



HAL
open science

**Do early diagenetic processes affect the applicability of commonly-used organic matter source tracking tools?
An assessment through controlled degradation
end-member mixing experiments**

Morgane Derrien, Heybin Choi, Emilie Jardé, Kyung-Hoon Shin, Jin Hur

► **To cite this version:**

Morgane Derrien, Heybin Choi, Emilie Jardé, Kyung-Hoon Shin, Jin Hur. Do early diagenetic processes affect the applicability of commonly-used organic matter source tracking tools? An assessment through controlled degradation end-member mixing experiments. *Water Research*, 2020, 173, pp.Art. n°115588. 10.1016/j.watres.2020.115588. insu-02475407

HAL Id: insu-02475407

<https://insu.hal.science/insu-02475407v1>

Submitted on 19 Mar 2020

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

1 **Do early diagenetic processes affect the applicability of**
2 **commonly-used organic matter source tracking tools? An**
3 **assessment through controlled degradation end-member mixing**
4 **experiments**

5
6
7 Morgane Derrien^{a,*}, Heybin Choi^b, Jardé Emilie^c, Kyung-Hoon Shin^b, and Jin Hur^{a,*}

8 ^a*Department of Environment and Energy, Sejong University, Seoul 143-747, South Korea*

9 ^b*Department of Environmental Marine Sciences, Hanyang University, Ansan, Gyeonggi do*
10 *15588, South Korea*

11 ^c *University of Rennes 1, CNRS, Géosciences Rennes, UMR 6118, Rennes, France*

12
13
14
15 Re-submitted to *Water Research*, January 2020.

16
17
18
19
20 * Corresponding author

21 Tel. +82-2-6935-2634

22 E-Mail: morganederrien@sejong.ac.kr; jinhur@sejong.ac.kr

23 **ABSTRACT**

24 In the development of organic matter (OM) source tracking tools, it is critical to validate
25 if (1) the tracers are conservative with source mixing, and (2) they can be conservative under
26 diagenetic processes (e.g., microbial degradation). In this study, these two critical points were
27 rigorously tested for three commonly-used source tracking tools (i.e., absorbance and
28 fluorescence proxies, stable carbon isotopes and lipid biomarkers) via a controlled experiment at
29 laboratory scale. To this end, two end-members (e.g., soil and algae), which represent the most
30 common and contrasted sources of OM to sediments in an aquatic environment, were mixed in
31 different ratios and then incubated under different oxygen conditions (oxic versus anoxic) in the
32 dark at 25°C for 60 days. The initial and final signals of the source tracking tools were analyzed
33 and compared for each mixing ratio. Based on three evaluation criteria concerning the linearity
34 of the relationships, discrimination sensitivity, and conservative mixing behavior, we evaluated
35 the applicability of the tools to trace the sediment organic matter in the aquatic environment.
36 Although most of the source tracking proxies evaluated in this study showed a conservative
37 nature after incubation, there are only a few that demonstrated both conservative behaviors with
38 the sources mixing and under early diagenetic processes. The fluorescence proxies such as the
39 relative distribution of a humic-like component associated with refractory source material
40 (Ex/Em: 220/430nm), modified fluorescence index (YFI), humification index (HIX), and carbon
41 stable isotope ratios were identified to be the most reliable tracers for tracking sedimentary OM
42 sources under early diagenetic processes. This study provides strong insights into the validation
43 of common OM source tracking tools for sediment and a reasonable guideline to select the
44 optimum indices for source discrimination via end-member mixing analysis.

45

46 **Keywords:** Sources tracking tools; Organic matter; Sediments; Biodegradation; End-member
47 mixing.

48

49 **1.INTRODUCTION**

50 Natural organic matter (NOM) is generated by the breakdown and degradation of
51 organisms involving hydrosphere, biosphere, and geosphere through diverse biological,
52 chemical, and physical processes in the natural environment (Sillanpää, 2015). NOM sources are
53 usually classified as allochthonous or autochthonous. Allochthonous OM, which occurs from
54 outside the aquatic environment, consists of terrigenous materials, such as vascular plants,
55 leaves, root exudates, and soils, exported from the upstream catchment into rivers and lakes. By
56 contrast, autochthonous OM, which is produced within water bodies, derives from aquatic biota
57 (e.g., algae, bacteria, plankton, macrophytes, and nekton) (Derrien et al., 2019a; Volkman and
58 Tanoue, 2002). In aquatic systems, sediments represent a large reservoir of nutrients and NOM
59 from both inputs in various proportions (Briand et al., 2015; Fisher et al., 2005; Southwell et al.,
60 2010; Waterson and Canuel, 2008; Zhang et al., 2009). Indeed, sedimentary OM is derived from
61 bacteria or plankton formed in situ, but also receives allochthonous OM from the upstream
62 catchment. Soil OM is a representative allochthonous OM source and it is easily transported into
63 the rivers and ends up in sediments through hydrological processes (van der Meij et al., 2018).
64 Nowadays, terrestrial organic matter input into surface waters is one of the major concerns in the
65 context of the current climate changes as it is observed in polar regions with the release of
66 “dormant” carbon from permafrost (Bischoff et al., 2016; Vonk et al., 2015; Wild et al., 2019).
67 Sediments are also a reactive compartment where diagenetic processes occur inducing changes.
68 The processes may be physical, chemical, and/or biological in nature and may occur at any time

69 subsequent to the arrival of a particle at the sediment-water interface (Henrichs, 1992;
70 Kuznetsova et al., 2019; Milliken, 2003). Among the early diagenetic processes, biodegradation
71 plays a key role as it is one of the main processes causing changes in amount, composition and
72 properties of OM in sediment (Arndt et al., 2013; Derrien et al., 2019a; Guenet et al., 2014).

