1 Bacterioplankton dynamics driven by inter-annual variation in phytoplankton spring bloom

2 communities in the Baltic Sea

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15 Running title: Bacterioplankton dynamics and phytoplankton composition in the Baltic Sea

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26   **Abstract**

27   In the Baltic Sea, climate change has caused shifts in the phytoplankton spring bloom

28   communities with co-occurrence of diatoms and dinoflagellates. Such changes likely affect

29   the composition and function of associated bacterioplankton, key members of the carbon

30   cycling, although the actual effects are unknown. To understand how changes in

31   phytoplankton impact on bacterioplankton composition and function, we analysed

32   bacterioplankton communities and their production during different phases of the spring

33   bloom in four consecutive years across the Baltic Sea, and related them to environmental

34   variables. Phytoplankton communities varied largely in composition, modifying the

35   taxonomic structure and richness of the associated bacterioplankton assemblages. In presence

36   of certain diatoms (*Achnanthes taeniata, Skeletonema costatum* and *Chaetoceros* spp.),

37   bacterial production and diversity were high and with more relative abundance of

38   Flavobacteriia, Gammaproteobacteria and Betaproteobacteria. This bacterial community

39   structure correlated positively with high diatom biomass and with high bacterial production

40   rates. In contrast, during dinoflagellate-dominated blooms or when the diatom *Thalassiosira*

41   *baltica* was abundant, both bacterial production rates and diversity were low, with bacterial

42   communities dominated by SAR11 and Rhodobacteraceae. Our results demonstrate that,

43   changes in the phytoplankton spring bloom will have profound consequences on bacterial

44   community structure and their role in carbon cycling.

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51   **Introduction**

52   The Baltic Sea is suffering from a progressive increase in surface water temperature, which

53   has been suggested to alter the food web structure, favoring small flagellates and

54   dinoflagellates [1]. During the last decades, shifting phytoplankton spring bloom communities

55   from diatom-dominated blooms towards higher abundances of dinoflagellates have been

56   reported in some subbasins of the Baltic Sea [2-4], likely due to the increase in frequency of

57   milder winters [1,5]. However, the consequences of these changes in spring blooms on the

58   structure and functioning of the associated bacterioplankton communities are still poorly

59   understood.

60   The nature and the timing of the spring bloom in the Baltic Sea vary between sub-basins [6].

61   The bloom typically starts with the light promoting net primary production in the

62   southernmost Baltic Sea in February/March, reaching the Gulf of Finland in April, and the

63   Gulf of Bothnia in May (Map in Fig. 1). The bloom reaches the peak at the time when

64   inorganic nutrients have been depleted; N-limitation prevails in most of the Baltic Sea except

65   for the P-limited Bay of Bothnia [7-9]. The subsequent decline phase is characterized by rapid

66   sinking of the phytoplankton cells [10].

67   Diatoms and dinoflagellates have different ecological features, as diatoms grow faster

68   than co-occurring dinoflagellates [11], they also differ in terms of quality and quantity of

69   dissolved organic matter (DOM) release [12-14], and have different sedimentation patterns [10,15].

70   Since bacteria are the main consumers of the DOM produced by phytoplankton, also in the

71   Baltic Sea [1,16], understanding how the predicted changes in the spring bloom may impact

72   bacterial community composition and activity is essential to anticipate the biogeochemical

73   consequences of this global warming driven process.

74   Assessing these issues in the Baltic Sea is complex due to the large environmental and

75   spatial-temporal heterogeneity between subbasins, i.e. the pronounced salinity (2 - 20) and

76 temperature gradients (0 - 20°C) [8]. Thus, the taxonomic composition of bacteria varies along

77 the salinity gradient with Alphaproteobacteria and Gammaproteobacteria dominating in more

78 saline waters whereas Actinobacteria, Betaproteobacteria and Planctomycetes are favored by

79 lower salinity [17-19]. The strong seasonality in the Baltic Sea also shapes bacterial community

80 structure and dynamics: Alphaproteobacteria are present throughout the year [16,19],

81 Bacteroidetes, Beta- and Gammaproteobacteria are associated with phytoplankton spring

82 blooms [17,20,21] and Verrucomicrobia, Actinobacteria and Planctomycetes appear later in the

83 year [18,22]. Although it is clear that both physical and chemical properties affect bacterial

84 communities, most of these studies have been restricted to specific sub-basins or did not

85 capture the full development of the spring bloom. The potential of diatoms to shape the

86 bacterial communities towards the dominance of Gamma-, Betaproteobacteria and

87 Bacteriodetes and to increase heterotrophic production in the Baltic Sea was recently

88 demonstrated a in mesocosm study [23]. However, so far no study has addressed this issue in

89 natural conditions or across the naturally occurring environmental gradients in the Baltic Sea.

90 The aim of this study was to investigate how differences in the taxonomic composition of

91 phytoplankton spring bloom communities (diatom- or dinoflagellate-dominated), affect the

92 community structure and dynamics of the associated bacterioplankton across different areas

93 of the Baltic Sea. We collected samples from four consecutive years (2013–2016) during and

94 after the spring bloom, from the northern Gulf of Bothnia to the southern Baltic Proper.

95 Based on our previous observations from laboratory studies [23], we hypothesized that the

96 predicted increase in dinoflagellates may lead to less productive bacterial communities

97 dominated by less copiotrophic bacterial groups.

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99 **Material and Methods**

100 **Study area, sampling design and environmental variables**

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101 The water samples were collected during four cruises (2013–2016) on board of the R/V

102 Aranda. In total, 127 stations were sampled and the area spanned from the southern Baltic

103 Proper (BP, 55°22'N) to the northern Bay of Bothnia (BoB, 65°53'N, Fig 1). At all stations,

104 the water was collected from 3 m depth either using an oceanographic rosette with Niskin

105 bottles of 5 L (n = 122) or from the flow-through system on board (n = 5). Salinity,

106 temperature, and depth, were recorded *in situ*. Inorganic nutrients, measured as nitrogen

107 (NO_2+NO_3-N, NH_4-N), phosphorus (PO_4-P), and dissolved silica (DSi), particulate organic

108 carbon (POC), biogenic silicate (BSi), dissolved organic carbon (DOC), dissolved organic

109 nitrogen (DON) and chlorophyll *a* (Chl *a*) concentrations were measured as described by

110 Camarena-Gómez, et al. [23]. The particulate organic carbon to chlorophyll *a* (POC:Chl *a*)

111 ratio, based on weight ($\mu g\ L^{-1}$), was calculated as an indicator of the trophic status

112 (autotrophic or heterotrophic) of the system, considering heterotrophic status those values

113 larger than 200 [24].

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115 **Plankton community composition, carbon biomass and productivity**

116 Samples for the determination of nano- and microplankton, including phytoplankton, were

117 determined as described by Lipsewers and Spilling [25]. A modified (carbon biomass instead of

118 biovolume) diatom/dinoflagellate (Dia/Dino) index = diatom biomass/diatom + dinoflagellate

119 biomass, was calculated for each station according to Wasmund, et al. [26]. The values higher

120 than 0.5 indicate diatom dominance whereas values lower than 0.5 indicate dinoflagellate

121 dominance. Primary production (PP) was estimated by measuring the incorporation of ^{14}C-

122 labelled sodium bicarbonate (DHI, Denmark) according to Gargas [27] and following the

123 method described in Camarena-Gómez, et al. [23].

124   The percent extracellular release of ^{14}C (PER) was calculated based on the dissolved

125 organic fraction ($DO^{14}C < 0.2\ \mu m$) of the total ^{14}C fixation after 24 h incubation. Since

126   substantial heterotrophic consumption of DOC (on average, 30–50 %) might take place

127   during the incubation period [28], the results are regarded as net DOC production rates.

128   Four different stages of the spring bloom were defined based on the concentration of

129   inorganic nutrients (PO_4^{3-} and $NO_3^-+NO_2^-$) and Chl a (Table S1): the bloom growth phase

130   (Growth), the peak of the bloom phase (Peak), the declining bloom phase (Decline) and after

131   the bloom phase (Postbloom). The $NO_3^-+NO_2^-$ was used for the N-limited sub-basins Gulf of

132   Finland (GoF), BP, Archipelago Sea (ArS), Åland Sea (ÅS) and Bothnian Sea (BS); and

133   PO_4^{3-} for the P-limited BoB subbasin. High Chl a was defined as within 20 % of the average

134   peak concentration during spring, which is different in the different sub-basins.

135

Bacterial heterotrophic production, abundance and community composition

137   Bacterial cell production (BPT) and protein production (BPL) were measured in triplicates by

138   using the dual labeling with [metil-^{3}H]-thymidine and [^{14}C (U)]-leucine (PerkinElmer Inc.,

139   Waltham, MA, USA) of samples processed by the centrifugation method as described in

140   Camarena-Gómez, et al. [23]. Bacterial abundances (BA) were determined in duplicates by flow

141   cytometry according to Gasol and Del Giorgio [29], applying some modifications of the method

142   described in Camarena-Gómez, et al. [23]. Bacterial community analysis including DNA

143   extraction and *Illumina* sequencing were done as described by Camarena-Gómez, et al. [23].

