
856 S.T., 2001. North African wetland lakes: Characterization of nine sites included in the
857 CASSARINA Project. *Aquat Ecol* 35, 281-302.

858 Reyes-Sosa, M.B., Apodaca-Hernandez, J.E., Arena-Ortiz, M.L., 2018. Bioprospecting for
859 microbes with potential hydrocarbon remediation activity on the northwest coast of the
860 Yucatan Peninsula, Mexico, using DNA sequencing. *Sci Total Environ* 642, 1060-1074.

861 Rillig, M.C., Antonovics, J., Caruso, T., Lehmann, A., Powell, J.R., Veresoglou, S.D.,
862 Verbruggen, E., 2015. Interchange of entire communities: Microbial community coalescence.
863 *Trends in Ecology and Evolution* 30, 470-476.

864 Schultz, P., Urban, N.R., 2008. Effects of bacterial dynamics on organic matter
865 decomposition and nutrient release from sediments: A modeling study. *Ecol Model* 210, 1-14.

866 Segata, N., Izard, J., Waldron, L., Gevers, D., Miropolsky, L., Garrett, W.S., Huttenhower, C.,
867 2011. Metagenomic biomarker discovery and explanation. *Genome Biol* 12.

868 Sercu, B., Jones, A.D., Wu, C.H., Escobar, M.H., Serlin, C.L., Knapp, T.A., Andersen, G.L.,
869 Holden, P.A., 2013. The influence of in situ chemical oxidation on microbial community
870 composition in groundwater contaminated with chlorinated solvents. *Microb Ecol* 65, 39-49.

871 Song, B., Palleroni, N.J., Haggblom, M.M., 2000. Isolation and characterization of diverse
872 halobenzoate-degrading denitrifying bacteria from soils and sediments. *Appl Environ*
873 *Microbiol* 66, 3446-3453.

874 Stauffert, M., Cravo-Laureau, C., Duran, R., 2016. Structure of hydrocarbonoclastic nitrate-
875 reducing bacterial communities in bioturbated coastal marine sediments. *FEMS Microbiology*
876 *Ecology* 89, 580-593.

877 Stauffert, M., Cravo-Laureau, C., Jezequel, R., Barantal, S., Cuny, P., Gilbert, F., Cagnon, C.,
878 Militon, C., Amouroux, D., Mahdaoui, F., Bouyssiére, B., Stora, G., Merlin, F.X., Duran, R.,
879 2013. Impact of oil on bacterial community structure in bioturbated sediments. *PLoS One* 8,
880 e65347.

881 Stauffert, M., Duran, R., Gassie, C., Cravo-Laureau, C., 2014. Response of archaeal
882 communities to oil spill in bioturbated mudflat sediments. *Microb Ecol* 67, 108-119.

883 Stevenson, A., Battarbee, R., 1991. Palaeoecological and documentary records of recent
884 environmental change in Garaet El Ichkeul: a seasonally saline lake in NW Tunisia. *Biol*
885 *Conserv* 58, 275-295.

886 Sun, M.Y., Dafforn, K.A., Johnston, E.L., Brown, M.V., 2013. Core sediment bacteria drive
887 community response to anthropogenic contamination over multiple environmental gradients.
888 *Environmental Microbiology* 15, 2517-2531.

889 Terrisse, F., Cravo-Laureau, C., Noel, C., Cagnon, C., Dumbrell, A.J., McGenity, T.J., Duran,
890 R., 2017. Variation of Oxygenation Conditions on a Hydrocarbonoclastic Microbial
891 Community Reveals *Alcanivorax* and *Cycloclasticus* Ecotypes. *Front Microbiol* 8.

892 Thiyagarajan, V., Tsoi, M.M., Zhang, W., Qian, P.Y., 2010. Temporal variation of coastal
893 surface sediment bacterial communities along an environmental pollution gradient. *Mar*
894 *Environ Res* 70, 56-64.

895 Trabelsi, S., Driss, M.R., 2005. Polycyclic aromatic hydrocarbons in superficial coastal
896 sediments from Bizerte Lagoon, Tunisia. *Mar Pollut Bull* 50, 344-348.

897 Vasconcelos, R.P., Reis-Santos, P., Costa, M.J., Cabral, H.N., 2011. Connectivity between
898 estuaries and marine environment: Integrating metrics to assess estuarine nursery function.
899 *Ecological Indicators* 11, 1123-1133.

900 Vercaene-Eairmal, M., Lauga, B., Saint Laurent, S., Mazzella, N., Boutry, S., Simon, M.,
901 Karama, S., Delmas, F., Duran, R., 2010. Diuron biotransformation and its effects on biofilm
902 bacterial community structure. *Chemosphere* 81, 837-843.