73 Investigating the processes that control the composition of sedimentary OM is crucial for
74 a thorough understanding of OM dynamics and its role in the carbon cycle at a local and global
75 scale (Gordon and Goni, 2003; Pedrosa-Pàmies et al., 2015). Identifying, apportioning and
76 tracking the sources of OM in aquatic systems require robust, reliable and effective source
77 tracking tools. Absorbance and fluorescence proxies, stable carbon isotopes, and lipid
78 biomarkers are the most commonly-used OM source tracking tools in aquatic systems (Aiken,
79 2014; Bianchi and Canuel, 2011; Derrien et al., 2017; Meyers, 1994). These tools have been
80 applied in diverse environments including soils, sediments, pore-water, river, lakes, and oceans
81 for several decades (Amiotte-Suchet et al., 2007; Benner et al., 1997; Derrien et al., 2015; Fichot
82 et al., 2013; Jaffé et al., 2004; Meyers and Ishiwatari, 1993; Toming et al., 2013; Wakeham and
83 Canuel, 1990; Zimmerman and Canuel, 2001; Zsolnay et al., 1999). Although these tools have
84 been used for decades, their applicability and potential limitations have not been rigorously
85 examined yet. Nevertheless, to fully validate their use, two critical points need to be examined:
86 1) conservative behavior with sources mixing, and 2) conservative behavior under early
87 diagenetic processes (e.g., microbial degradation). In our previous study, based on a simple
88 mixing POM end-member experiment, we demonstrated that the conservative behavior of the
89 OM source tracking tools with the mixing source is not obvious and finally raised the question of
90 the reliability of some of the most commonly-used tools (Derrien et al., 2019b). More recently,
91 we also investigated the biodegradation-induced changes in the porewater DOM for different

92 sources (Derrien et al., 2019c). The results of the latter study suggested the existence of different
93 pathways of biodegradation with both positive and negative priming effects depending on the
94 sources and the ratio of labile material. All these observations clearly justify the questioning of
95 the applicability to these tools in natural systems and the necessity to test them for both criteria.

96 In this context, we designed a controlled degradation experiment at laboratory scale using
97 organic-rich sediments artificially composed of two contrasting OM end-members (i.e., soil and
98 algae) at known mixing ratios. The incubations were performed under oxic and anoxic conditions
99 in order to reproduce different configurations of natural sediment (Kristensen, 2000). Hence, this
100 study aimed to establish the applicability of 3 commonly-used source tracking tools (i.e.,
101 absorbance and fluorescence proxies, stable carbon isotopes, and lipid biomarkers) during early
102 diagenetic processes in the natural environment. As a result, the specific objectives of this study
103 were (i) to study the influence of oxygen on the degradation-induced changes, and (ii) to
104 examine the conservative/non conservative behaviors of the three source discrimination tools in
105 artificial sediments before and after biodegradation, and (iii) to evaluate the conservative/non
106 conservative behaviors of the three source discrimination tools with sources mixing in the
107 artificial sediments.

108

109 **2. MATERIAL AND METHODS**

110 **2.1. Experimental design of the incubation experiment**

111 Two end-members (i.e., soil and algae), as the major organic sources of sediments, were
112 mixed at soil to algae ratios of 100:0, 75:25, 50:50, 25:75, and 0:100, respectively, on the basis
113 of organic carbon (OC) concentrations, not their masses, after the OC contents of the two end-
114 members were taken into account. Briefly, a topsoil (0-10cm) sample was collected as the soil

115 end-member at Bukhansan National Park (37°43'37.0" N 127°00'50.9") in South Korea. A
116 commercial unicellular green alga (*Chlorella vulgaris*), which is commonly found in lakes and
117 ponds in South Korea, was purchased as the algae end-member from Aquanet Co., Ltd. in
118 Gyeongsangnam-do, South Korea. Detailed information on the two end-members can be found
119 in Derrien et al., 2019b.

