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Singlet oxygen-induced alteration of bacteria associated with phytodetritus: effect of irradiance

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Abstract. Contrasting irradiation of senescent cells of the diatom *Thalassiosira* sp. in association with the bacterium *Pseudomonas stutzeri* showed the effect of intensity of irradiance on the transfer of singlet oxygen ($^1\text{O}_2$) to bacteria attached to phytoplanktonic cells. Under low irradiances, $^1\text{O}_2$ is produced slowly, favors the oxidation of algal unsaturated lipids (photodynamic effect) and limits $^1\text{O}_2$ transfer to attached bacteria. However, high irradiances induce a rapid and intense production of $^1\text{O}_2$, which diffuses out of the chloroplasts and easily reaches the attached bacteria, where it efficiently oxidizes their unsaturated membrane components. Analysis of numerous sinking particle samples collected in different regions of the Canadian Arctic showed that the photooxidation state of attached bacteria increased from ice-covered areas to open water, in agreement with *in vitro* results. Photooxidation of bacteria appeared to be particularly intense in sea ice, where the sympagic algae–bacteria association is maintained at relatively high irradiances for long periods of time.

Keywords. Senescent phytoplankton; Attached bacteria; Type II photosensitized oxidation; Irradiance effect; Arctic; Sinking particulate matter; Sea ice.

44 1. Introduction

45 Type II photooxidation processes are light-induced indirect degradation reactions, that
46 involve the action of a photosensitizer molecule (typically pigments or aromatic compounds)
47 in its triplet state (Gollnick, 1968). Chlorophyll, which is an efficient photosensitizer (Foote,
48 1976), can trigger type II photooxidation processes in phytoplanktonic cells, especially in
49 late bloom phases when they are senescent (Nelson, 1993). Indeed, in senescent cells, the
50 cessation of photosynthetic reactions results in an accelerated rate of formation of ^3Chl and
51 reactive oxygen species (ROS), mainly singlet oxygen ($^1\text{O}_2$) (Knox and Dodge, 1985;
52 Nelson, 1993). The rate of formation of these potentially damaging species can then exceed
53 the quenching capacity of the photoprotective system, thus enabling the photodegradation of
54 cell components (photodynamic effect) to occur (Merzlyak and Hendry, 1994). $^1\text{O}_2$ reacts
55 with a broad range of cellular compounds but especially unsaturated lipids such as sterols
56 and unsaturated fatty acids (Rontani and Belt, 2020), proteins (Morgan et al., 2004) and
57 nucleic acids (Agnéz-Lima et al., 1999; Ravanat et al., 2000). The very high reactivity of
58 $^1\text{O}_2$ with these biomolecules is due to the lack of spin restriction that normally prevents $^3\text{O}_2$
59 from reacting with these compounds (Zolla and Rinalducci, 2002).

60 The $^1\text{O}_2$ thus formed reacts not only with membrane components of phytoplankton but
61 also with the monounsaturated fatty acids (MUFAs) of its neighboring attached bacteria
62 (Rontani et al., 2003; Christodoulou et al., 2010, Petit et al., 2013, 2015). Indeed, in
63 phototrophic cells, the radius of the sphere of activity of $^1\text{O}_2$ from its point of production was
64 estimated to be between 155 and 340 nm (Baier et al., 2005; Ogilby, 2010; Skovsen et al.,
65 2005), which is a **sufficient** distance to allow it to cross the cellular membranes (Ogilby,
66 2010) and thus attack attached bacteria.

67 It has previously been demonstrated that low solar irradiance favors slow production
68 and diffusion of $^1\text{O}_2$ through the membranes and thus increases photooxidative damage of

69 the unsaturated algal lipids (Amiriaux et al., 2016). However, the effects of intensity of solar
70 irradiance on the photooxidation of bacteria attached to phytodetritus are totally unknown.

71 Here, to clarify this issue, we incubated senescent cells of the diatom *Thalassiosira* sp.
72 (a **phytoplankton** genus widely distributed in oceans, Malviya et al., 2016) in association
73 with the bacteria *Pseudomonas stutzeri* under contrasted artificial light irradiances. Bacteria
74 of the *Pseudomonas* genus were selected because they are often associated with
75 *Thalassiosira* cells (Bidle and Azam, 2001; Schaum, 2019). To validate the results, we
76 compared the photooxidation state of diatoms and their attached bacteria in numerous
77 samples of Arctic sea ice and sinking particulate matter. Samples **were collected** at different
78 depths under open water. marginal ice zone and first-year ice).

79

80 **2. Material and methods**

81

82 *2.1 Collection of in situ samples*

83 Detailed descriptions of the Arctic marine particulate matter and sea ice sample
84 collection processes (e.g. sampling dates, depths, volumes, etc.) can be found in Rontani et
85 al. (2012, 2016, 2022) and Amiriaux et al. (2020). Briefly, sinking particles were collected i)
86 in Resolute Passage under first-year ice at 5 m and 30 m depth with Hydro-Bios MS12-
87 model multi-sediment traps (Rontani et al., 2016), ii) in central Baffin Bay under the
88 marginal ice zone at 25 m depth with a Technicap PPS4 drifting sediment trap (Rontani et
89 al., 2022), and iii) in the Beaufort Sea in open water at 100 m depth with Technicap PPS
90 traps (Rontani et al., 2012). Sea-ice samples were collected at GreenEdge Project landfast
91 ice station located near Broughton Island in the Davis Strait using a Kovacs Mark V 14-cm-
92 diameter corer and focusing on the bottommost 10 cm of sea ice (Amiriaux et al., 2020) (Fig.
93 1).

94

95 *2.2. Algal phytodetritus production*

96 The non-axenic diatom *Thalassiosira* sp. RCC1714 was obtained from the Roscoff
97 culture collection. Diatoms were grown in F/2 medium at 17°C in a constant-environment
98 cabinet under an irradiance of 18 W.m⁻² (Osram, Lumilux 12:12h light/dark cycle). After
99 two months of growth, cell density was measured by counting under microscope, and death
100 of the algal cells was induced by sonication. *Thalassiosira* sp. cells were sonicated on ice
101 using a Branson Sonifier 450 ultrasound generator equipped with a Branson S450A cell
102 disruptor probe at 240 W for 10 minutes (duty cycle 50%). The sonicated cells were then
103 sub-sampled in 50 mL Falcon tubes and stored at -20°C for later photooxidation
104 experiments.