903 Vigneron, A., Alsop, E.B., Chambers, B., Lomans, B.P., Head, I.M., Tsesmetzis, N., 2016.
904 Complementary Microorganisms in Highly Corrosive Biofilms from an Offshore Oil
905 Production Facility. *Appl Environ Microbiol* 82, 2545-2554.

906 Vitte, I., Duran, R., Jezequel, R., Caumette, P., Cravo-Laureau, C., 2011. Effect of
907 oxic/anoxic switches on bacterial communities and PAH biodegradation in an oil-
908 contaminated sludge. *Environ Sci Pollut R* 18, 1022-1032.

909 Wang, J., Li, Y., Wang, P., Niu, L., Zhang, W., Wang, C., 2016. Response of bacterial
910 community compositions to different sources of pollutants in sediments of a tributary of Taihu
911 Lake, China. *Environ Sci Pollut R* 23, 13886-13894.

912 Wang, J., Zhao, H.B., Zhuang, T., Qin, W.Q., Zhu, S., Qiu, G.Z., 2013. Bioleaching of Pb-
913 Zn-Sn chalcopyrite concentrate in tank bioreactor and microbial community succession
914 analysis. *T Nonferr Metal Soc* 23, 3758-3762.

915 Wang, S., Zhang, B.G., Diao, M.H., Shi, J.X., Jiang, Y.F., Cheng, Y.T., Liu, H., 2018.
916 Enhancement of synchronous bio-reductions of vanadium (V) and chromium (VI) by mixed
917 anaerobic culture. *Environ Pollut* 242, 249-256.

918 Warnecke, F., Sommaruga, R., Sekar, R., Hofer, J.S., Pernthaler, J., 2005. Abundances,
919 identity, and growth state of actinobacteria in mountain lakes of different UV transparency.
920 *Appl Environ Microbiol* 71, 5551-5559.

921 Weinstein, J.N., Myers, T.G., OConnor, P.M., Friend, S.H., Fornace, A.J., Kohn, K.W., Fojo,
922 T., Bates, S.E., Rubinstein, L.V., Anderson, N.L., Buolamwini, J.K., vanOsdol, W.W.,
923 Monks, A.P., Scudiero, D.A., Sausville, E.A., Zaharevitz, D.W., Bunow, B., Viswanadhan,
924 V.N., Johnson, G.S., Wittes, R.E., Paull, K.D., 1997. An information-intensive approach to
925 the molecular pharmacology of cancer. *Science* 275, 343-349.

926 Wen, J.G., Jirka, G.H., Raggio, G., 1994. Linked Sediment/Contaminant Transport Model for
927 Rivers with Application to the Buffalo River, New-York. *J Great Lakes Res* 20, 671-682.

928 Williams, R.J., Howe, A., Hofmockel, K.S., 2014. Demonstrating microbial co-occurrence
929 pattern analyses within and between ecosystems. *Front Microbiol* 5, 358.

930 Xi, X.F., Wang, L., Hu, J.J., Tang, Y.S., Hu, Y., Fu, X.H., Sun, Y., Tsang, Y.F., Zhang, Y.N.,
931 Chen, J.H., 2014. Salinity influence on soil microbial respiration rate of wetland in the
932 Yangtze River estuary through changing microbial community. *J Environ Sci-China* 26, 2562-
933 2570.

934 Xu, X.J., Chen, C., Guan, X., Yuan, Y., Wang, A.J., Lee, D.J., Zhang, Z.F., Zhang, J., Zhong,
935 Y.J., Ren, N.Q., 2017. Performance and microbial community analysis of a microaerophilic
936 sulfate and nitrate co-reduction system. *Chemical Engineering Journal* 330, 63-70.

937 Yan, Z., Hao, Z., Wu, H., Jiang, H., Yang, M., Wang, C., 2018. Co-occurrence patterns of the
938 microbial community in polycyclic aromatic hydrocarbon-contaminated riverine sediments. *J*
939 *Hazard Mater* 367, 99-108.

940 Yao, J., Sánchez-Pérez, J.M., Sauvage, S., Teissier, S., Attard, E., Lauga, B., Duran, R.,
941 Julien, F., Bernard-Jannin, L., Ramburn, H., Gerino, M., 2017. Biodiversity and ecosystem
942 purification service in an alluvial wetland. *Ecological Engineering* 103, 359-371.

943 Yergeau, E., Lawrence, J.R., Sanschagrin, S., Waiser, M.J., Korber, D.R., Greer, C.W., 2012.
944 Next-generation sequencing of microbial communities in the Athabasca River and its
945 tributaries in relation to oil sands mining activities. *Appl Environ Microbiol* 78, 7626-7637.