120 Incubation experiments in oxic and anoxic conditions were performed in pre-washed and
121 pre-combusted (450°C for 4 hours) 125 ml Wheaton® amber glass bottles with Teflon
122 screwcaps. Samples for anoxic incubation were prepared and sampled using sterile Aldrich®
123 AtmosBag two-hand glove bag. A mass (12 g) of the end-member mixture samples were mixed
124 with ultrapure water (Barnstead™ Easypure™ RoDi, Thermo Scientific) at a solid to solution
125 ratio of 1:4 and equilibrated for 48 h in the dark at room temperature after shaking at 100 rpm for
126 one hour. River water from Jungnang river in Seoul (37° 40' 16"N, 127° 04' 47"E) was used as
127 inoculum for microbial incubation. The collected river water sample was first passed through a
128 GF/C (pre-combusted, 1.2 µm pore-sized) and 10 mM of inorganic nutrients (NH₄NO₃ and
129 K₂HPO₄) was added to the filtered river water for sufficient microbial growth. Then, the
130 inoculum solution was covered by aluminum foil to protect the solution from light to avoid the
131 growth of algae and finally kept at room temperature under slow shaking (80 rpm) for 2 days.
132 Prior to incubation, all artificial sediment samples were spiked with 3% (v/v) of the prepared
133 inoculum, and a sufficient amount (1%, v/v) of nutrients (NH₄NO₃ and K₂HPO₄ at 10 mM) was
134 added to each sample to avoid nutrient limitations during the incubation. Finally, the samples in
135 oxic and anoxic conditions were incubated in the dark at 25°C for 60 days (Guenet et al., 2014;
136 Navel et al., 2012). No control was prepared because it required the poisoning of the samples
137 with either mercuric chloride or sodium azide, which are extremely toxic and environmentally

138 harmful and more importantly result in fluorescence quenching (Park and Snyder, 2018;
139 Retelletti Brogi et al., 2019). Samples under oxic conditions were aerated every 3 days under a
140 fume hood in order to minimize the contamination by microorganisms in the air. Samples were
141 sacrificed for sampling on day 0 and day 60, with “day 0” corresponding to the day when the
142 samples were inoculated. The degradation experiments were carried out in duplicate.

143 The sampling was performed according to the following. First, samples were taken from the
144 incubator and the overlying water was carefully removed not to disturb the sediment. The
145 artificial sediment samples were then centrifuged at 5000 rpm for 20 min to remove the
146 porewater. Finally, the porewater-free sediment samples were freeze-dried, ground, and
147 homogenized for further analyses.

148

149 **2.2. Spectroscopic measurements**

150 To investigate the absorbance and fluorescence properties of the artificial sediment, it is
151 necessary to perform an extraction of the OM (e.g., extraction of the dissolved OM from the
152 sedimentary OM). Extraction can be performed with different solvents. According to our
153 previous results comparing common solvent-based OM extraction methods, the water extraction
154 was identified as the preferred extraction method for the application of the spectroscopic tool to
155 POM source discrimination (Derrien et al., 2019b). In this study, the same procedure was used to
156 obtain the water-extractable organic matter (WEOM) of the artificial sediment samples.

157 The dissolved organic carbon (DOC) concentrations were measured using a Shimadzu V-
158 CPH TOC analyzer. The absorption spectra were scanned from 200 to 800 nm at 0.5 nm-interval
159 using an ultraviolet-visible (UV-vis) spectrometer (Shimadzu UV-1300). The fluorescence
160 excitation-emission matrices (EEMs) were obtained with a luminescence spectrometer (Hitachi

161 F7000, Japan) following the procedure previously described by Retelletti Brogi et al. (2019). A
162 total of 82 EEMs were collected for the PARAFAC modeling, and it was processed using
163 MATLAB R2013b (Mathworks, USA) with the drEEM toolbox (Murphy et al., 2013). The
164 validation of the PARAFAC model was made by split-half analysis and percentage of explained
165 variance (>99.5%). By using corrected EEMs, several classical fluorescence indices (e.g.,
166 fluorescence index and modified fluorescence index: FI and YFI, the humification index: HIX,
167 and the index of recent autochthonous contribution BIX) were calculated (Heo et al., 2016;
168 Huguet et al., 2009; Zsolnay et al., 1999). The maximum fluorescence intensities (F_{\max}) of the
169 identified PARAFAC components were used to represent their relative abundance (%).

170

171 **2.3. Carbon and nitrogen stable isotope ratio analyses**

172 Before carbon stable isotope analysis, inorganic carbon was removed by 1N HCl
173 treatment, whereas untreated samples were directly used for the nitrogen isotope ratio analysis
174 (Carabel et al., 2006). The total organic carbon (TOC) and the carbon stable isotope ratio ($\delta^{13}\text{C}$)
175 were measured using an elemental analyzer that was coupled with an isotope ratio mass
176 spectrometer (EA-IRMS; EuroEA-Isoprime IRMS, GV Instruments, UK). Stable isotope ratios
177 were calculated using the standard d notation:

$$\delta^{13}\text{C}(\text{‰}) = \left((R_{\text{sample}}/R_{\text{reference}}) - 1 \right) \times 1000 \quad (1)$$

178 where R is the corresponding ratio of $^{13}\text{C}/^{12}\text{C}$. The standard reference material was IAEA-CH-3,
179 Cellulose ($\delta^{13}\text{C} = -24.724 \pm 0.041$ (vs VPDB, Vienna Pee Dee Belemnite)). The analytical
180 precision was 0.05‰.