105

106 *2.3. Culturing of P. stutzeri*

107 Pre-culture of *P. stutzeri* ATCC 14405 was grown aerobically overnight on a rotary
108 shaker in the dark at 17°C on marine broth medium (US Biological™) diluted 1:10 with
109 sterilized artificial sea water (Kester et al., 1967) therefore not adding large amounts of
110 assimilable organic matter from the inoculum in the further steps. The culture was inoculated
111 with the aerobic pre-culture to obtain an initial OD_{620nm}, about 0.020. At the mid growth
112 exponential phase, the incubation was stopped and cells counted based on epifluorescence
113 in the presence of fluorochrome (4P,6-diamidino-2P-phenylindole dihydrochloride, DAPI)
114 as described in Otto (1990).

115

116 *2.4. Photooxidation experiments*

117 Phytodetritus and bacteria were mixed (ratio algal:bacterial cells = 1:5) in K medium
118 (Keller et al., 1987) to obtain a final volume of 100 mL. The experiments were performed in

119 triplicate. After taking three 10 mL sub-samples (controls before irradiation) the reactional
120 systems were exposed to high or low irradiance.

121

122 2.4.1. *High irradiance experiments*

123 The high-irradiance experiments were carried out in sterilized quartz and glass tubes
124 (100 mL) with an Atlas Suntest CPS + solar simulator equipped with a Xenon NXE 1500
125 lamp producing a high solar irradiance (500 W.m^{-2}) in the 300–800 nm range, i.e. close to
126 that generally observed at noon at the surface of the Mediterranean Sea (Sarraf et al., 2019)
127 and not far from the one observed at the surface of Arctic waters in summer (up to 350 W
128 m^{-2} , Horvat et al., 2020). To eliminate UV radiation, experiments were also performed in
129 glass tubes reducing the irradiance to 446 W.m^{-2} . After irradiation with a light dose of 1000
130 KJ.m^{-2} (corresponding to an irradiation time of 0.5 and 15 h at the surface and at the deep
131 chlorophyll maximum (DCM) of the Mediterranean Sea, respectively), 10 mL was collected.
132 All the samples were filtered on $0.8 \mu\text{m}$ glass fiber filters (GF/F) to concentrate bacteria
133 attached on phytodetritus and stored at -20°C until further lipid extraction.

134 Controls were also conducted in triplicate with no phytodetritus. Bacteria (*P. stutzeri*)
135 were mixed with sterilized artificial sea water complemented with 0.1 g.L^{-1} of lactate/acetate
136 (50:50) as carbon source, in 100 mL sterilized quartz or glass tubes and exposed to light in
137 the solar simulator for the same duration as the tubes containing phytodetritus and bacteria.
138 After irradiation, 50 mL was sampled and bacteria were concentrated by centrifugation
139 (Beckman & Coulter Allegra X-22 centrifuge at $20,000 \text{ g}$ for 30 min). The irradiated
140 bacterial cells were then stored at -20°C until further lipid extraction.

141

142 2.4.2. *Low irradiance experiments*

143 Low-irradiance photosynthetically active radiation (PAR) (18 W.m^{-2}) in the range
144 400–750 nm, i.e. comparable to that observed at the DCM in the Mediterranean Sea
145 (Marañón et al., 2020), was obtained with a Binder incubator equipped with Osram Lumilux
146 lamps. The vials were exposed to a light dose of 1000 KJ.m^{-2} in the incubator at 17°C . After
147 irradiation, 10 mL samples was collected, filtered on $0.8 \mu\text{m}$ GF/F and stored at -20°C until
148 further lipid extraction. Control experiments without phytodetritus were also carried out as
149 described above.

150

151 *2.5. Lipid extraction and derivatization*

152 Samples (GF/F filters or bacterial pellets) were reduced with excess NaBH_4 after
153 adding MeOH (25 mL, 30 min) to reduce labile hydroperoxides (resulting from
154 photooxidation) to alcohols, which are more amenable to gas chromatography-mass
155 spectrometry (GC-MS) analysis. Water (25 mL) and KOH (2.8 g) were then added and the
156 resulting mixture was saponified by refluxing (2 h), then cooled, acidified (HCl, 2 N) to pH
157 1, and extracted with dichloromethane ($3 \times 20 \text{ mL}$). The combined extracts were then
158 concentrated by rotary evaporation at 40°C to give total lipid extracts (TLEs).

159 Dry TLEs were derivatized by dissolving them in $300 \mu\text{L}$ of a mixture of pyridine/bis-
160 (trimethylsilyl)trifluoroacetamide (BSTFA; Supelco; 2:1, v/v) and silylated in a heating
161 block (50°C , 1 h). After evaporation to dryness under a stream of N_2 , the derivatized residue
162 was dissolved in ethyl acetate/BSTFA (to avoid desilylation) and analyzed by GC-MS/MS.