144   The post-processing of the sequences was done according to the pipeline described by

145   Logares [30]. The Operational Taxonomic Units (OTUs) were clustered at 99 % of similarity.

146   After the removal of singletons as well as the OTUs classified as chloroplasts and

147   mitochondria, based on the classification with SILVA v123, 2 247 OTUs and 2 250 000 reads

148   were retained. Samples with less than 7 000 reads (4 samples) were removed. The remaining

149   samples (122) were rarefied using *rrarefy* in R to the lowest number of reads (n = 7 024

150   reads). Downstream analyses were carried out in the rarefied table, including 122 samples

151 and 2 128 OTUs. Raw sequence data will be deposited in the European Nucleotide Archive

152 (acc. nun. XXXXX).

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154 **Statistical analyses**

155 To assess whether there were differences in the diatom and dinoflagellate carbon biomass,

156 between the different phytoplankton bloom phases a non-parametric Wilcoxon rank sum test

157 was applied. This test was also applied to the bacterial diversity in order to test whether there

158 were differences between the two main bacterial community clusters observed. Non-metric

159 multidimensional scaling (NMDS) analysis was done on the Bray-Curtis dissimilarity matrix.

160 Mantel tests with Spearman correlation were applied to study the correlation between the

161 composition of bacterial and phytoplankton communities. Redundancy analysis (RDA) with

162 forward selection was applied to obtain the most important environmental variables that

163 explain bacterial community variance. This analysis included an ANOVA test in each step

164 (permutation test for RDA under reduced model, permutations: free, number of permutations:

165 9 999) to identify the significant environmental variables ($p < 0.05$). Spearman correlation

166 was used to obtain the correlation between the main bacterial groups and the significant

167 environmental variables as well as the bacterial activity. All figures and analyses were

168 performed with the software RStudio (RStudio Team 2015) with the packages *colorRamps,*

169 *plotrix, rworldmap, rworldxtra*, *vegan, ggplot2*, *ade4*, *ggpubr* and *Hmisc* and

170 *ComplexHeatmap.*

171

172 **Results**

173 **Inter-annual and spatial variability in environmental variables and bloom dynamics**

174 The environmental conditions varied largely within and between the years due to the location

175 of the different subbasins and time of the sampling (Table 1, Fig. 1A). For instance, salinity

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176   ranged from ~ 8 in the southernmost station of the BP to ~2 in the northernmost part of the

177   BoB (Table 1). In general, the $NO_2^-+NO_3^-$ was depleted after the bloom with the exception of

178   the P-limited BoB, where the $NO_2^-+NO_3^-$ concentration was similar to the stations in the

179   Growth phase and phosphate was low. The lowest dissolved DSi concentration was measured

180   in the GoF in 2014 while the maximum DSi concentration was detected in the Kemi river

181   plume in the northernmost BoB. The DOC and DON were highest in the ArS in 2014, and

182   higher in the GoF and the Gulf of Bothnia compared with the BP.

183   Overall, diatom carbon biomass was significantly higher in the Growth and Peak

184   phytoplankton bloom phases compared to the Decline and Postbloom phases (Wilcoxon test,

185   $p < 0.05$), whereas dinoflagellate carbon biomass was also significantly high in the Decline

186   phase (Wilcoxon test, $p < 0.05$). However, no clear increasing trend in the biomass of

187   dinoflagellates compared to diatoms was observed across the studied years. In 2013, we

188   sampled right after the ice melt in the Gulf of Finland and the water temperature was low in

189   the entire sampling area (Table 1). The spring bloom was in the Growth to Decline phase in

190   the 2013 cruise (Table 1), and was largely dominated by diatoms, displaying much higher

191   carbon contribution to total biomass than in the other three cruises (Fig. 1B). In 2014, the

192   temperature was 4-5.5 °C and the spring bloom was in a more advanced phase with low

193   phytoplankton biomass (Fig. 1B). In 2015, the conditions were similar to 2014, with the

194   exception of the Gulf of Bothnia where the spring bloom was in the Peak to Decline phase at

195   the time of the sampling. In 2016, we sampled the same areas and time of the year as 2013,

196   but the temperature was warmer in the BP subbassin and the spring bloom was more

197   advanced in the latest year than in the first sampling cruise (Table 1). In these warmer

198   cruises, dinoflagellates contributed > 300 µg C L^{-1} to the carbon biomass at some stations

199   (Fig. 1B).

200   High PP was observed during the Growth and Peak phases (Fig. 1C), following the

201   tendency of the Chl *a* and coinciding with the stations that presented high carbon biomass

202   (Fig. 2A, Table S2). The PER, measured after 24 h, was high in the Decline and Postbloom

203   phase in the BP and the BoB, in contrast to PP (Fig. 1C, Table S2). The trophic status,

204   estimated as the POC:Chl *a* ratio, was in general low, except at some stations in the BP and

205   the BoB (Table 1).

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207   **Changes in plankton community composition**

208   We observed pronounced differences in taxonomic composition of the studied plankton

209   communities (mainly phytoplankton and nano-microzooplankton here) across the different

210   subbasins and years, which appeared mainly shaped by the inorganic nutrient concentrations

211   and the water temperature (Fig. 2A, B). Diatoms largely dominated in the GoF and ÅS (Table

212   1), with high contributions of *Achnanthes taeniata*, *Skeletonema marinoi*, *Thalassiosira*

213   *levanderi*, and *Chaetoceros* spp., whereas in the Bothnian Sea, *T. baltica* dominated during

214   the Peak bloom phase (Fig. 2A). Dinoflagellates were dominated by the dinoflagellate

215   complex, mainly formed by *Biecheleria baltica* and/or *Gymnodinium corollarium* in the GoF

216   and BP. The chain forming *Peridiniella catenata* was present in all the subbasins except at

217   the stations with clearly low carbon biomass (Fig. 2A). Other autotrophic dinoflagellates

218   (*Heterocapsa* spp.) and heterotrophic/mixotrophic dinoflagellates (e.g. *Protoperidinium* spp.

219   and *Dinophysis* spp., respectively) were also present, but their contribution to the total

220   plankton carbon biomass was < 5 %. The contribution of other autotrophic plankton

221   organisms, such as Crytophyceae, Chrysophyceae, Prasinophyceae, Prymnesiophyceae,

222   Chlorophyceae, to the total carbon biomass was also generally low (< 2 µg C L^{-1}).

223   The mixotrophic ciliate, *Mesodinium rubrum,* was present across all sub-basins having

224   highest contribution in the BS and being in general more abundant in 2016 compared with

225   other years (Fig. 2A). Heterotrophic ciliates were most abundant during the late phase of the

226   bloom in the GoF and the northern BP. The silicoflagellate *Ebria tripartita* and the

227   heterotrophic nanoflagellates (HNF) had generally low abundance but contributed more than

228   10 µg C L^{-1} to the total carbon biomass during the Peak and Decline phases of the bloom also

229   at some stations in the GoF and the BP.

230

231   **Bacterial community composition and its environmental and biological drivers**

232   The taxonomic composition of the studied bacterial communities varied largely across the

233   stations sampled (Fig. 3). Overall, Alphaproteobacteria, mainly SAR11, dominated in all the

234   sampled years (Fig. 3A). Bacteroidetes were dominated by the class Flavobacteriia, which

235   comprised on average ~ 25 % of the total relative abundance. Within Flavobacteriia, the

236   genus *Flavobacterium* contributed 8–12 % to the total relative abundance during the

237   Peak/Decline phase in 2013 and 2016, together with *Fluviicola*, *Polaribacter* and NS3 marine

238   group, whereas *Owenweeksia* was found in the Growth phase. Gammaproteobacteria

239   (*Crenothrix*) had higher relative abundance (> 10 %) in the GoF and the ÅS in 2013 and

240   2016. The classes Actinobacteria (hgcl clade) and Acidimicrobiia (CL500-29 marine group)

241   had higher relative abundance in 2014 and 2015. The phylum Phycisphaerae (CL500-3) was

242   scarce but reached its highest values (~ 8 %) in the Gulf of Bothnia in 2015.

243   Based on their overall taxonomic structure, the communities were clustered into two main

244   groups (Wilcoxon; *p* < 0.001) differing largely in taxonomic richness (Fig. 3B) and diversity

245   (Shannon index; table S2) as well as in the bacteria abundances of the major bacterial groups.

246   The group with high taxonomic diversity comprised communities sampled during 2013 and

247   2014, and showed higher relative abundances of Bacteroidetes, Gammaproteobacteria and

248   Actinobacteria, whereas the low taxonomic richness group (2015–2016) showed a dominance

249   of Alphaproteobacteria (SAR11 and Rhodobacteraceae) and Cyanobacteria (Fig. 3B).