181

182 **2.4. The sterols/stanols analysis**

183 The total lipid fraction of the artificial sediments was extracted with dichloromethane
184 using an accelerated solvent extractor (ASE 200, Dionex). Conditions of the extraction are
185 described in Derrien et al. (2011). The total lipid extract was then fractionated using solid/liquid
186 chromatography (silica column) to isolate the polar fraction. The polar fraction was derivatized
187 with a mixture of N,O-bis-(trimethylsilyl)trifluoroacetamide and trimethylchloro-silane (BSTFA
188 + TMCS, 99/1, v/v, Supelco) after addition of 5 α -cholestane (CDN isotope) as an internal
189 standard (IS). Derivatized samples were analyzed by gas chromatography-mass spectrometry
190 (GC-MS) using a Shimadzu QP2010plus equipped with a capillary column (Supelco,
191 60m \times 0.25mm ID, 0.25 μ m film thickness). The temperature of the transfer line was set at 280 $^{\circ}$ C,
192 and molecules were ionized by electron impact using the energy of 70 eV. The temperature of
193 the ionization source was set at 200 $^{\circ}$ C. Samples were injected in splitless mode at 310 $^{\circ}$ C. The
194 oven temperature was programmed from an initial temperature of 200 $^{\circ}$ C (held for 1 min) then
195 rising to 310 $^{\circ}$ C at 15 $^{\circ}$ C/min (held for 35 min). Helium was used as the carrier gas, with a flow
196 rate of 1.0 mL/min. The analyses were made in selective ion monitoring mode (SIM). The
197 compound identification was performed based on the comparison of the retention times and the
198 mass spectra with the available standards or the literature data (Table S1) (Debieu et al., 1992;
199 Derrien et al., 2019b; Harrault et al., 2019). The quantification was achieved with a 5-point
200 internal calibration curve (two series of calibration solutions: 0.1, 0.3, 0.5, 0.7 and 1ppm or 1, 3,
201 5, 7, and 10 ppm) of sterol and stanol standards with a constant IS concentration at 0.5 or 5 ppm,
202 respectively.

203

204 **2.6. Conservative mixing relationship and statistical analyses**

205 In order to assess the applicability of the different source tracking tools during early
206 diagenetic processes, several evaluation criteria were established, which were already presented
207 and used in a previous study (Derrien et al., 2019b). First, the linearity of the measured
208 parameters in artificial sediments was examined with respect to the increasing abundance of one
209 end-member (e.g., soil or algae). The relationship was considered “linear” for a high coefficient
210 of determination $R^2 > 0.8$ and an associated p-value < 0.01 . Then, the regression slope of the
211 measured values (S_m) normalized by the standard deviation of the measurement (S_m/SD) was
212 used to evaluate the sensitivity of the source discrimination. A value of the $S_m/SD \geq 0.5$ was
213 considered as an acceptable range for the sensitivity regarding the source discrimination relative
214 to the measurement uncertainty. Finally, the deviation from the ideal values for a conservative
215 mixing relationship was estimated in calculating the percent difference (%difference) between
216 the slopes for the measured (S_m) and the predicted (S_p) values. The predicted values were
217 calculated using the pure end member's values and the mixing ratios under a linear conservative
218 mixing assumption (Table S2). A %difference of $< 10\%$ was considered to be an acceptable
219 degree of the deviation for the conservative mixing relationship.

220

221 **3.RESULTS AND DISCUSSION**

222 **3.1. Spectroscopic indices**

223 3.1.1. Identification of different fluorescent components via EEM-PARAFAC

224 Four different components were identified, which included two humic-like components
225 (C1 and C2) and two protein-like components (C3 and C4) (Fig. S1). All the identified
226 components were consistent with those previously reported and/or matched well with the Open
227 Fluor database with similarity scores > 0.95 (Table S3). Component 1 (C1), which had a

228 maximum at 220/430 nm (Ex/Em), can be assigned to a typical terrestrial humic-like component
229 (Graeber et al., 2012; Retelletti Brogi et al., 2019b; Yamashita et al., 2010). Component 2 (C2),
230 which exhibited the maxima at 220/547 nm (Ex/Em) can be associated with a humic-like
231 component or aromatic conjugated macromolecular substances of terrestrial origin (Galletti et
232 al., 2019; Osburn et al., 2016b; Wünsch et al., 2017). Component 3 (C3, peaked at 220,265/361
233 nm (Ex/Em)) and component 4 (C4, peaked at 220/309 nm (Ex/Em)) were reported as protein-
234 like substances, namely tryptophan-like and tyrosine-like components, respectively (Cawley et
235 al., 2012; D'Andrilli et al., 2017; Derrien et al., 2019c; Yamashita et al., 2013).

236

237 3.1.2. Changes in the absorbance and fluorescence proxies of the end-members after incubation

238 The original (at 0-day of incubation) spectroscopic characteristics of the end-members for
239 this study were consistent with those reported in our previous study (Derrien et al., 2019b)
240 (Table 1). Briefly, the soil end-member was mainly characterized by a high abundance of both
241 humic-like components, C1 and C2, with the percentages of > 47% and > 34%, respectively. On
242 the contrary, the algae end-member consisted only of the two protein-like components, C3 and
243 C4, in equal proportions. Higher values of $SUVA_{254}$ ($> 2 \text{ L}\cdot\text{mg}^{-1}\cdot\text{m}^{-1}$) and HIX (> 15) were
244 observed for the soil end-member corroborating the high content of humic substances (Derrien
245 et al., 2017; Zsolnay et al., 1999) while higher values of YFI (>3.5) and BIX (>1) were for the
246 algae end-member reflecting its allochthonous origin (Heo et al., 2016; Huguet et al., 2009).