163

164 *2.6. Gas chromatography-tandem mass spectrometry (GC-MS/MS)*

165 GC-MS/MS analyses were performed on an Agilent 7890A/7010A tandem-quadrupole gas
166 chromatograph system (Agilent Technologies, Les Ulis, France) using a cross-linked 5%
167 phenyl-methylpolysiloxane (Agilent; HP-5MS ultra inert, $30 \text{ m} \times 0.25 \text{ mm}$, $0.25 \mu\text{m}$ film

168 thickness) capillary column. Analyses were performed with an injector operating in pulsed
169 splitless mode set at 270°C. Oven temperature was ramped from 70°C to 130°C at 20°C.min⁻¹,
170 then to 250°C at 5°C.min⁻¹ and finally to 300°C at 3°C.min⁻¹. Pressure of the carrier gas
171 (He) was maintained at 0.69×10^5 Pa until the end of the temperature program, then ramped
172 from 0.69×10^5 Pa to 1.49×10^5 Pa at 0.04×10^5 Pa.min⁻¹. The following mass spectrometer
173 conditions were used: electron energy 70 eV, source temperature 230°C, quadrupole 1
174 temperature 150°C, quadrupole 2 temperature 150°C, collision gas (N₂) flow 1.5 mL.min⁻¹,
175 quench gas (He) flow 2.25 mL.min⁻¹, mass range 50–700 Daltons, cycle time 313 ms.
176 Vaccenic acid photooxidation products were quantified in multiple reaction monitoring
177 (MRM) mode using the transitions m/z 339 → 131 for the parent vaccenic acid and m/z 199
178 → 129, m/z 213 → 129, m/z 357 → 129 and m/z 371 → 129 for its oxidation products
179 (Rontani, 2021). Precursor ions were selected from the most intense ions and specific
180 fragmentations observed in electron ionization (EI) mass spectra. Collision-induced
181 dissociation (CID) was optimized by using collision energies at 5, 10, 15 and 20 eV.
182 Quantification involved peak integration and determination of individual response factors
183 using standard compounds and Mass Hunter software (Agilent Technologies, Les Ulis,
184 France).

185

186 *2.7. Standard compounds*

187 Vaccenic and palmitoleic acids were obtained from Sigma-Aldrich. Vaccenic acid
188 oxidation was achieved using Fe²⁺/ascorbate (Loidl-Stahlhofen and Spiteller, 1994).
189 Subsequent reduction of the resulting hydroperoxides in methanol with excess NaBH₄
190 afforded the corresponding hydroxyacids.

191

192 *2.8. Statistical analysis*

193 Results of statistical tests were considered to be significant at a confidence level of
194 95% ($\alpha = 0.05$). All tests were performed using XL-Stat Statistics software.

195

196 **3. Results and Discussion**

197

198 *3.1. In vitro experiments*

199 To determine the effect of solar irradiance on the efficiency of transfer of $^1\text{O}_2$ from
200 phytodetritus to their attached bacteria, senescent *Thalassiosira* sp. cells associated with *P.*
201 *stutzeri* were irradiated by a light dose of 1000 KJ.m^{-2} under contrasting irradiances, i.e. 18
202 (PAR), 446 (PAR) and 500 (UV+PAR) W.m^{-2} . The efficiency of $^1\text{O}_2$ transfer to bacteria was
203 estimated based on the measured concentration of photooxidation products of vaccenic acid
204 (pmol.L^{-1}). Vaccenic acid was present in high proportion in *P. stutzeri* (Rainey et al., 1994)
205 but only in weak amount in the unknown bacterial community associated with the non-
206 axenic strain of *Thalassiosira* sp. employed (amount estimated to be more than one order of
207 magnitude lesser than this present in the cells of *P. stutzeri* added during the different
208 experiments).

209 Type-II photosensitized oxidation of vaccenic acid involves the addition of $^1\text{O}_2$ to the
210 two carbon atoms of its Δ^{11} double bond, and leads to the formation of 11-
211 hydroperoxyoctadec-12(*trans*)-enoic and 12-hydroperoxyoctadec-10(*trans*)-enoic acids
212 (Rontani, 2021). These hydroperoxides can subsequently undergo stereoselective radical
213 allylic rearrangement to afford 13-hydroperoxyoctadec-11(*trans*)-enoic and 10-
214 hydroperoxyoctadec-11(*trans*)-enoic acids, respectively (Porter et al., 1995). *Trans* allylic
215 hydroxy acids arising from NaBH_4 reduction of these hydroperoxyacids are sufficiently
216 stable for use as tracers of type-II photosensitized oxidation processes (Marchand and
217 Rontani, 2001).

218 Interestingly, we detected only trace amounts of vaccenic acid photooxidation
219 products before irradiation (under the detection limit) and in the different controls carried
220 out without algae (Table 1, Fig. 2). Significant increases of vaccenic acid photooxidation
221 products were observed **with algae amendment after 0.5 and 15H for low and high irradiance**
222 **respectively** (Wilcoxon test, $n=6$, $p=0.031<0.05$). As previously observed (Rontani et al.,
223 2003; Petit et al., 2015), photooxidation of heterotrophic bacteria membranes requires the
224 presence of senescent phytoplankton cells. Our results showed a strong effect of intensity of
225 irradiance on the photooxidation of the bacterial membranes (Table 1, Fig. 2). Indeed, the
226 concentration of vaccenic acid photooxidation products reached $2741 \pm 21 \text{ nmol.L}^{-1}$ under
227 high PAR irradiance but only $464 \pm 190 \text{ nmol.L}^{-1}$ under low irradiance (Fig. 2). The highest
228 concentration of vaccenic acid oxidation products ($3998 \pm 1010 \text{ nmol.L}^{-1}$) was observed
229 under high UV+PAR irradiance (Table 1, Fig. 2) and may be attributed to the presence of
230 additional UV radiations. In phytodetritus, chlorophyll can act as photosensitizer not only
231 under PAR (Nelson, 1993) but also under UV (Ying He and Häder, 2002), since they absorb
232 in the UV range (Queseda and Vincent, 1997; Ehling-Schulz and Scherer, 1999). This
233 assumption is well supported by the profile of photooxidation products obtained under these
234 conditions, showing the presence of a high proportion of allylic 11- and 12-hydroxyacids
235 with a cis double bond (Fig. S1) typical of the UV-induced photooxidation of MUFAs
236 (Christodoulou et al., 2010).

237 Amiraux et al. (2016) previously showed in senescent diatom cells that low irradiances
238 induce a slow but long-lasting production of $^1\text{O}_2$ that strongly oxidizes algal unsaturated
239 lipids and sterols, through a process called “photodynamic effect” (Knox and Dodge, 1985;
240 Skovsen et al., 2005), whereas high irradiances lead to stronger but shorter production of $^1\text{O}_2$
241 that strongly photooxidizes the sensitizer (chlorophyll) through a process called
242 “photobleaching” but with only a weak photodynamic effect (Amiraux et al., 2016; Rontani

243 et al., 2021). This effect of the intensity of solar irradiance on the efficiency of the
244 photodynamic effect in senescent phytoplankton cells was then confirmed *in situ* (Rontani
245 et al., 2021).