946 Yin, H., Niu, J., Ren, Y., Cong, J., Zhang, X., Fan, F., Xiao, Y., Zhang, X., Deng, J., Xie, M.,
947 He, Z., Zhou, J., Liang, Y., Liu, X., 2015. An integrated insight into the response of
948 sedimentary microbial communities to heavy metal contamination. *Scientific Reports* 5,
949 14266.

950 Zaaboub, N., Martins, M.V., Dhib, A., Bejaoui, B., Galgani, F., El Bour, M., Aleya, L., 2015.
951 Accumulation of trace metals in sediments in a Mediterranean Lagoon: Usefulness of metal
952 sediment fractionation and elutriate toxicity assessment. *Environ Pollut* 207, 226-237.

953 Zapata-Peñasco, I., Salazar-Coria, L., Saucedo-García, M., Villa-Tanaka, L., Hernández-
954 Rodríguez, C., 2013. Bisulfite reductase and nitrogenase genes retrieved from biocorrosive

955 bacteria in saline produced waters of offshore oil recovery facilities. *Int Biodeterior*
956 *Biodegrad* 81, 17-27.
957 Zeng, Y., Zou, Y., Chen, B., Grebmeier, J.M., Li, H., Yu, Y., Zheng, T., 2011. Phylogenetic
958 diversity of sediment bacteria in the northern Bering Sea. *Polar Biology* 34, 907-919.
959 Zhang, B., Xu, X., Zhu, L., 2017. Structure and function of the microbial consortia of
960 activated sludge in typical municipal wastewater treatment plants in winter. *Sci Rep* 7, 17930.
961 Zhang, B., Xu, X., Zhu, L., 2018. Activated sludge bacterial communities of typical
962 wastewater treatment plants: distinct genera identification and metabolic potential differential
963 analysis. *AMB Express* 8, 184.

964

965

966 **Figure legends**

967

968 **Figure 1. Map of the Ichkeul Lake / Bizerte Lagoon hydrological system.** The sampling
969 stations (ST, red) are indicated by numbers.

970

971 **Figure 2. Comparison of bacterial communities from stations of the Ichkeul Lake /**
972 **Bizerte Lagoon hydrological system.** The canonical correspondence analysis shows the
973 correlations between bacterial communities structures (T-RFLP patterns) and environmental
974 chemical data in a) the Bizerte Lagoon-Ichkeul Lake complex, b) in Ichkeul Lake sediments,
975 and c) in Bizerte Lagoon sediments. The analyses were performed in September 2011 (wet
976 season) and April 2012 (dry season). Lake September, pink circles; Lake April, blue circles;
977 Lagoon September, red diamonds; Lagoon April brown diamonds. The analyses are based on
978 biological triplicates for each station. Σ Pesticides, total pesticides; Σ PAHs, total PAHs; Σ
979 PCBs, total PCB; Σ Sn, total Sn organic compounds; T°, temperature; m-ERM-Q, mean
980 effect range medium quotient; P, phosphate; DO, dissolved oxygen. The names of the stations
981 further characterized in this study are indicated ST5 and ST8 for the Ichkeul Lake, and ST12
982 and ST17 for the Bizerte Lagoon.

983

984 **Figure 3. Composition of bacterial communities from stations of the Ichkeul Lake (ST5**
985 **and ST8) and Bizerte Lagoon (ST12 and ST17) hydrological system.** The analysis was
986 performed at the phylum level applying a threshold similarity of 97 % for OTU identification.
987 The average of three replicates was used for the analysis. Other, correspond to phyla with
988 relative abundance < 5%.

989

990 **Figure 4. Comparison of bacterial communities from the different stations of the Ichkeul**
991 **Lake / Bizerte Lagoon hydrological system.** a) Venn diagram showing the number of shared
992 OTUs between stations as well as the specific OTUs for each station. b) Linear discriminant
993 analysis effect size (LEfSe) of the habitat (lake / lagoon). c) LEfSe analysis of the
994 contamination. LEfSe analysis shows the significant differential abundance distribution of
995 bacterial genera between the samples of environmental groups (lake / lagoon; high

996 contamination / low contamination). The analysis was performed applying a threshold
997 similarity of 97 % for OTU identification. The contamination level (high / low) refers to mean
998 effect range medium quotients (m-ERM-Q) presented in Table 1.

999

1000 **Figure 5. Heatmap comparing the bacterial communities from the different stations of**
1001 **the Ichkeul Lake / Bizerte Lagoon hydrological system.** The heatmap with clustering
1002 analysis based on Pearson correlation shows the repartition of the 50 most abundant genera.
1003 The analysis was performed applying a threshold similarity of 97 % for OTU identification.
1004 The triplicates for each station are shown in the heatmap.