247 The biodegradation led to changes in most of the spectroscopic parameters for both end-
248 members (Table 1). Although there were some differences between the two oxygen conditions,
249 these differences were not statistically significant as the p-values were > 0.95 (e.g., 0.95 and 0.99
250 for soil and algae end-members, respectively). After 60 days of incubation, the soil end member

251 exhibited a partial (~30%) to complete decrease in the abundance for both protein-like
252 components (C3 and C4). For the main fluorescent components, biodegradation induced a slight
253 depletion of the component C1 (< 10%) while the second humic-like component (C2) became
254 more enriched by 20 to 30 %. Regarding the spectroscopic indices, substantial changes were
255 mainly observed for HIX with an increase of the value from 16.9 ± 0.7 to 21.3 ± 0.2 under oxic
256 conditions. However, the increase was slight under anoxic conditions. These results suggest the
257 initial consumption of labile substances as reflected by the partial to complete consumption of
258 the two protein-like components, and a production of high molecular weight and aromatic
259 substances (C2) through the alteration of existing compounds (C1) and/or the production of new
260 compounds by autotrophic organisms (Derrien et al., 2019a; Hansen et al., 2016; Kinsey et al.,
261 2018).

262 The algae end member also displayed biodegradation-induced changes in the spectroscopic
263 parameters. The most notable change was the decrease in the relative abundance of the C3
264 component (i.e., %C3) changing from 46.0 ± 0.8 to $42.4 \pm 1.1\%$ in parallel to the increase of the
265 %C4 values from 52.5 ± 0.6 to $56.5 \pm 1.1\%$. The two fluorescence indices, FI and YFI, showed
266 an increasing trend over incubation, with highest increasing trend for FI under oxic conditions
267 (0.7 ± 0.0 to 1.0 ± 0.0) and YFI under anoxic conditions (3.6 ± 0.0 to 4.1 ± 0.0). The rest of the
268 indices exhibited little to slight changes in the values specifically under anoxic conditions. For
269 instance, BIX presented an increase of ~20% after incubation under anoxic conditions compared
270 to 3% under oxic conditions. Although the results could be apparently interpreted as the
271 occurrence of a potential effect of the oxygen condition, the t-test demonstrated that these
272 changes were not significant enough (p-values >0.95). These observations suggest that the

273 biodegradation-induced changes in the spectroscopic source indices were less pronounced for the
274 algae end-member than for the soil counterpart.

275

276 3.1.3. Biodegradation-induced deviations from a relationships

277 Biodegradation-induced changes in the absorbance and fluorescence proxies with the
278 increasing of the algal content in the artificial sediments are shown in Figs. 1 and 2. The oxygen
279 condition does not seem to be a critical factor for the biodegradation-induced changes as the
280 results were similar between the oxic and anoxic conditions (p-value of >0.96). The trends of the
281 proxies at 0 day and 60 days of incubation (i.e., %C1, %C2, YFI, HIX, and BIX; Fig. 1 and Fig.
282 2c, d, e) with increasing algal content in the mixture mostly remained the same (p-values > 0.6
283 for each proxy), which demonstrates a minor effect of the degradation on the changing trends of
284 the signals with source mixing. By contrast, deviations between both incubation times were
285 observed for the proxies including %C3, %C4, $SUVA_{254}$, and FI. $SUVA_{254}$ and FI (Fig 1 and Fig
286 2a, b) displayed a deviation after biodegradation along with the increase of the algae fraction
287 except for the mixing ratio soil/algae 25/75. However, for %C3 and %C4, the deviation is the
288 most pronounced at the particular mixing ratio (i.e., 25/75).

289 In order to evaluate the applicability of spectroscopic indices to identify OM sources
290 during early diagenesis, the initial relationships between the absorbance/fluorescence proxies and
291 the abundance of algal content in the mixture were investigated (Table 2) and then compared to
292 the relationship after the biodegradation (Table 3). At 0-day of incubation (Table 2), only %C1,
293 %C2, %C4, and $SUVA_{254}$ satisfied the threshold values of the three evaluation criteria (i.e., R^2
294 with p-value < 0.01 , $Sm/SD > 0.5$, and % difference $> 10\%$) for one (e.g. %C2 and $SUVA_{254}$) or
295 both of the oxygen conditions (e.g. %C1 and %C4). The parameters of YFI and HIX can also be