246 The effect of irradiance on the efficiency of photooxidation of attached bacteria thus
247 appears to be exactly the opposite of the effect on phytoplankton. Under low irradiance, the
248 low flux of $^1\text{O}_2$ produced reacts with the unsaturated membrane components of phytodetritus
249 and thus gets quenched before it reaches attached bacteria (Fig. 3). In contrast, under strong
250 irradiance, a higher proportion of $^1\text{O}_2$ is efficiently transferred to the attached bacteria,
251 strongly oxidizing their unsaturated lipids (Fig. 3). This new concept opens interesting
252 perspectives in the study of phytoplankton-bacteria relationships.

253

254 *3.2. Confirmation of the results obtained in vitro in samples collected in situ*

255 To confirm the enhancement of the efficiency of photooxidation processes on bacteria
256 attached to phytoplankton cells, we analyzed a variety (n=58) of sinking particulate matter
257 and sea ice samples collected in the Arctic (Fig. 1), where irradiance at the sea ice–water
258 interface and in the water column varies strongly according to the albedo of snow and ice
259 and to snow and ice thickness (Chresten Lund-Hansen et al., 2021). Sinking particulate
260 matter samples were collected: (i) in summer at 100 m in open water in the Beaufort Sea
261 (Rontani et al., 2012), (ii) in summer at 25 m in the marginal ice zone of central Baffin Bay
262 (Rontani et al., 2022), and (iii) in spring at 5 m and 30 m in ice-covered Resolute Passage
263 (Rontani et al., 2016). Sea-ice samples (0–10 cm) were collected in spring during the
264 GreenEdge campaign at Qikiqtarjuaq (Baffin Bay, Arctic Ocean; Amiraux et al., 2020). To
265 compare the efficiency of type-II photooxidation processes in algal and bacterial material,
266 we used the ratio of % vaccenic acid photooxidation to % palmitoleic acid photooxidation.
267 Vaccenic acid is generally considered to be specific to bacteria (Lambert and Moss, 1983;

268 Sicre et al., 1988), whereas palmitoleic acid is the main fatty acid component of sympagic
269 diatoms (Fahl and Kattner, 1993; Falk-Petersen et al., 1998; Leu et al., 2010). Although
270 palmitoleic acid is also present in several bacteria (e.g. de Carvalho and Caramujo, 2014),
271 the contribution of bacteria to palmitoleic acid in the samples analyzed (dominated by
272 diatoms) is negligible relative to the contribution of sympagic algae.

273 The results obtained show higher % vaccenic acid photooxidation/% palmitoleic
274 photooxidation ratios in sinking particles collected in open-water Beaufort Sea than in ice-
275 covered Resolute Passage (Fig. 4A). This difference may be attributed to the mean PAR
276 irradiance in the mixed surface layer, which is considerably higher in open water than in ice-
277 covered zones (e.g. 365 ± 62 and $10.9 \pm 2.7 \mu\text{mol photons.m}^{-2}.\text{s}^{-1}$ during the spring-summer
278 transition in Beaufort Sea, respectively; Alou-Font et al., 2016). The ratios observed in the
279 Baffin Bay marginal ice zone (where ice concentration ranges from 15% to 80%) were
280 logically intermediate (Fig. 4A). The decrease in the ratio observed between 5 and 30 m in
281 the Resolute Passage (Fig.4A) could be attributed to the presence in the deeper trap of highly
282 aggregated senescent sea-ice algae settling sufficiently rapidly out of the euphotic zone to
283 avoid significant photooxidation (Rontani et al., 2016), however these differences **were not**
284 **significant** (p-value > 0.05, see Fig. 4A, Table S1).

285 Very high ratios were found in the bottommost 10 cm of sea ice collected at the
286 GreenEdge ice camp in 2016 (Fig. 4B). Under 75 cm of sea ice, irradiance can reach 105.9
287 $\mu\text{mol photons m}^{-2}.\text{s}^{-1}$ without snow cover and decrease to $51.9 \mu\text{mol photons m}^{-2}.\text{s}^{-1}$ with a
288 snow cover of 1 cm (Lund-Hansen et al., 2021). In this solid habitat, the bacteria–sympagic
289 algae association may thus be maintained at relatively high irradiances during long periods,
290 these conditions strongly favoring the photooxidation of bacteria. It was previously
291 demonstrated that during the early stages of ice melt, sympagic bacteria undergo an intense
292 osmotic stress in hypersaline ice brines (Amiriaux et al., 2017) and are affected later in the

293 season by the release of bactericidal free palmitoleic acid by sympagic algae under the effect
294 of light stress (Amiriaux et al., 2020). Our results show that these organisms may also
295 undergo a strong photooxidative stress in the ice resulting from a strong and efficient transfer
296 of $^1\text{O}_2$ from senescent sympagic algae. These different stresses are thought to strongly
297 decrease the mineralization potential of bacteria attached to ice algae and thus favor the
298 preservation of sympagic material in Arctic sediments (Amiriaux et al., 2021; Rontani et al.,
299 2022).

300

301 **4- Conclusion**

302 Incubation of senescent cells of the diatom *Thalassiosira* sp. associated with the
303 bacteria *P. stutzeri* under **contrasting artificial** light irradiances made it possible to
304 demonstrate that high irradiances strongly favor photooxidation of the unsaturated
305 membrane components of bacteria attached to senescent phytoplankton cells. Under these
306 conditions, only a small part of $^1\text{O}_2$ produced in chloroplasts reacts with membrane
307 components of senescent algae, and so the remainder is efficiently transferred to their
308 attached bacteria, inducing strong **oxidative damage**. Conversely, low irradiances favor
309 photooxidation of the unsaturated algal lipids rather than attached bacteria. Indeed, $^1\text{O}_2$
310 slowly generated under these conditions reacts with unsaturated lipids of the algal cells and
311 is thus quenched before it can reach bacterial membranes. These findings were also
312 confirmed *in situ*, where we observed the same tendencies in a variety of Arctic
313 environmental samples (sea ice and sinking particulate matter). The transfer of $^1\text{O}_2$ from
314 senescent algae to their attached bacteria appeared to be particularly efficient in sea ice where
315 the sympagic algae–bacteria association is maintained at relatively high irradiances during
316 long periods of time.