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250   Although the bacterial community structure was strongly influenced by the salinity gradient

251   (Fig. 3C), the shift in community structure observed between 2013–2014 and 2015–2016

252   seemed to be largely driven by certain phytoplankton groups such as the diatoms *A. taeniata,*

253   *S. marinoi, T. levanderi* and *Chaetoceros* spp*.,* as well as temperature and the POC:Chl *a*

254   ratio (Fig. 3C). The role of the phytoplankton community composition in shaping bacterial

255   assemblages was further supported by the strong correlations between bacterial and

256   phytoplankton taxonomic dissimilarity matrices found across the four years of study

257   (Spearman r values 0.6–0.8, Mantel test *p* = 0.0001; Table S3).

258   When exploring the individual correlations between different environmental and

259   biological factors and the abundances of the main bacterial groups (Fig. 4), we observed two

260   groups of bacterial taxa showing fundamentally contrasting relationships. Bacteroidetes

261   (Flavobacteriia, Cytophagia and Sphingobacteriia), Gamma- and Betaproteobacteria were

262   strongly correlated with diatoms and *E. tripartita,* and negatively correlated with temperature

263   and POC:Chl *a* ratios (Fig. 4). In contrast, Rhodobacteraceae, SAR11 and Cyanobacteria

264   correlated positively with temperature, POC:Chl *a* ratios, HNFs and ciliates, whereas the

265   dominance of certain diatoms or dinoflagellates caused decreases in their abundances (Fig.

266   4). Phycisphaerae correlated strongly with the diatom *T. baltica*. Within dinoflagellates, the

267   dinoflagellate complex was the only group that correlated positively with all the bacterial

268   taxa dominant in 2013–2014, but also with Rhodobacteraceae (Fig. 4), which was more

269   prevalent in 2015–2016 (Fig. 3B).

270   The observed changes in composition of the phytoplankton communities were

271   accompanied by a large spatial-temporal variation of bacterial abundances and production

272   (Fig. 5). Overall, the bacterial production rates measured with leucine (BPL) were highest in

273   2013 (Fig. 5A, Table S2), having similar pattern of the PP (Fig. 2A), while when measured

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274   with thymidine (BPT), the rates were highest in 2014 (Fig. 5A). In contrast, the highest BA

275   values were recorded at the stations sampled during the 2015 cruise (Fig. 5B; Table S2).

276   The differences in BCC had an impact on community functioning. Most of the bacterial

277   groups that were prevalent in the high diversity assemblages from 2013 and 2014, i.e. Beta-,

278   Gammaproteobacteria, Actinobacteria, Bacteroidetes, and more specifically Flavobacteriia

279   showed strong positive correlations with BPL and BPT (Fig. 5C−D; Table S4). In contrast,

280   the abundance of Alphaproteobacteria (SAR11 in particular), was negatively correlated with

281   bacterial production rates (Fig. 5E, Table S4)**.**

282

283   **Discussion**

284   Our results demonstrate that the bacterioplankton community structure was strongly affected

285   by the different phytoplankton communities, either dominated by diatoms or dinoflagellates,

286   and by the different phases of the phytoplankton bloom. Although our results do not evidence

287   a clear trend of increasing dominance of dinoflagellates from 2013 to 2016, we did capture

288   much higher carbon biomass of diatoms during the 2013 cruise than in the other years. This

289   heterogeneity allowed us to explore how changes in the dominant phytoplankton groups are

290   translated into changes in the structure and functioning of the associated bacterioplankton

291   communities.

292

293   **Dynamics of environmental variables and phytoplankton biomass**

294   We found that the different environmental conditions, primarily temperature and inorganic

295   nutrient concentrations such as NO_3^- and PO_4^{3-}, shaped the phytoplankton community

296   structure and bloom dynamics in terms of Chl *a* concentration, PP and dominant species, in

297   agreement with previous studies in the Baltic Sea [9,31]. The phytoplankton community during

298   the early bloom development, with low temperature and high inorganic nutrient

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299   concentrations, was clearly dominated by diatoms, in particular in the GoF. Diatom

300   dominance was also associated with high primary productivity suggesting an actively

301   growing community. The prevailing diatoms were the taxa that typically occur during the

302   cold water season [32-34]. In the BS, the Peak phase was dominated by the single diatom species

303   *Thalassiosira baltica* along with high carbon biomass of the mixotrophic ciliate *Mesodinium*

304   *rubrum,* which both seem to be common in this sub-basin [7]. The diatom-dominated

305   communities co-occurred to a varying extent with several dinoflagellates, most likely

306   *Gymnodinium corollarium* and *Biecheleria baltica* [35,36] in the GoF and the BP, similarly to

307   the earlier observations [2,3]. The heterotrophic *Ebria tripartita* was more abundant during

308   diatom dominance, likely because it grazed on diatom species such as *Skeletonema* sp. and

309   *Thalassiosira* spp. [37].

310   During the declining phytoplankton bloom, the water was warmer, the Chl *a*

311   concentration and the total carbon biomass were lower and dinoflagellates contributed more

312   to the biomass than in the Growth and Peak phases. This is the typical situation in the GoF

313   with diatoms being more abundant in early spring and dinoflagellates gradually becoming

314   more dominant as the nutrients are depleted [10,38]. For instance, the stations sampled during

315   2014 presented low phytoplankton biomass and also low DSi concentration in the GoF,

316   suggesting that a diatom-dominated bloom had settled out of the surface layer before the time

317   of sampling. This probably explained the similarity in structure between the bacterial

318   communities and productivity found in 2014 and those in the diatom-dominated stations of

319   2013. In the BoB, the low phytoplankton carbon biomass was limited by phosphorus with

320   excess NO_3^- and DSi and high DOC and DON concentrations. This sub-basin is P-limited [9]

321   and is more turbid due to terrestrial DOM inputs, which limits light penetration resulting in

322   light-limited primary production [39-41].

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323   The abundance of nano- and microzooplankton (HNF, ciliates and *M. rubrum*) were also

324   higher during the Decline phase of the bloom, which, together with higher POC:Chl *a* ratios (

325   > 175), points to a more heterotrophic plankton community [24]. A decrease in phytoplankton

326   biomass and an increase in the proportion of dinoflagellates and heterotrophic organisms

327   have been projected for a future, warmer Baltic Sea [1], in addition to the earlier onset of the

328   spring bloom [42]. Our results represent different temperature scenarios, for example the 2013

329   and 2016 cruises sampled the same subbasins during varying environmental conditions with

330   2016 being warmer and the bloom in a more advance stage, in line with these predictions.

331

332   **Bacterial community dynamics and their environmental drivers**

333   The changes in the phytoplankton community composition between the bloom phases

334   resulted in pronounced shifts in the structure of the associated bacterial communities and

335   likely related to the availability of the released DOM, largely produced by phytoplankton

336   [43,44]. The availability of this DOM can lead to changes in bacterial community dynamics [45-47],

337   which have been also reported in several field studies during phytoplankton spring blooms

338   [16,17,48].

339   The studied bacterial communities clustered into two clearly different groups of high and

340   low taxonomic richness and diversity, which corresponded to the communities sampled

341   during 2013−2014, and those sampled during 2015−2016, respectively. Salinity and

342   temperature appeared as relevant factors in shaping the bacterial communities in the Baltic

343   Sea, similar to previous studies [20,22,49]. However, in our study, the salinity was not responsible

344   for the segregation observed in the bacterial communities. Instead, changes in the carbon

345   biomass of specific phytoplankton groups such as *Achnanthes taeniata*, *Chaetoceros* spp.,

346   *Skeletonema marinoi*, and *T. levanderi* emerged as some of the most important drivers in the

347   configuration of bacterial communities. Thus, the diatom bloom observed in 2013 and the

348   diatom bloom that had likely recently settled out of the surface layer in 2014 promoted a

349   higher diversity of associated bacterial communities with more Gammaproteobacteria,

350   Betaproteobacteria, and Bacteroidetes. This conclusion was further strengthened by the

351   strong and positive correlations between the diatom species and these copiotrophic bacteria,

352   in accordance with their preference for productive or bloom-like conditions. All these groups

353   are known to be favored during diatom blooms [23,50]. In contrast, the lower diatom carbon

354   biomass observed during 2015 and 2016 were associated with less diverse communities

355   dominated by the SAR11 group in 2015 and also by Rhodobacteraceae (genus *Gemmobacter*)

356   in 2016, supported by the strong negative correlations between this bacterial community

357   structure and the phytoplankton carbon biomass. The conditions within the less productive

358   stations sampled during 2015 and 2016 likely promoted the dominance of SAR11, which

359   typically occurs in nutrient-poor environments [51,52]. Other groups like Phycisphaerae, and in

360   particular CL500-3, appeared to follow more clearly the salinity gradient and was strongly

361   associated to the presence of *T. baltica*. The presence of Phycisphaerae has been associated to

362   the lower salinity of the Gulf of Bothnia and to a higher influence of allochthonous DOM [19].