296 suggested as good indices in terms of the relationship linearity as they meet 2 out of the 3 criteria
297 without any effect of the oxygen condition, while %C3 and FI seem to be reliable indices for
298 anoxic and oxic conditions, respectively. BIX does not validate any of the criteria. Among these
299 spectroscopic indices, after incubation, only %C1, YFI and HIX still satisfied all the evaluation
300 criteria for both oxygen conditions after biodegradation except HIX under anoxic condition
301 which did not validate the sensitivity criterion ($S_m/SD = 0.169$) (Table 3). The other indices
302 failed to meet the criteria in terms of linearity, sensitivity and exhibited a higher degree of the
303 deviation from the ideal end-member mixing ratios (i.e., %C3, %C4, $SUVA_{254}$, FI) after
304 microbial degradation. The non-conservative behavior of the latter indices is probably related to
305 the labile characteristic of these compounds (protein-like). From the comparison of all
306 spectroscopic proxies using the evaluation criteria at 0-day and 60-days incubation, it can be
307 concluded that the relative abundance of C1 and the two fluorescence indices, YFI and HIX,
308 represent the optimum fluorescence proxies for the OM source discrimination with varying end-
309 member mixing ratios.

310

311 **3.2. Carbon isotope ratios**

312 3.2.1. Changes for carbon isotope ratios of the end-members after incubation

313 As has been found for the spectroscopic characteristics, the isotope results of the end-
314 members at 0-day of incubation were similar to those reported in our previous study (Derrien et
315 al., 2019b) and in lines with other literature (Pardue et al., 1976; Yu et al., 2015). The values for
316 the soil end-member were $-27.2 \pm 0.1\text{‰}$ (oxic) and $-26.7 \pm 0.1\text{‰}$ (anoxic), while those of the
317 algae end-member were $-11.9 \pm 0.0\text{‰}$ (oxic) and $-11.4 \pm 0.1\text{‰}$ (anoxic).

318 After incubation, the values of each end-member remained invariant suggesting no effect
319 of the biodegradation on the carbon isotopic composition. In addition, no significant difference
320 was observed between the two oxygen conditions as the p-values were >0.3 and >0.9 for soil and
321 algae end-members, respectively. These results confirm the chemical stability of the $\delta^{13}\text{C}$ to trace
322 the sedimentary OM under the early diagenetic processes irrespective of oxygen presence.

323

324 3.2.2. Biodegradation-induced deviations from end member mixing analysis (EMMA)
325 relationships

326 Biodegradation-induced changes in the $\delta^{13}\text{C}$ values of the artificial sediment mixtures
327 with the increase of the algal content are shown in Fig. 3. The variation of the $\delta^{13}\text{C}$ values
328 exhibited a linear relationship with the increase of the algal end member fraction both before and
329 after biodegradation (Tables 2 and 3). In addition, the trend of the carbon stable isotope with
330 increasing algal content in the mixture mostly remained the same before and after 60-days of
331 incubation, demonstrating the resistance to isotopic alteration of $\delta^{13}\text{C}$ during sedimentary
332 diagenesis. However, a deviation was exceptionally noted for a mixing ratio with an equal
333 proportion of both end-members (soil/algae, 50/50). The decomposition of OM, mediated by a
334 variety of microbial processes, can progressively modify the bulk composition of the organic
335 substrates because different OM fractions do not have the same degradation rate (Lehmann et al.,
336 2002; Meyers, 1994). Such selective losses of certain OM fractions may lead to a diagenetic shift
337 in $\delta^{13}\text{C}$ (Benner et al., 1997; Harvey et al., 1995), leaving irreversible changes in the original
338 signature. This is probably what was observed in this present study for the 50/50 artificial
339 sediment mixture. Although the stable isotopes are widely recognized as a reliable and robust
340 tool to trace sediment OM sources and their linear and conservative behavior in a context of

341 artificial end-member mixing (Derrien et al., 2019b), their specificity to individual sources might
342 be weakened by early diagenesis of sediments (Bianchi and Canuel, 2011; Xiao and Liu, 2010).

343 Despite this slight deviation, the $\delta^{13}\text{C}$ tool satisfied the three (and 2 for anoxic condition)
344 established criteria (e.g., linearity, sensitivity, and deviation), evidencing its linear conservative
345 mixing behavior after biodegradation, with the R^2 values ranging from 0.969 (oxic) and 0.898
346 (anoxic) with the p-values of ≤ 0.01 , %difference of $<10\%$ and a sensitivity S_m/SD of 1.431
347 (oxic condition) (Table 3). These results clearly demonstrate the robustness and reliability of the
348 $\delta^{13}\text{C}$ tool for source discrimination of the sedimentary OM even after diagenetic processes.

349

350 **3.3. Sterols/stanols biomarkers**

351 3.3.1. Changes in the sterol/stanols distributions of the end-members after incubation

352 The original (at 0-day of incubation) biomarker distribution of the end-members for this
353 study was consistent with those reported in our previous study (Derrien et al., 2019b). Briefly,
354 the soil end member was characterized by abundant amounts ($> 80\%$) of terrestrial sterols, such
355 as sitosterol, stigmasterol, and campesterol (Table 1), which agreed with the previous literature
356 (Bouloubassi et al., 1997; Derrien et al., 2011; Volkman, 2005). Fecosterol, a fungi or lichens
357 sterol biomarker (Debieu et al., 1992), sitostanol (i.e., a by-product of sitosterol) and cholesterol,
358 the most ubiquitous sterol (Leeming and Nichols, 1998), contributed $<10\%$ of the total
359 sterol/stanol distribution. Unlike the soil end-member, the algae was characterized by a high
360 abundance ($>70\%$) of ergosterol (i.e., a green algae sterol biomarker) and the occurrence of
361 sitosterol and cholesterol, which corresponded to $\sim 18\%$ and $\sim 5\%$ of the sterol compounds in the
362 distribution, respectively.