317

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335

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337

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- 485

486 **FIGURE CAPTIONS**

487

488 **Fig. 1.** Map showing the location of the different stations investigated. Sinking particles were
489 collected in open water, first-year ice, and marginal ice zones in Beaufort Sea, Resolute
490 Passage and central Baffin Bay, respectively. Sea ice samples originated from Davis Strait.

491

492 **Fig. 2.** Measurement of vaccenic acid photooxidation products in *P. stutzeri* cultures with
493 and without addition of *Thalassiosira* sp. cells after irradiation (1000 KJ.m⁻²) under low
494 PAR, high PAR and high PAR+UV irradiances (n=3).

495

496 **Fig. 3.** Conceptual scheme of the effect of ¹O₂ on bacteria attached to senescent
497 phytoplankton cells under low or high irradiance.

498

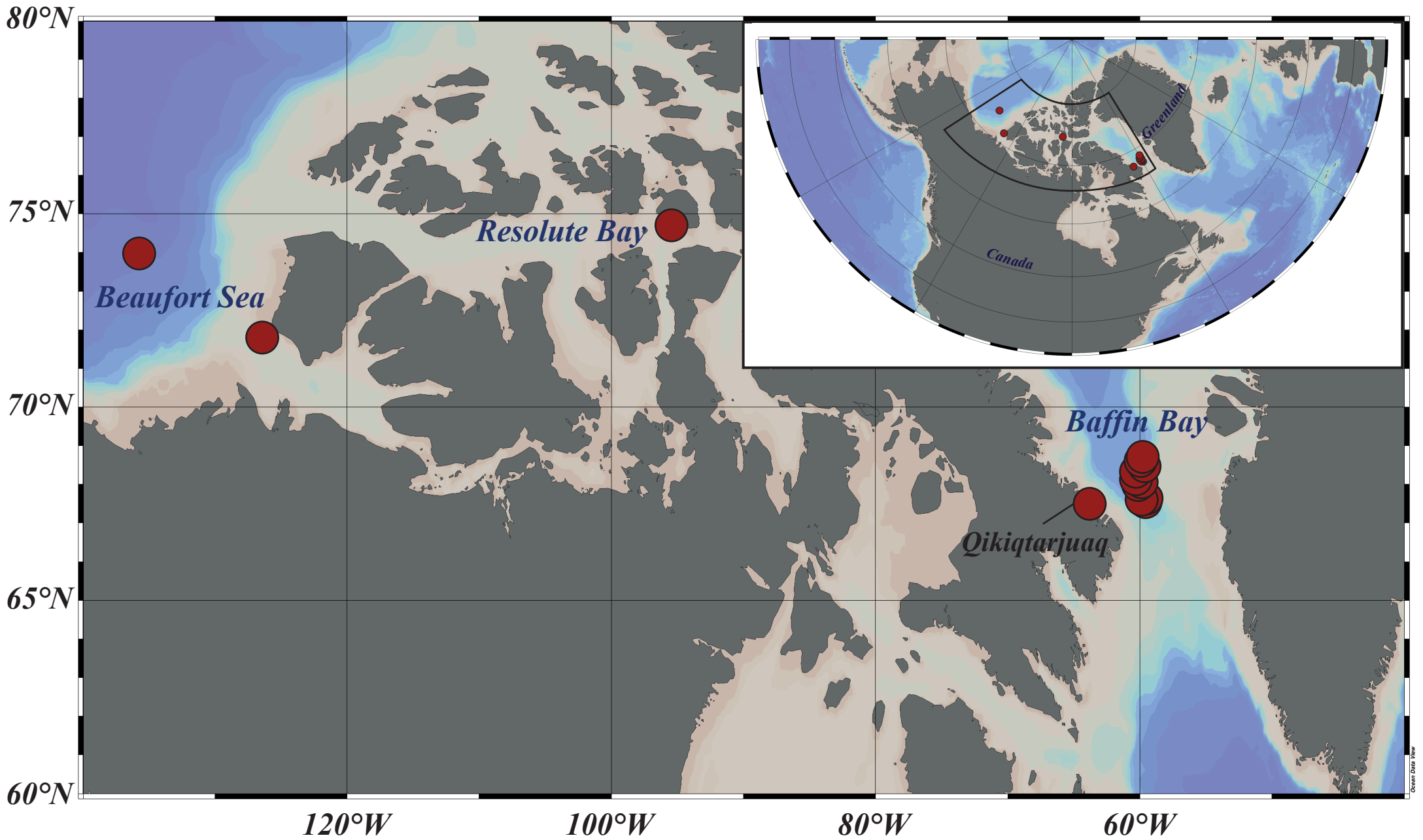
499 **Fig. 4.** Values of the ratio of % vaccenic acid photooxidation to % palmitoleic acid
500 photooxidation in particulate matter samples collected in the Arctic under different
501 irradiance conditions (A) and in sea-ice samples collected at the GreenEdge 2016 ice camp
502 (B). Kruskal-Wallis test,