363   Interestingly, temperature emerged as a one of the main drivers for the structure of both, the

364   phytoplankton and bacterioplankton communities, suggesting that the predicted increases in

365   water temperature along the Baltic Sea [53,54] may lead to dramatic changes in the planktonic

366   assemblages inhabiting this vulnerable system.

367   Within the high-diversity bacterial communities found in 2013 and 2014, we observed

368   that the dynamics of Gammaproteobacteria and Bacteroidetes differed between bloom phases

369   and subbasins. Bacteroidetes were always present with fluctuating relative abundance,

370   whereas Gammaproteobacteria, dominated by the genus *Crenothrix,* peaked shortly and were

371   restricted to those stations with complex diatom communities. These bacterial groups are

372   known to occupy different niches in terms of metabolic affinities: Flavobacteriia are capable

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373   of degrading high molecular weight compounds, such as proteins, chitin and polysaccharides

374   [48,55], whereas Gammaproteobacteria are more specialized on breaking down low molecular

375   weight substrates, peaking shortly during increased organic and inorganic substrate

376   availability [56,57]. Thus, their different patterns were likely related to the DOM availability and

377   niche partitioning during the highly dynamic phytoplankton blooms. This was supported by

378   the lower net PER during diatom dominance compared with dinoflagellate dominance,

379   suggesting that diatoms release more labile DOC, and therefore more rapidly consumed, than

380   dinoflagellates (Lignell 1990).

381   The succession of taxa within Bacteroidetes in the different phases of the bloom was

382   similar to that observed in a mesocosm experiment simulating diatom and dinoflagellate

383   blooms with Baltic Sea bacterial communities [23]. In the Growth phase, *Owenweeksia*,

384   *Flabovacterium* and *Fluviicola* were more dominant, whereas the genera *Polaribacter* and

385   NS3 marine group were favored in the declining phase. Within Gammaproteobacteria, the

386   presence of *Crenothrix* in surface waters was surprising since this taxon is a methane-

387   oxidizer specialized in the uptake of single-carbon substrate, such as methane and methanol,

388   but also acetate and glucose [58]. This filamentous bacterium may have benefited by the

389   increase of the suitable substrates, either by fresh and labile DOM released or as a product

390   from a metabolic cascade after the degradation process carried out by other bacterial groups.

391   A lower diversity of Bacteroidetes was observed in the presence of the diatom *T. baltica*

392   in the BS. A previous study has reported the production of certain polyunsaturated aldehydes

393   (PUAs) by *Thalassiosira spp.* [59], which can affect the bacterial protein production of

394   Bacteroidetes negatively [60]. This could have played a role in the reduced growth of this

395   bacterial group. Any anti-bacterial effects of phytoplankton are not well studied, but there are

396   indications that some species inhibit bacterial growth [50].

397

Links between bacterial community composition and ecosystem functioning

399   The observed changes in bacterial community structure resulted in pronounced changes in

400   the bacterial production of the studied communities. Most stations sampled during 2013 and

401   2014 showed much higher rates of BPL compared to the stations sampled in 2015 and 2016

402   and were similar or, in some cases, larger than those reported by previous studies conducted

403   at similar temperatures during the spring in the Baltic Sea [16,61]. The larger values of BPL

404   coincided with the higher abundances of copiotrophic groups such as Betaproteobacteria,

405   Gammaproteobacteria and Bacteroidetes associated to diatom dominance. The variation in

406   BPL between studies conducted at similar temperatures may be due to the different

407   dominating phytoplankton groups that produce DOM of different lability. This emphasizes

408   the importance of the phytoplankton taxonomical composition as the driver of the bacterial

409   carbon cycling mediated by the presence of specific bacterial taxa. Large peaks in BPT were

410   also observed in 2014 indicating an actively dividing community and mostly related to the

411   increases in relative abundance of Actinobacteria, which is known to occur after

412   phytoplankton blooms [20] and correlate positively with thymidine incorporation [62]. In contrast,

413   SAR11 correlated negatively with BPL, which agrees with the small and slow-growing

414   pattern of this clade typically occurring in oligotrophic ecosystems [51,52]. Similar bacterial

415   responses were observed in a mesocosm study, where the growth of diatoms resulted in

416   higher bacterial production and higher dominance of Flavobacteriia, Gammaproteobacteria

417   and Cytophagia than in the presence of dinoflagellates [23]. The robustness of the observed

418   patterns in our study, linking BCC and functioning, are essential in microbial ecology since

419   the studies that explore these relationship are scarce in the Baltic Sea [63].

420   Gammaproteobacteria and Bacteroidetes are known to be quickly grazed by HNF [51],

421   which prefer to graze on actively growing bacteria. This may explain the low bacterial

422   abundance of the active community observed in 2013. The dinoflagellate complex also

423   correlated positively with the relative abundances of these bacterial groups. However,

424   previous experimental observations reported a decrease in bacterial productivity upon the

425   addition of *Biecheleria baltica* [23], one of the species forming the dinoflagellate complex.

426

Conclusions

428   Our results during and after phytoplankton spring blooms highlight the differences in BCC,

429   diversity and productivity under distinct phytoplankton communities dominated by diatoms

430   and/or dinoflagellates. Diatoms like *Skeletonema marinoi, Achnanthes taeniata* and

431   *Chaetoceros* spp. likely release high-quality DOM that boosts the bacterial production and

432   enhances the development of highly diverse bacterial communities dominated by Gamma-,

433   Betaproteobacteria and Bacteroidetes. In contrast, less diverse and less productive

434   bacterioplankton communities dominated by SAR11 and Cyanobacteria are observed in

435   presence of more dinoflagellates. Based on our results, it is not the overall dominance of

436   dinoflagellates or diatoms what drives the largest changes in bacterial community structure,

437   but rather the presence of particular taxonomic groups. Finally, although temperature appears

438   as a main factor shaping both bacterial and phytoplankton communities, higher temperatures

439   are not necessarily related to higher bacterial production rates. Thus, it is needed to

440   incorporate knowledge on the changes in plankton community composition to be able to

441   understand and predict the biogeochemical consequences of expected temperature increases

442   and associated changes in phytoplankton blooms in Baltic Sea waters.

443

Conflict of interest

445   The authors declare no conflict of interest

446

447

454   **References**

455   1   Legrand C., Fridofsson E., Bertos-Fortis M., Lindehoff E., Larsson P., Pinhassi J., *et*

456   *al.* Interannual variability of phyto-bacterioplankton biomass and production in

457   coastal and offshore waters of the Baltic Sea. *Ambio* 2015; **44:** 427–438.

458   2   Klais R., Tamminen T., Kremp A., Spilling K., Olli K. Decadal-scale changes of

459   dinoflagellates and diatoms in the anomalous Baltic Sea spring bloom. *PLoS One*

460   2011; **6**: e21567

461   3   Wasmund N., Tuimala J., Suikkanen S., Vandepitte L., Kraberg A. Long-term trends

462   in phytoplankton composition in the western and central Baltic Sea *J Mar Syst* 2011;

463   **87**: 145–159.

464   4   Wasmund N., Uhlig S. Phytoplankton trends in the Baltic Sea. *ICES J Mar Sci* 2003;

465   **60**: 177–186.

466   5   Klais R., Tamminen T., Kremp A., Spilling K., An B.W., Hajdu S., *et al.* Spring

467   phytoplankton communities shaped by interannual weather variability and dispersal

468   limitation: Mechanisms of climate change effects on key coastal primary producers.

469   *Limno Oceanogr* 2013; **58**: 753–762.

470   6   Kahru M., Nômmann S. The phytoplankton spring bloom in the Baltic Sea in 1985,

471   1986: multitude of spatio-temporal scales. *Cont Shelf Res* 1990; **10**. 329–354.

472   7   Andersson A., Hajdu S., Haecky P., Kuparinen J., Wikner J. Succession and growth

473   limitation of phytoplankton in the Gulf of Bothnia (Baltic Sea). *Mar Biol* 1996; **126**:

474   791–801.

475   8   Hagström Å., Azam F., Kuparinen J., Zweifel U. L. in *A systems analysis of the Baltic*

476   *Sea*. Springer 2001: 177–210.

477   9   Tamminen T., Andersen T. Seasonal phytoplankton nutrient limitation patterns as

478   revealed by bioassays over Baltic Sea gradients of salinity and eutrophication. *Mar*

479   *Ecol Prog Ser* 2007; **340**: 121–138.

480   10   Heiskanen A. in *Monographs of the Boreal Environment Research 8*. Finnish

481   Environment Institute, 1998.

482   11   Spilling K., Markager S. Ecophysiological growth characteristics and modeling of the

483   onset of the spring bloom in the Baltic Sea. *J Mar Syst* 2008; **73**: 323–337.

484   12   Castillo C. R., Sarmento H., Alvarez-Salgado X. A., Gasol J. M., Marraséa C.

485   Production of chromophoric dissolved organic matter by marine phytoplankton.

486   *Limno Oceanogr* 2010; **55**: 446–454.