363 After biodegradation, the distribution in sterols/stanols in the end-members remained the
364 same, suggesting no effect of biodegradation such as it was observed for the carbon isotopic
365 composition. Likewise, no significant difference was observed between the two oxygen
366 conditions with the p-values of > 0.90 for both end-members. These results confirm the chemical
367 stability of the biomarkers to trace the sedimentary OM sources under 60 days of diagenesis
368 dominated by biodegradation.

369

370 3.3.2. Biodegradation-induced deviations from EMMA relationships

371 Biodegradation-induced changes in the distribution of sterol/stanol biomarkers in the
372 artificial sediments with increasing algal content are shown in Fig. 4. In most cases, the oxygen
373 condition does not seem to be a critical factor for the biodegradation-induced changes, which
374 was observed for all the biomarkers (p-value > 0.99). For all the biomarkers, the observed
375 distribution values at all varying algal content between 0-day and 60-days of incubation were the
376 same based on paired t-test (p-value > 0.5). These observations demonstrate the chemical
377 stability of these lipid biomarkers to biodegradation-induced alterations.

378 Regarding the applicability of the lipid biomarker to identify the source of the
379 sedimentary OM during early diagenesis, none of them satisfy the established evaluation criteria
380 either at 0-day or 60-days of incubation (Table 2 and 3). Surprisingly, the source-specific
381 biomarkers, found only in one of the two end-members (Table 1) such as ergosterol, fecosterol,
382 campesterol, stigmasterol or sitostanol, even did not follow a conservative behavior with sources
383 mixing before biodegradation as the highest R^2 were still < 0.8 with p-values > 0.05 . For
384 example, campesterol, presented a R^2 of 0.695 with a p-values of 0.079 and 0.722 (p-
385 value=0.068) in oxic and anoxic conditions, respectively (Table 2). It is noteworthy that

386 biodegradation-induces changes on the sitostanol resulted in improved linearity criterion with the
387 R^2 values changing from 0.530 (0.163) to 0.638 (0.105) and 0.544 (0.155) to 0.723 (0.068) for 0-
388 day and 60-days of incubation, respectively and specifically the reduction in the deviation under
389 anoxic condition (i.e., 11.8% to 6.2%). This singular behavior can be related to the nature of the
390 by-products of the biomarker showing a higher sensitivity to microbial alterations (Bull et al.,
391 2002). On the other hand, the incapability of the sterols and stanols lipid biomarker to reflect a
392 conservative behavior with sources mixing was already suggested by previous studies (Derrien et
393 al., 2019b; Shah et al., 2007). Nevertheless, it is noteworthy to observe the conservative behavior
394 of these lipid biomarkers under early diagenetic processes.

395

396 **3.4. Implications for the applicability of the organic matter source tracking tools**

397 In this study, the discrimination capacities of the commonly used source tracking tools
398 during biodegradation were evaluated and compared using artificial sediments with different
399 mixing ratios of two contrasting OM sources. Two modes were observed: (i) conservative
400 behavior in terms of both sources mixing and biodegradation and (ii) single conservative
401 behavior in terms of biodegradation only. The results of this study demonstrated the strong
402 capacity and the robustness of the carbon stable isotope ratios to identify and estimate the source
403 contribution in artificial sediment mixtures under diagenesis processes. Several spectroscopic
404 indices, such as the relative distribution of the specific fluorescent components (i.e., C1: typical
405 terrestrial humic-like component), YFI, and HIX also represent a robust source tracking tools
406 that were applicable for source apportionment to the artificial sediment mixtures samples under
407 the influence of biodegradation. Regrettably, the lipid biomarkers failed in source apportionment
408 in the artificial sediment mixtures. However, they demonstrated a resistance to microbial

409 alterations in time. Regarding the oxygen conditions, this study provides, for the first time, clear
410 evidence of little to slight effect on the efficacy of the 3 tested sources tracking tools in artificial
411 sediment mixtures.

412 All of these tools were widely utilized to track and estimate OM sources in various
413 aquatic environments, including rivers, groundwater, lakes, coastal area and oceans (Affouri and
414 Sahraoui, 2017; Derrien et al., 2018; Li et al., 2015; Ogrinc et al., 2005; Osburn et al., 2016a;
415 Toming et al., 2013; Zhang et al., 2017). They were used with the assumption of a conservative
416 mixing behavior and stability under diagenetic processes without firstly evaluating the potential
417 limited applicability. But this study provides strong evidence that the application of these
418 commonly-used tools for OM source tracking could lead to a disruption of the source assignment
419 to a wrong estimation of OM source contribution in the context of early diagenesis. Aside from
420 demonstrating a certain weakness to a complete failure of some of the commonly-used source
421 tracking tools to identify and estimate OM source contribution, this study also raises the question
422 of temporal scale. 60 days of incubation correspond to a short period of time at the diagenesis
423 scale. Therefore, it is reasonable to presume that in higher temporal scale, the limitations of the
424 tools may increase to reach the point where the source tracking tools will not be sustainable
425 anymore, leading to misinterpretations of the biogeochemical processes and dynamics of the OM
426 in specific environments.