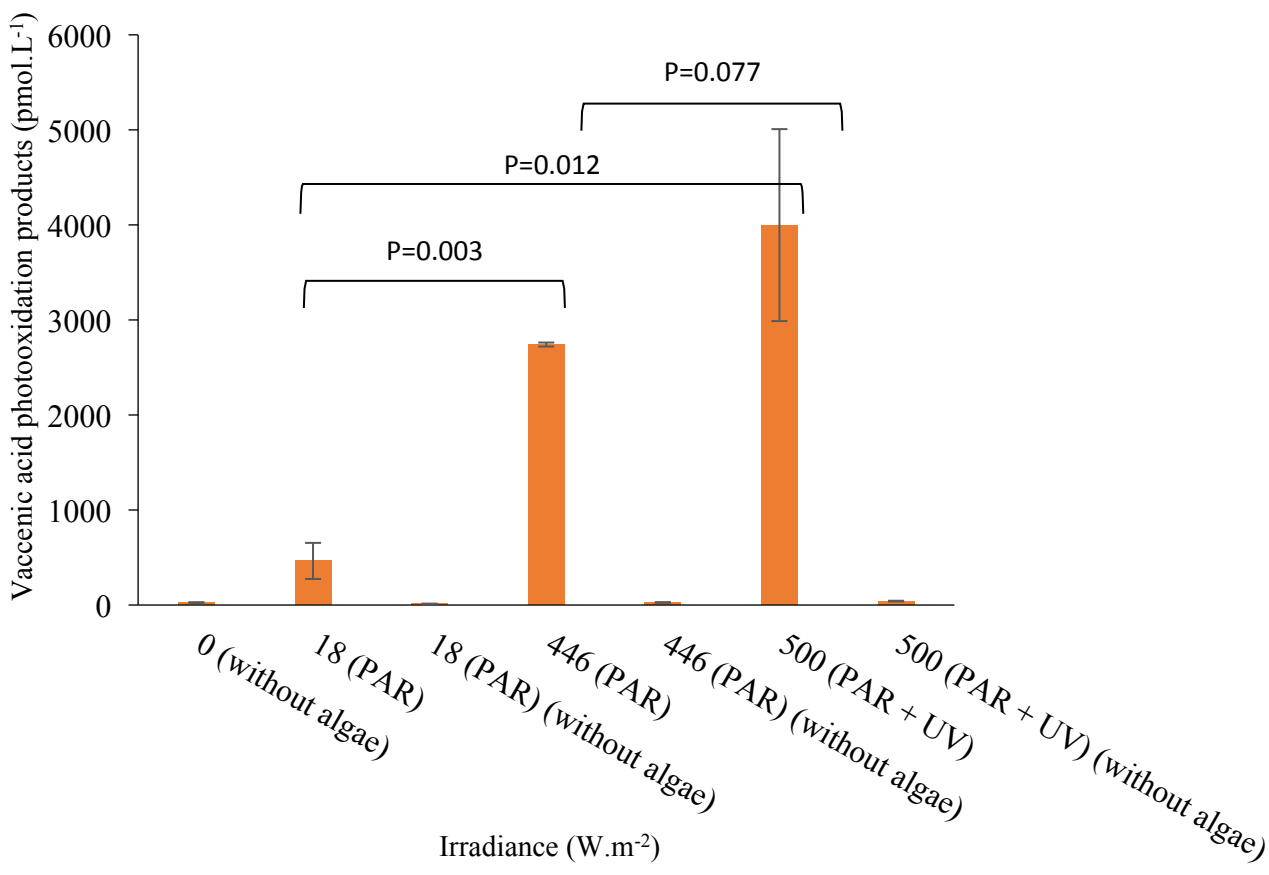
503 p-values : 0 < "****" < 0.001 < "***" < 0.01 < "*" < 0.05 < " "

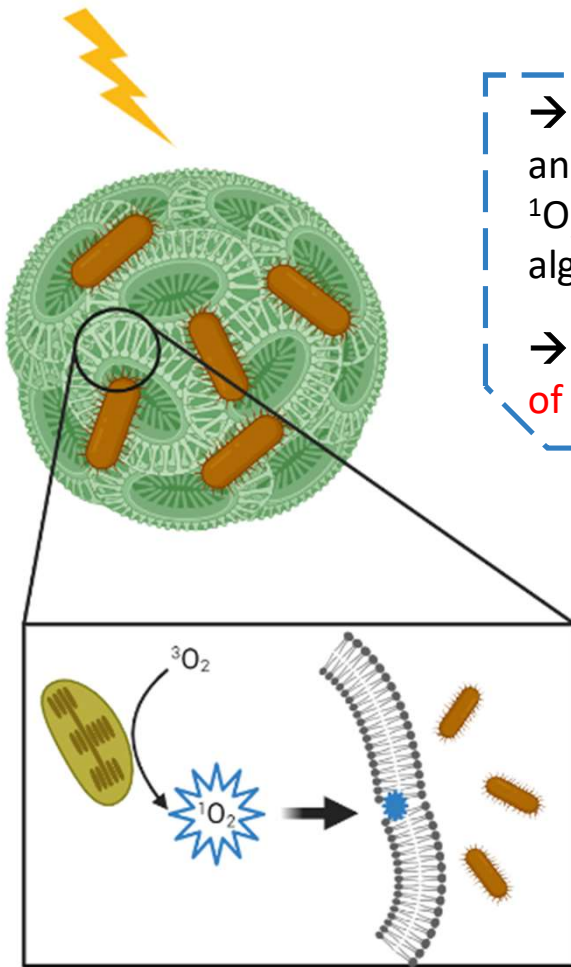
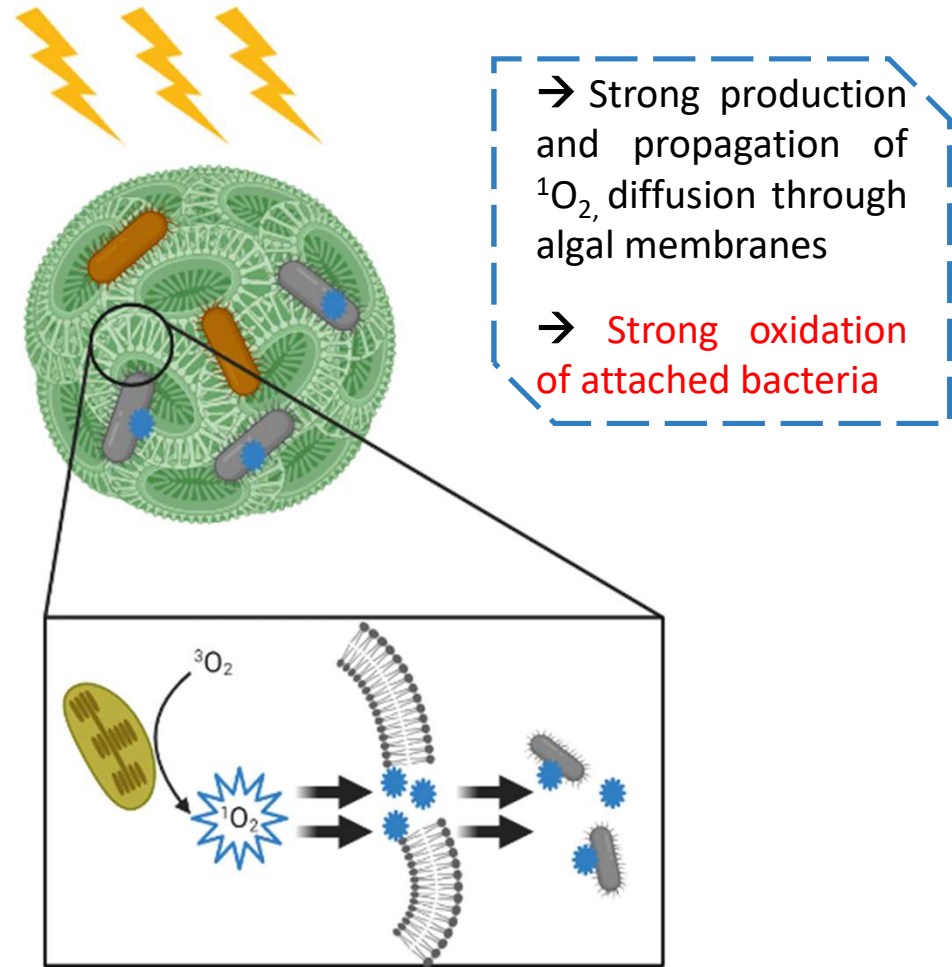
504