487   13   Myklestad S. M. Release of extracellular products by phytoplankton with special

488   emphasis on polysaccharides. *Sci Total Environ* 1995; **165**: 155–164.

489   14   Spilling K., Kremp A., Klais R., Olli K., Tamminen T. Spring bloom community

490   change modifies carbon pathways and C: N: P: Chl a stoichiometry of coastal material

491   fluxes. *Biogeosciences* 2014; **11**: 7275-7289.

492   15   Spilling K., Lindström M. Phytoplankton life cycle transformations lead to species-

493   specific effects on sediment processes in the Baltic Sea. *Con Shelf Res* 2008; **28**:

494   2488–2495.

495   16   Lindh M. V., Sjöstedt J., Andersson A. F., Baltar F., Hugerth L. W., Lundin D., *et al.*

496   Disentangling seasonal bacterioplankton population dynamics by high-frequency

497   sampling. *Environ Microbiol* 2015; **17**: 2459–2476.

498   17   Bunse, C. Bertos-Fortis M., Sassenhagen I., Sildever S., Sjöqvist C., Godhe A., *et al.*

499   Spatio-temporal interdependence of bacteria and phytoplankton during a Baltic Sea

500   spring bloom. *Front Microbiol* 2016; **7**: 517.

501   18   Herlemann D. P., Labrenz M., Jürgens K., Bertilsson S., Waniek J. J., Andersson A.

502   F. *et al.* Transitions in bacterial communities along the 2000 km salinity gradient of

503   the Baltic Sea. *ISME J* 2011; **5**: 1571–1579.

504   19   Rieck A., Herlemann D. P., Jürgens K., Grossart H. P. Particle-associated differ from

505   free-living bacteria in surface waters of the Baltic Sea. *Front Microbiol* 2015; **6**:

506   1297.

507   20   Hugerth L. W., Larsson J., Alneberg J., Lindh M. V., Legrand C., Pinhassi J., *et al.*

508   Metagenome-assembled genomes uncover a global brackish microbiome. *Genome*

509   *Biol* 2015; **16**: 279.

510   21   Laas P., Simm J., Lips I., Lips U., Kisand V., Metsis M. Redox-specialized

511   bacterioplankton metacommunity in a temperate estuary. *PloS One* 2015; **10**:

512   e0122304.

513   22   Andersson A. F., Riemann L., Bertilsson S. Pyrosequencing reveals contrasting

514   seasonal dynamics of taxa within Baltic Sea bacterioplankton communities. *ISME J*

515   2010; **4**: 171–181.

516   23   Camarena-Gómez M. T., Lipsewers T., Piiparinen J., Eronen-Rasimus E., Perez-

517   Quemaliños D., Hoikkala L., *et al.* Shifts in phytoplankton community structure

518   modify bacterial production, abundance and community composition. *Aquat Microb*

519   *Ecol* 2018; **81**: 149–170.

21

520   24   Vargas C. A., Martínez R. A., Cuevas L. A., Pavez M. A., Cartes C., González H. E.,

521   *et al.* The relative importance of microbial and classical food webs in a highly

522   productive coastal upwelling area. *Limnol Oceanogr* 2007; **52**,:1495–1510.

523   25   Lipsewers T., Spilling K. Microzooplankton, the missing link in Finnish plankton

524   monitoring programs. *Boreal Environ Res* 2018; **23**, 127–137.

525   26   Wasmund N., Kownacka J., Göbel J., Jaanus A., Johansen M., Jurgensone I., *et al.* The

526   diatom/dinoflagellate index as an indicator of ecosystem changes in the Baltic Sea. 1.

527   Principle and handling instruction. *Front Mar Sci* 2017; **4**: 22.

528   27   Gargas E. A manual for phytoplankton primary production studies in the Baltic. *Baltic*

529   *Mar Biologists* 1975; **2**: 1–88. (Copies available from: Water Quality Institute, 11

530   Agern Allé, DK-2970 Hørsholm, Denmark).

531   28   Morán G. X. A., Estrada M., Gasol J. M., Pedrós-Alió C. Phytoplanktonic DOC and

532   POC production in the Bransfield and Gerlache Straits as derived from kinetic

533   experiments of 14C incorporation. *Deep Sea Research Part II: Topical Studies in*

534   *Oceanography* 2002; **49**: 769–786.

535   29   Gasol J. M., Del Giorgio P. A. Using flow cytometry for counting natural planktonic

536   bacteria and understanding the structure of planktonic bacterial communities. *Sci Mar*

537   2000; **64**: 197–224.

538   30   Logares R. Workflow for analysing MiSeq amplicons based on Uparse v1. 5. 2017

539   *Zenodo10* **5281**.

540   31   Fleming V., Kaitala S. Phytoplankton spring bloom intensity index for the Baltic Sea

541   estimated for the years 1992 to 2004. *Hydrobiologia* 2006; **554**: 57–65.

542   32   Ikävalko J., Thomsen H. A. The Baltic Sea ice biota (March 1994): a study of the

543   protistan community. *Eur J Protistol* 1997; **33**: 229–243.

544 33 Lignell R. Excretion of organic carbon by phytoplankton: its relation to algal biomass,

545   primary productivity and bacterial secondary productivity in the Baltic Sea. *Mar Ecol*

546   *Prog Ser* 1990; **68**: 85–99 *Oldendorf.*

547 34 Spilling K. On the ecology of cold-water phytoplankton in the Baltic Sea. 2007.

548 35 Olli K., Trunov, K. Abundance and distribution of vernal bloom dinoflagellate cysts

549   in the Gulf of Finland and Gulf of Riga (the Baltic Sea). *Deep Sea Research Part II:*

550   *Topical Studies in Oceanography* 2010; **57**: 235–242.

551 36 Sundström A. M., Kremp A., Daugbjerg N., Moestrup Ø., Ellegaard M., Hansen R., *et*

552   *al. Gymnodinium corollarium* sp. nov.(dinophyceae)—a new cold-water

553   dinoflagellate responsible for cyst sedimentation events in the baltic sea 1. *J Phycol*

554   2009; **45**: 938–952.

555 37 Hargraves P. E. The ebridian flagellates Ebria and Hermesinum. *Plankton Biol Ecol*

556   2002; **49**: 9–16.

557 38 Spilling K., Olli K., Lehtoranta J., Kremp A., Tedesco L., Tamelander T., *et al.*

558   Shifting diatom-dinoflagellate dominance during spring bloom in the Baltic Sea and

559   its potential effects on biogeochemical cycling. *Front Mar Sci* 2018; **19**.

560 39 Andersson A., Meier H. E. M., Ripszam M., Rowe O., Wikner J., Haglund P., *et al.*

561   Projected future climate change and Baltic Sea ecosystem management. *Ambio* 2015;

562   **44**: 34–356.

563 40 Rowe O. F., Dinasquet J., Paczkowska J., Figueroa D., Riemann L., Andersson A.

564   Major differences in dissolved organic matter characteristics and bacterial processing

565   over an extensive brackish water gradient, the Baltic Sea. *Mar Chem* 2018; **202**: 27–

566   36.

23

567 41   Sandberg J., Andersson A., Johansson S., Wikner, J. Pelagic food web structure and

568   carbon budget in the northern Baltic Sea: potential importance of terrigenous carbon.

569   *Mar Ecol Prog Ser* 2004; **268**: 13–29.

570 42   Groetsch P. M., Simis S. G., Eleveld M. A., Peters S. W. Spring blooms in the Baltic

571   Sea have weakened but lengthened from 2000 to 2014. *Biogeosciences* 2016; **13:**

572   4959–4973.

573 43   Ducklow H. W. Production and fate of bacteria in the oceans. *Bioscience* 1983; **33**:

574   494–501.

575 44   Kirchman D. Measuring bacterial biomass production and growth rates from leucine

576   incorporation in natural aquatic environments. *Methods in microbiology* 2001; **30**:

577   227–237.

578 45   Pinhassi J., Sala M. M., Havskum H., Peters F., Guadayol O., Malits A., *et al.*

579   Changes in bacterioplankton composition under different phytoplankton regimens.

580   *Appl Environ Microbiol* 2004; **70**: 6753–6766.

581 46   Riemann L., Steward G. F., Azam F. Dynamics of bacterial community composition

582   and activity during a mesocosm diatom bloom. *Appl Environ Microbiol* 2000; **66**:

583   578–587.

584 47   Sarmento H., Romera-Castillo C., Lindh M., Pinhassi J., Sala M. M., Gasol J. M., *et*

585   *al.* Phytoplankton species-specific release of dissolved free amino acids and their

586   selective consumption by bacteria. *Limnol Oceanogr* 2013; **58**: 1123–1135.