1005

1006 **Figure 6. Clustered correlation (ClusCor) between bacterial OTUs and pollutants**
1007 **parameters.** The analysis is based on Pearson correlation, red indicating high positive
1008 correlation and blue high negative correlation (p value <0.01). Clustering was performed
1009 applying Pearson correlations and Ward algorithm as cluster method. Abbreviations of
1010 pollutants are those indicated in Table 1. The analysis was performed at the genus level
1011 applying a threshold similarity of 97 % for OTU identification. The specific OTUs revealed
1012 by LEfSe analysis are coloured according to their specificity: lake (khaki), lagoon (red), high
1013 contamination (orange), and low contamination (blue).

1014

1015 **Table 1: Chemical analyses of contaminants in the Ichkeul Lake / Bizerte Lagoon**
 1016 **hydrological system and sediment toxicity potential.** The data corresponds to the sum of
 1017 the different types of pollutants obtained in April 2012 described in details by Ben Salem et
 1018 al. (2017). Average \pm SD are presented (n=3).

Pollutants (ng g ⁻¹ dry wt)	Stations			
	ST5 Lake	ST8	ST12 Lagoon	ST17
Organic Sn^a	143 \pm 0.3	83 \pm 0.3	154 \pm 0.1	102 \pm 0.5
PCBs^b	4.66 \pm 0.7 (I) ^h	5.58 \pm 1 (II) ^h	4.54 \pm 0.1 (I) ^h	2.48 \pm 0.8 (I) ^h
Heptachlor	10.7 \pm 0.7	14 \pm 1.6	11 \pm 4	12 \pm 4
Lindane^d	268 \pm 6	283 \pm 13	101 \pm 5	95 \pm 14
Other OCPs^e	586 \pm 0.03 (IV) ^h	1717 \pm 65 (IV) ^h	53 \pm 3.4 (III) ^h	86 \pm 7.4 (III) ^h
PAHs^f	1362 \pm 560 (II) ^h	444 \pm 24 (II) ^h	17988 \pm 1183 (IV) ^h	137 \pm 26 (I) ^h
m-ERM-Q^g	8.73 \pm 0.03	13.30 \pm 0.4	5.30 \pm 0.06	3.90 \pm 0.1

1019 ^asum of organic forms of tin including methyl, butyl, and phenyl derivatives.
 1020 ^bSum of PCB congeners including PCB18, PCB28, PCB31, PCB44, PCB52, PCB101, PCB138, PCB118,
 1021 PCB153, PCB149, PCB180, PCB194.
 1022 ^cSum of heptachlor and its epoxide derivative.
 1023 ^dSum of lindane and its metabolites.
 1024 ^eSum of endosulfan, dieldrin, aldrin, chlordane, and their derivatives.
 1025 ^fSum of PAHs including naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene,
 1026 fluoranthene, pyrene, benzo[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene,
 1027 benzo[a]pryrene, indenol[1,2,3-cd]pyrene, dibenz[a,h]anthracene, and benzoghiperylene.
 1028 ^gm-ERM-Q, mean effect range medium quotient calculated according to (Long et al., 1998).
 1029 ^hQuality classification according to Bakke et al. (2010).
 1030

1031 **Table 2: Alpha diversity indices (mean \pm SD, n=3) of microbial community from the**
 1032 **Ichkeul Lake / Bizerte Lagoon stations**

Indices	Station			
	ST5 Lake	ST8 Lake	ST12 Lagoon	ST17 Lagoon
Reads^a	22368	19247	22101	25662
Trimmed sequences^b	16378	14217	15185	18502
Species richness (<i>R</i>)	347 \pm 32	359 \pm 28	341 \pm 27	356 \pm 40
Coverage	0.93 \pm 0.01	0.93 \pm 0.01	0.94 \pm 0.01	0.94 \pm 0.01
Simpson (<i>I-D</i>)	0.97 \pm 0.01	0.98 \pm 0.01	0.96 \pm 0.01	0.98 \pm 0.01
Shannon (<i>H</i>)	4.51 \pm 0.1	4.58 \pm 0.1	4.37 \pm 0.1	4.51 \pm 0.05
Evenness (<i>E</i>)	0.26 \pm 0.04	0.27 \pm 0.01	0.23 \pm 0.03	0.26 \pm 0.02
Chao-1	373 \pm 39	386 \pm 27	362 \pm 27	379 \pm 45

1033 ^aSD: ST5, \pm 1966; ST8, \pm 1866; ST12, \pm 3380; ST17, \pm 1688.

1034 ^bSD: ST5, \pm 1481; ST8, \pm 1406; ST12, \pm 2708; ST17, \pm 1611.