505 **Fig. S1.** MRM chromatograms (with the transitions *m/z* 357 → 129 and *m/z* 199 → 129)
506 showing the production of significant proportions of 12-hydroxyoctadec-10(*cis*)-enoic and
507 11-hydroxyoctadec-12(*cis*)-enoic acids (indicative of an effect of UV radiation,
508 Christodoulou et al., 2010), respectively, after irradiation of *Thalassiosira* sp. cells
509 associated with *P. stutzeri* under high solar irradiance (500 W.m⁻²) for 3h 30.

510





Low irradiance**High irradiance**

Algal cell



Healthy bacteria



Lipid membrane



Chloroplast



Singlet oxygen

Bacteria damaged by $^1\text{O}_2$

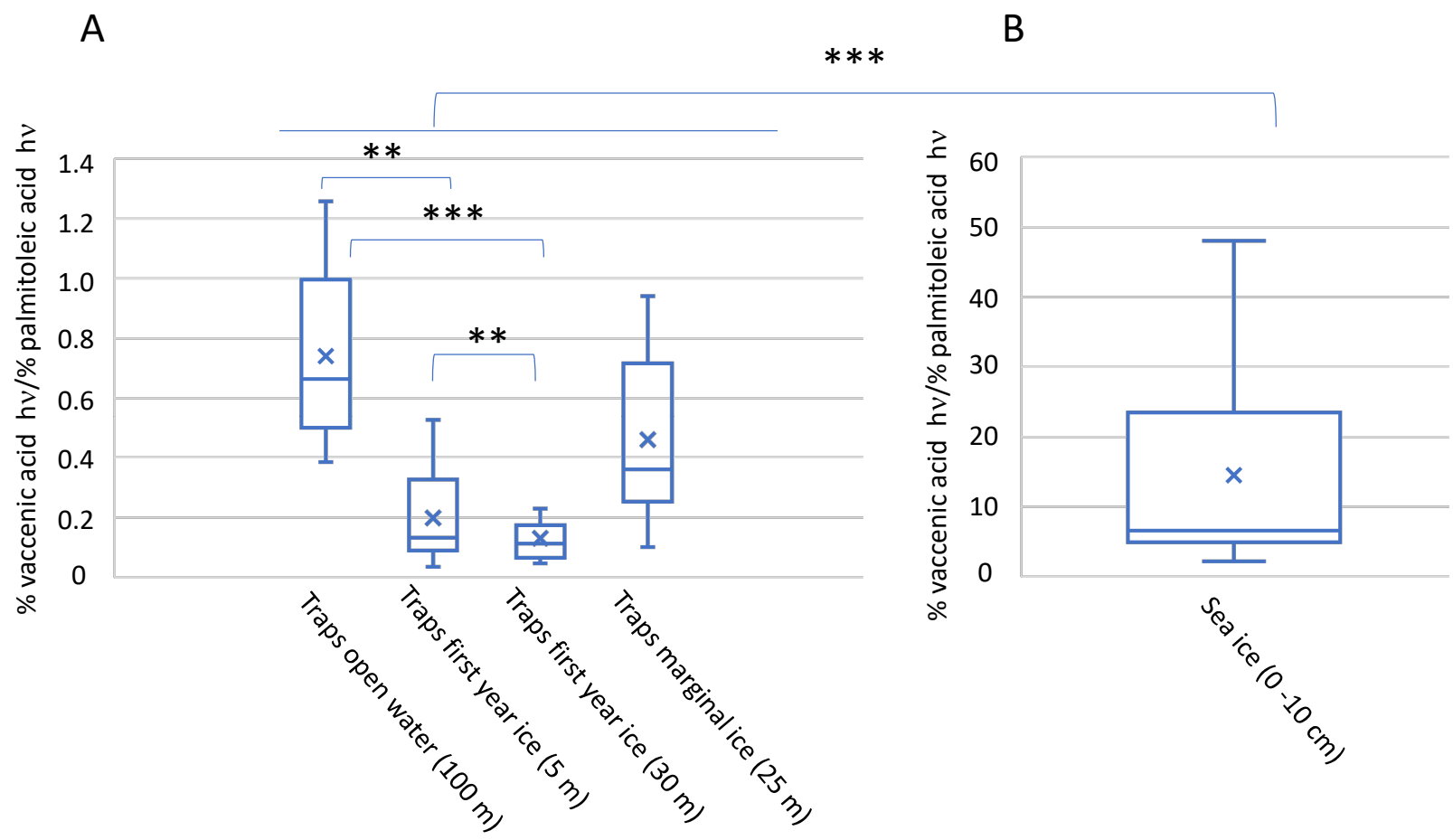


Table 1. Concentration of vaccenic acid photooxidation products in *P. stutzeri* cultures with and without addition of *Thalassiosira* sp. cells after irradiation (1000 KJ.m⁻²) under low PAR, high PAR and high PAR+UV irradiances (n=3).