587 48   Teeling H., Fuchs B. M., Becher D., Klockow C., Gardebrecht A., Bennke C. M., *et*

588   *al.* Substrate-controlled succession of marine bacterioplankton populations induced by

589   a phytoplankton bloom. *Science* 2012; **336**: 608–611.
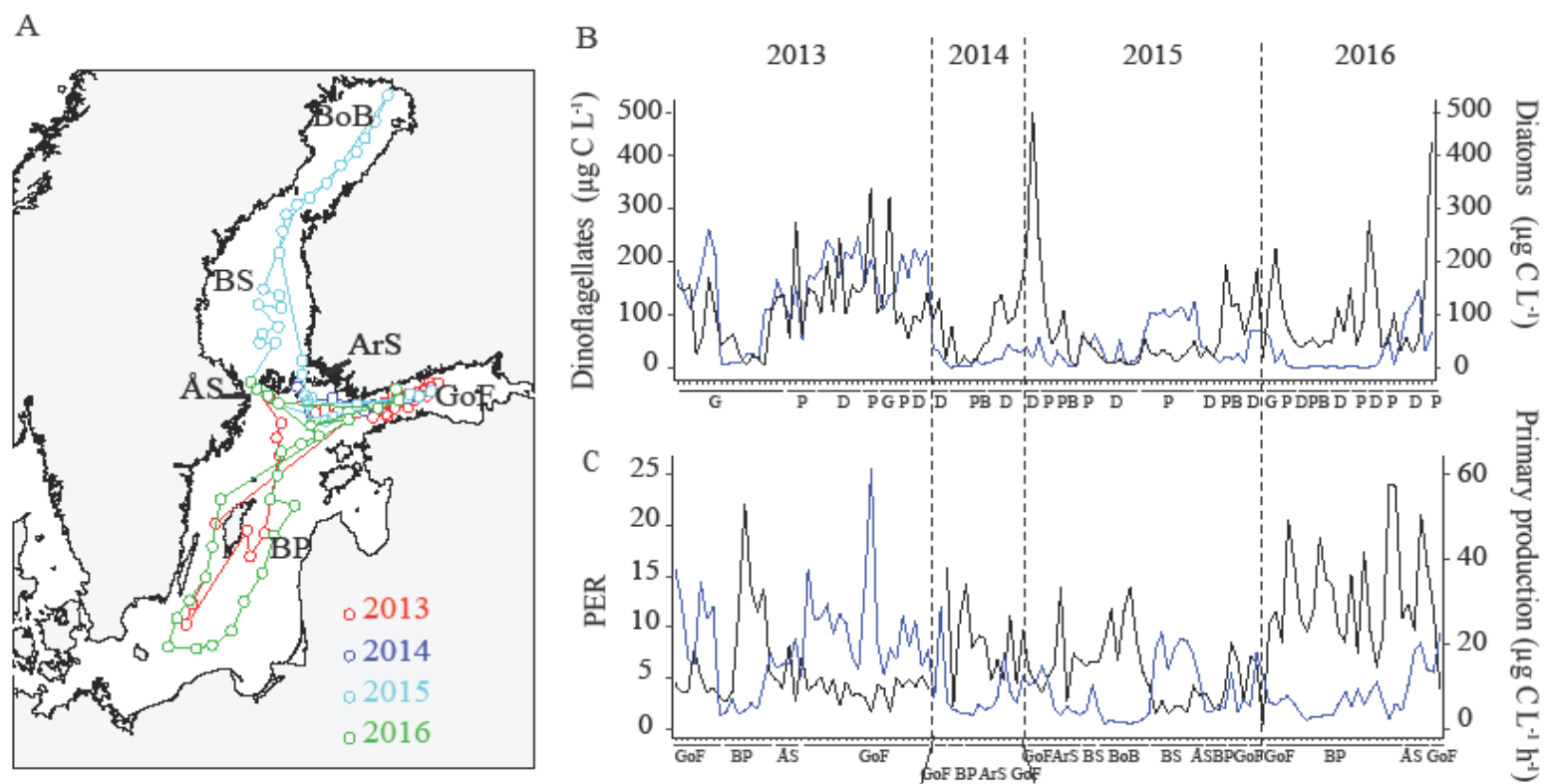
24

590   49   Herlemann D. P., Lundin D., Andersson A. F., Labrenz M., Jürgens, K. Phylogenetic

591   signals of salinity and season in bacterial community composition across the salinity

592   gradient of the Baltic Sea. *Front Microbiol* 2016; **7**.

593   50   Amin S. A., Parker M. S., Armbrust E. V. Interactions between diatoms and bacteria.

594   *Microbiol Mol Biol Rev* 2012; **76**: 667–684.

595   51   Alonso-Sáez L., Arístegui J., Pinhassi J., Gómez-Consarnau L., González J. M.,

596   Vaqué D., *et al.* Bacterial assemblage structure and carbon metabolism along a

597   productivity gradient in the NE Atlantic Ocean. *Aquat Microb Ecol* 2007; **46**: 43–53.

598   52   Giovannoni S. J., Tripp H J., Givan S., Podar M., Vergin K. L., Baptista D., *et al.*

599   Genome streamlining in a cosmopolitan oceanic bacterium. *Science* 2005; **309**: 1242–

600   1245.

601   53   Meier H. E. M., Hordoir R., Andersson H. C., Dieterich C., Eilola K., Gustafsson B.

602   G., *et al.* Modeling the combined impact of changing climate and changing nutrient

603   loads on the Baltic Sea environment in an ensemble of transient simulations for 1961–

604   2099. *Clim Dynam* 2012; **39**: 2421–2441.

605   54   Thomas D. N., Kaartokallio H., Tedesco L., Majaneva M., Piiparinen J., Eronen-

606   Rasimus E., *et al.* Life associated with Baltic Sea ice. *Biological oceanography of the

607   Baltic Sea*; 2017; 333–357.

608   55   Cottrell M. T., Kirchman D. L. Natural assemblages of marine proteobacteria and

609   members of the Cytophaga-Flavobacter cluster consuming low-and high-molecular-

610   weight dissolved organic matter. *Appl Environ Microbiol* 2000; **66**: 1692–1697.

611   56   Gómez-Consarnau L., Lindh M. V., Gasol J. M., Pinhassi J. Structuring of

612   bacterioplankton communities by specific dissolved organic carbon compounds.

613   *Environ Microbiol* 2012; **14**: 2361–2378.

614 57 Pinhassi J., Berman T. Differential growth response of colony-forming α-and γ-
615   proteobacteria in dilution culture and nutrient addition experiments from Lake
616   Kinneret (Israel), the eastern Mediterranean Sea, and the Gulf of Eilat. *Appl Environ*
617   *Microbiol* 2003; **69**: 199–211.

618 58 Stoecker K., Bendinger B., Schöning B., Nielsen P. H., Nielsen J. L., Baranyi C., *et*
619   *al.* Cohn's Crenothrix is a filamentous methane oxidizer with an unusual methane
620   monooxygenase. *Proceedings of the National Academy of Sciences* 2006; **103**: 2363–
621   2367.

622 59 Wichard T., Poulet S. A., Pohnert G. Determination and quantification of α, β, γ, δ-
623   unsaturated aldehydes as pentafluorobenzyl-oxime derivates in diatom cultures and
624   natural phytoplankton populations: application in marine field studies. *J Chromatogr*
625   *B* 2005; **814**: 155–161.

626 60 Balestra C., Alonso-Sáez L., Gasol J. M., Casotti, R. Group-specific effects on coastal
627   bacterioplankton of polyunsaturated aldehydes produced by diatoms. *Aquat Microb*
628   *Ecol* 2011; **63**: 123–131.

629 61 von Scheibner M., Herlemann D. P., Lewandowska A. M., Jürgens, K. Phyto-and
630   bacterioplankton during early spring conditions in the Baltic Sea and response to
631   short-term experimental warming. *Front Mar Sci* 2018; **5**: 231.

632 62 Pérez M. T., Hörtnagl P., Sommaruga, R. Contrasting ability to take up leucine and
633   thymidine among freshwater bacterial groups: implications for bacterial production
634   measurements. *Environ Microbiol* 2010; **12**: 74–82.