Irradiance (W m ⁻²)	Vaccenic acid hv (pmol.L ⁻¹) ^a	Standard deviation
0 (without algae)	24	6
18 (PAR)	464	190
18 (PAR) (without algae)	13	0
446 (PAR)	2741	21
446 (PAR) (without algae)	27	3
500 (PAR + UV)	3998	1010
500 (PAR + UV) (without algae)	41	4

^a Mean values.

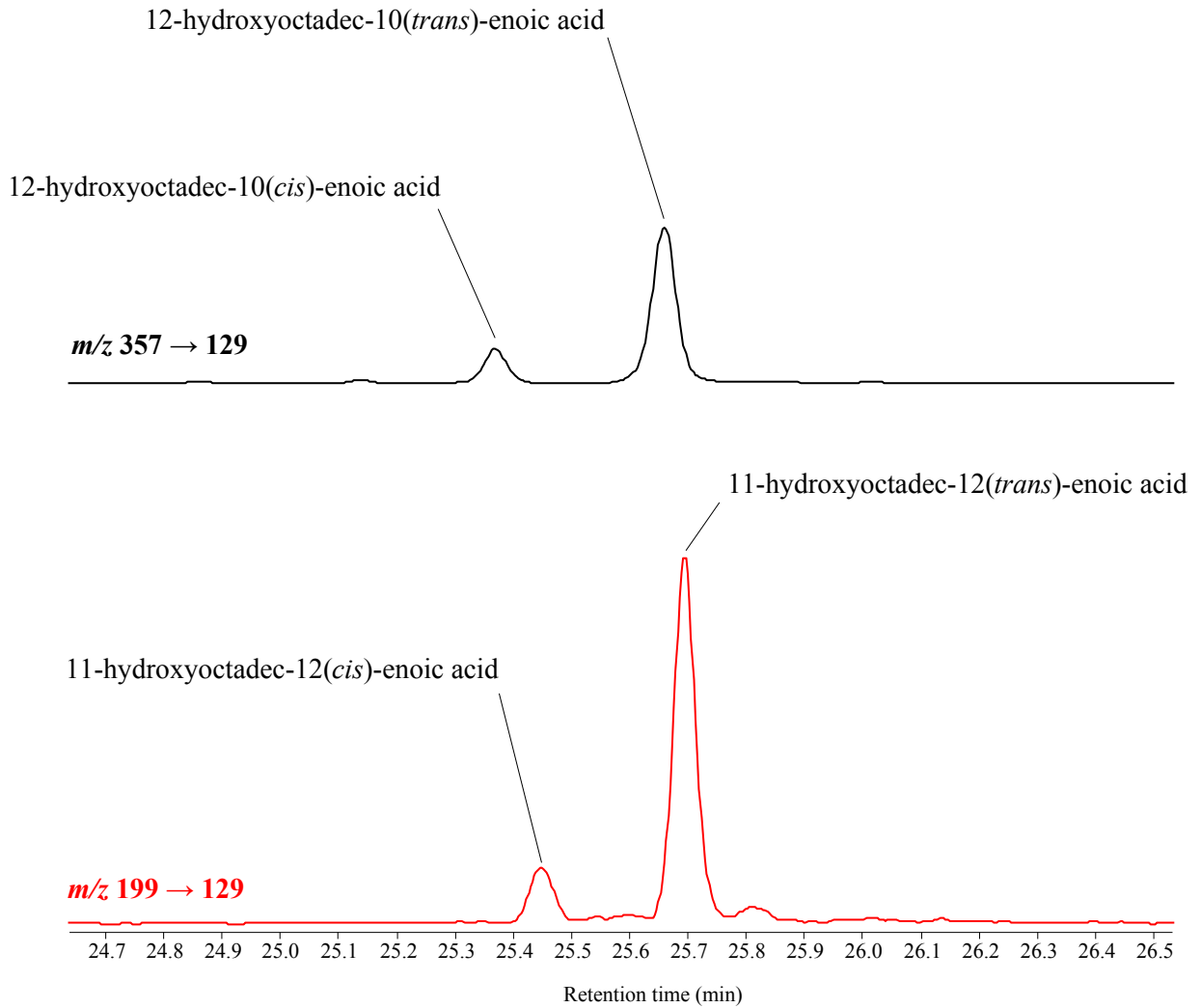


Table S1: Significant differences between % vaccenic acid photooxidation to % palmitoleic acid photooxidation ratios measured in the Arctic under different irradiances (P-Values, n = 3, KW test)

	Traps under open water (100 m) Malina	Traps under first year ice (5 m) (Resolute Passage)	Traps under first year ice (30 m) Resolute passage	Traps under marginal ice zone (30 m) Baffin Bay	Sea ice
Traps under open water (100 m) Malina	1	0,005	0,000	0,109	0,001
Traps under first year ice (5 m) (Resolute Passage)	0,005	1	0,997	0,111	0,002
Traps under first year ice (30 m) Resolute passage	0,000	0,997	1	0,004	0,001
Traps under marginal ice zone (30 m) Baffin Bay	0,109	0,111	0,004	1	0,001
Sea ice	0,001	0,002	0,001	0,001	1