635 63 Lindh M. V., Pinhassi J. Sensitivity of Bacterioplankton to Environmental
636   Disturbance: a Review of Baltic Sea Field Studies and Experiments. *Front Mar Sci*
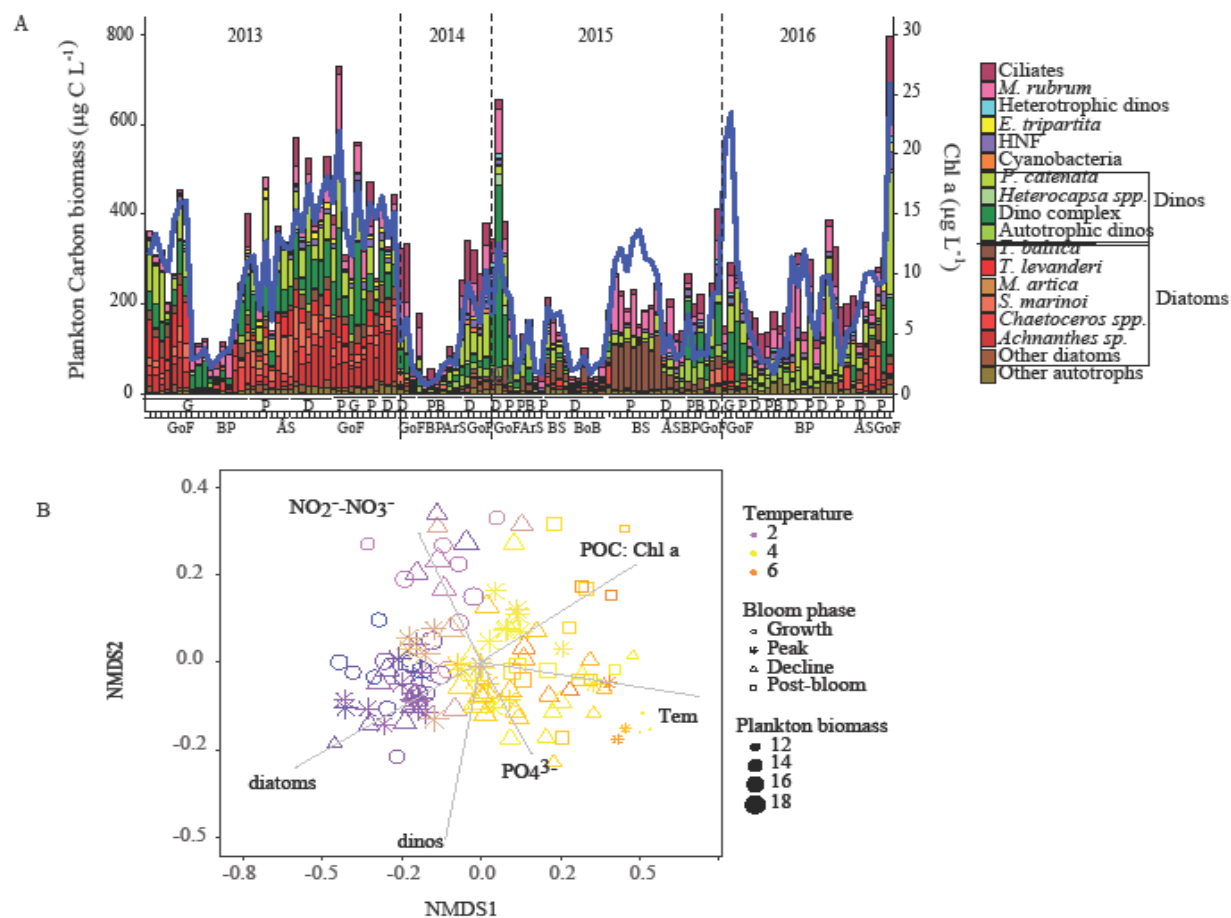637   2018; **5**: 361.

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Table 1. Environmental and biological variables measured along the four sampling transects (2013−2016). Values represent the range between the minimum and maximum values, in bold, within the four years. The different sub-basins covered are indicated: Gulf of Finland (GoF), Baltic Proper (BP), Åland Sea (ÅS), Archipelago Sea (ArS), Bothnian Sea (BS), Kvark (Kv) and Bay of Bothnia (BoB). The different phytoplankton bloom phases are indicated: Growth (G), Peak (P), Decline (D) and Post-bloom (PB), defined based on the Chl *a*, nitrate (NO_3^-) and phosphate (PO_4^{3-}) concentrations during the sampling at each subbasin (see Methods and Table S1). The inorganic nutrients and dissolved organic nitrogen are presented in µmol L^{-1} and the dissolved organic carbon is presented in mmol L^{-1}. The particulate organic carbon to Chl *a* (POC:Chl *a*) ratio indicates the trophic status (considering heterotrophic status those values larger than 200) and the diatoms:dinoflagellates index (Dia/Dino index) indicates diatom dominance or dinoflagellates dominance with values higher or lower than 0.5, respectively.

	#of Stations	Subbasins	Bloom phase	Temp (°C)	Saliniy (psu)	NO_2^--NO_3^-	NH_4^+	PO_4^{3-}	DSi	DOC	DON	POC:Chl *a*	Dia/Dino index
2013 April 10th-26th	29	GoF	G, P, D	0.76-1.98	5.09-6.38	0-5.85	0.10-0.39	0.19-0.48	7.84-19.75	0.30-1.08	10.06-45.57	33.36-52.47	0.30-0.86
	11	BP	G, P	1.62-2.72	5.97-7.24	0-2.08	0.04-0.19	0.06-0.33	8.27-14.24	0.41-1.01	13.49-30.78	38.96-100.35	0.09-0.94
	4	ÅS	G, P	1.29-1.59	5.45-6.16	0-2.59	0.07-0.13	0.02-0.35	8.92-13.10	0.35-0.51	16.07-17.85	44.54-56.39	0.36-0.74
2014 May 5th-10th	9	GoF	D, PB	4.25-5.18	5.03-6.01	0-0.26	0.5-0.24	0.12-0.33	2.27-9.10	0.50-1.05	16.07-28.70	49.43-80.00	0.11-0.29
	2	BP	D, PB	4.62-5.69	6.56-6.63	0	0.05	0.28	7.60-11.60	0.44-0.52	15.16-16.06	106.82-176.30	0.01-0.36
	4	ArS	D, PB	4.58-5.17	5.90-6.29	0-0.4	0.08-0.11	0.20-0.33	6.01-8.10	0.60-1.12	16.02-67.86	88.28-208.84	0.05.0.49
2015 May 4th-15th	5	GoF	D, PB	3.91-5.35	4.96-5.86	0-0.02	0.08-0.44	0.18-0.36	5.50-11.20	0.46-1.07	16.36-25.40	75.28-119.28	0.12-0.40
	6	ArS	P, PB	4.41-6.09	5.75-6.38	0-0.23	0.06-0.13	0.06-0.33	5.60-11.10	0.32-0.64	11.33-18.58	58.81-190.20	0.04-0.32
	4	ÅS	D	5.03-6.22	5.41-6.06	0	0-0.06	0.05-0.22	6.70-8.70	0.37-0.47	12.73-13.97	76.75-108.31	0.17-0.68
	4	BP	D, PB	4.52-5.69	5.72-6.56	0-0.44	0.05-0.34	0.27-0.40	7.6-11.8	0.37-0.55	15.16-18.95	83.49-149.96	0.09-0.33
	10	BS	P	3.59-4.16	5.12-5.54	0-0.38	0.05-0.13	0-0.21	8.10-12.70	0.33-1.07	12.40-18.66	31.93-82.12	0.48-0.90
	9	Kv, BoB	P, D	1.35-4.73	2.20-5.09	0-7.47	0.04-0.35	0-0.02	9.40-55.70	0.35-0.68	15.73-55.53	70.71-223.37	0.36-0.68
2016 April 4th-15th	4	GoF	G, P, D	1.56-2.89	4.95-5.20	0-6.64	0.14-0.22	0.34-0.61	12.21-19.09	0.44-0.58	14.71-31.52	25.85-57.10	0.13-0.77
	20	BP	G, P, D, PB	2.47-5.90	5.92-7.77	0-1.89	0-1.19	0.16-0.57	11.77-17.65	0.32-0.62	10.73-22.10	44.49-260.31	0.01-0.58
	4	ÅS	P	2.84-3.28	5.51-5.68	0-0.17	0.11-0.18	0.12-0.18	15.57-18.17	034-0.99	12.40-14.54	58.41-67.04	0.61-0.80

650 **Figure 1.** Sampling transects and phytoplankton dynamics across the studied period. A) Map with the sampling transects during the four cruises: 2013 (red), 2014 (dark
651 blue), 2015 (light blue) and 2016 (green). B) Total carbon biomass of dinoflagellates (black) and diatoms (blue) during the cruises and bloom phases. C) Percentage of
652 extracellular release (PER, black) and primary production (PP, blue) in the different sub-basins. See Table 1 for more details of the x-axis abbreviations.
653

654

655 **Figure 2.** Plankton dynamics in A) total carbon biomass of the main plankton groups and the chlorophyll *a* (Chl *a*) concentration at each station. The years are indicated on
656 top of the figure and the bloom phases and the sampled subbasins are at the bottom of the figure. See Table 1 for more details of x-axis abbreviations. B) Non-metric
657 multidimensional scaling (NMDS) plot based on Bray-Curtis distances between plankton community taxonomic composition (N = 126, stress = 0.1602). The vectors indicate
658 the environmental factors most strongly explaining the observed distribution based on RDA forward selection (NO_2^--NO_3^{-1}; nitrate-nitrite, Tem; temperature, PO_4^{3-};
659 phosphate, POC:Chl *a;* particulate organic carbon to Chl *a* ratio, diatoms and dinos; total carbon of diatoms and dinoflagellates). The colour gradient indicates the water
660 temperature, shape indicates the different bloom phases and size indicates the log-transformed total carbon biomass.

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Figure 3. Bacterial community dynamics and their environmental drivers. A) Relative abundance of the bacterial groups (class-family and genus level) that contribute more than 0.5 % of the total OTUs. Abbreviations as in figure 1. B) Non-metric multidimensional scaling (NMDS) plot based on Bray-Curtis distances of bacterial community structure (N = 122, stress = 0.134). Colour-code indicates the years (see Fig. 1) and size indicates the OTU richness in each community. Vectors indicate the main bacterial groups found in the four years. The abbreviations in the legend of Fig. A) and in Fig. B) indicate the main clades found during the sampling: Acidi; Acidimicrobiia, Actino; Actinobacteria, Flavo; Flavobacteriia; Sphingo; Sphingobacteriia; Cyano; Cyanobacterias, Alpha; Alphaproteobacteria; Beta; Betaproteobacteria; Gamma; Gammaproteobacteria; Phycis; Phycisphaerae (Phy). C) Non-metric multidimensional scaling (NMDS) plot linking bacterial community structure with environmental variables and plankton community. Colour-code indicates the salinity gradient and shape indicates the phytoplankton bloom phase. The vectors indicate the significant environmental variables as follow: *Achnanthes taeniata*, *Skeletonema marinoi*, *Thalassiosira levanderi*, *Chaetoceros* spp., Dinos (dinoflagellate complex), *Ebria tripartite*, *T. baltica*, HNF (heterotrophic nanoflagellates), temperature, POC Chl *a*, *Mesodinium rubrum*, ciliates, other autotrophs and salinity.
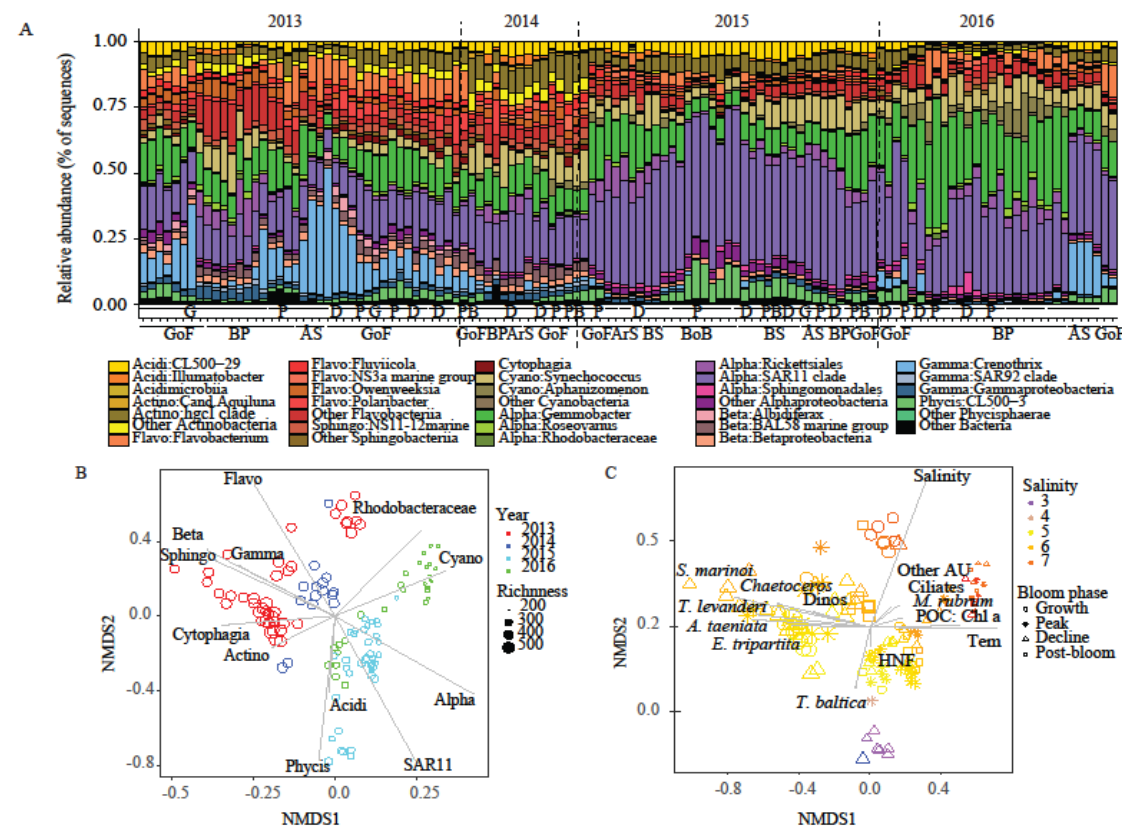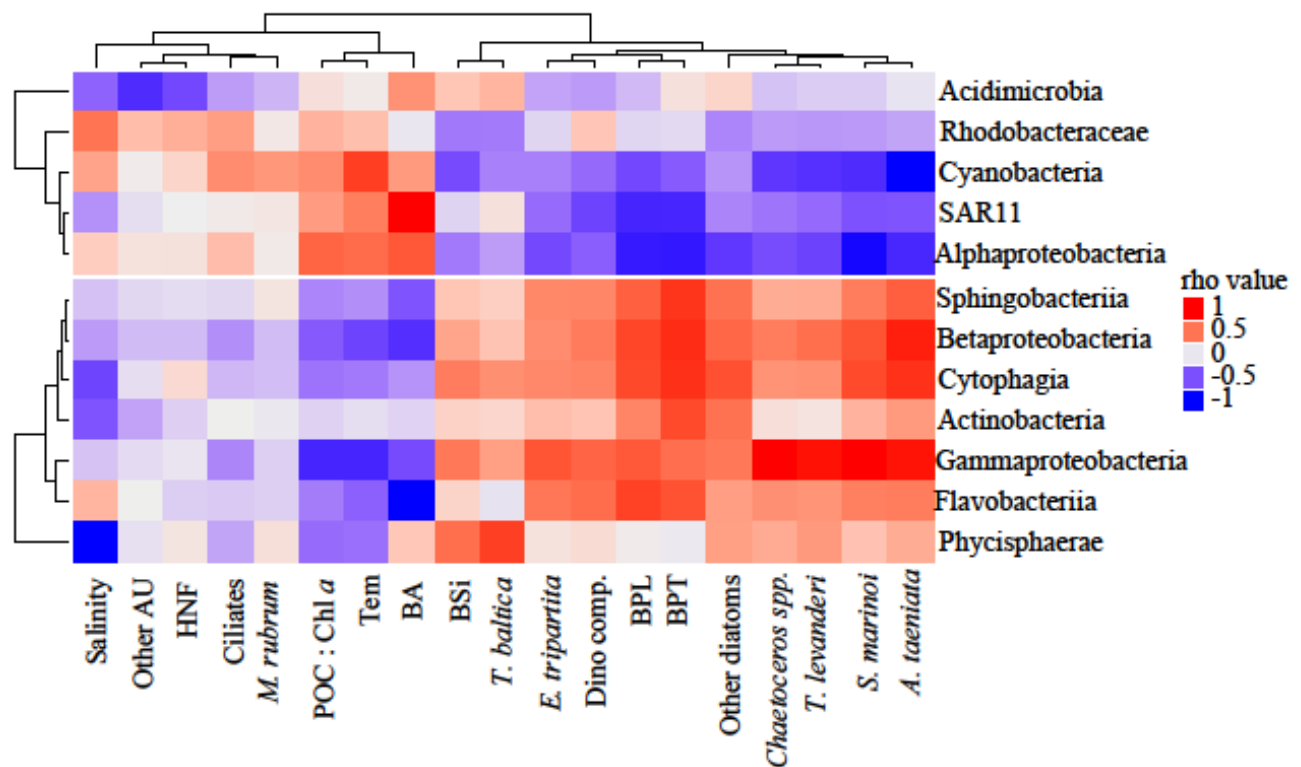
Figure 4. Heatmap with Spearman correlations between significant environmental variables, phytoplankton or heterotrophic plankton groups and the main bacterial groups. Colour-code indicates the correlation coefficients (rho values). The environmental factors (columns) are as in Fig. 3. In addition, BA = bacterial abundance; BSi = biogenic silicate; BPT = bacterial production thymidine and BPL = bacterial production leucine. N = 122.

683　**Figure 5.** Bacterioplankton productivity and abundance during the sampled spring blooms and the link to bacterial community. A) Bacterial heterotrophic production based
684　on leucine incorporation (BPL, black) and thymidine incorporation (BPT, blue), and B) bacterial abundance. The x-axis indicates the sub-basins across the sampling. See Fig.
685　1 for more detail of legend abbreviation. The dashed lines separate the different years. C-E) Spearman correlations between bacterial production measured as leucine uptake
686　(BPL) or thymidine uptake (BPT) and the relative abundance (%) of Flavobacteriia (C), Actinobacteria (D) and SAR11 (E), during the study. Rho values and p values are
687　presented on the figure. N = 122; p < 0.05.

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