427 Experiments carried out under controlled conditions can help in identifying the effects
428 that certain factors have on OM dynamics and properties by excluding any interferences that
429 might exist in real environments (Derrien et al., 2019a). In the present case, we tried to elucidate
430 the combined effect of multiple factors (i.e., microbial degradation, mixing sources ratios and
431 oxygen condition). Although the results of this study provide significant insights for the

432 characterization of the OM and more generally for the biogeosciences field, it is necessary to be
433 aware of some limitations of this kind of approach (i.e., a limited number of OM sources (i.e.,
434 soil and algae) and choice of the degradation time). Nevertheless, in this current framework,
435 carbon stable isotope ratios, the relative distribution of refractory material fluorescent
436 component, YFI and HIX indices could be suggested as a reasonable tracer with the most
437 conservative behavior and the highest sensitivity to source discrimination in the period of early
438 diagenesis.

439

440 **4. CONCLUSION**

441 The applicability of three commonly-used source tracking tools (i.e., fluorescence
442 proxies, carbon stable isotope ratios, and lipid biomarkers) was tested on artificial sediments of
443 different mixing ratios of two contrasting OM sources and after biodegradation for both oxygen
444 conditions (i.e., oxic and anoxic). Based on three evaluation criteria concerning the linearity of
445 the relationships, the discrimination sensitivity, and the conservative mixing behavior, the
446 following conclusions can be drawn:

- 447 • The oxygen condition seems to make a limited contribution to biodegradation-induced
448 changes by comparing initial and final tested properties after 60 days of degradation.
- 449 • Most of the sources tracking proxies evaluated in this study show a conservative behavior
450 with respect to biodegradation. By contrast, only a few demonstrated a linear relationship
451 with the increasing portion of one end-members in the mixtures.
- 452 • The fluorescence proxies such as the relative distribution of a humic-like component
453 associated with refractory source material (C1, Ex/Em: 220/430nm), YFI, and HIX
454 indices were found to be reliable proxies for source tracking after biodegradation.

- 455 • Carbon stable isotope ratios exhibited a linear conservative relationship with varying
456 mixing ratios of end members before and after biodegradation, demonstrating its strong
457 capacity for identifying the source's contribution at any time under diagenesis.
- 458 • The biomarkers of sterols/stanols did show a conservative nature under biodegradation
459 however they did not present a good linear and conservative behavior with varying
460 mixing ratios of end members.

461 Although these results clearly demonstrate the sensitivity to early diagenesis processes to
462 most of the commonly used sources tracking tools, further studies need to be performed to
463 rigorously evaluate the effects of the incubation time, temperature, sediment depth to fully
464 validate these results.

465 **Acknowledgments**

466 This work was supported by the National Research Foundation of Korea (NRF) grants and was
467 funded by the Korean government (MSIP) (No. 2017R1A4A1015393 and
468 2017R1D1A1B033546).

470 **References**

- 471 Affouri, H., Sahraoui, O., 2017. The sedimentary organic matter from a Lake Ichkeul core (far
472 northern Tunisia): Rock-Eval and biomarker approach. *J. African Earth Sci.* 129, 248–259.
473 <https://doi.org/10.1016/j.jafrearsci.2017.01.017>
- 474 Aiken, G., 2014. Fluorescence and Dissolved Organic Matter, in: Baker, A., Reynolds, D.M.,
475 Lead, J., Coble, P.G., Spencer, R.G.M. (Eds.), *Aquatic Organic Matter Fluorescence*.
476 Cambridge University Press, Cambridge, pp. 35–74. <https://doi.org/DOI:>

477 10.1017/CBO9781139045452.005

478 Amiotte-Suchet, P., Linglois, N., Leveque, J., Andreux, F., 2007. ¹³C composition of dissolved
479 organic carbon in upland forested catchments of the Morvan Mountains (France): Influence
480 of coniferous and deciduous vegetation. *J. Hydrol.* 335, 354–363.
481 <https://doi.org/http://dx.doi.org/10.1016/j.jhydrol.2006.12.002>

482 Arndt, S., Jørgensen, B.B., LaRowe, D.E., Middelburg, J.J., Pancost, R.D., Regnier, P., 2013.
483 Quantifying the degradation of organic matter in marine sediments: A review and synthesis.
484 *Earth-Science Rev.* 123, 53–86. <https://doi.org/10.1016/J.EARSCIREV.2013.02.008>

485 Benner, R., Biddanda, B., Black, B., McCarthy, M., 1997. Abundance, size distribution, and
486 stable carbon and nitrogen isotopic compositions of marine organic matter isolated by
487 tangential-flow ultrafiltration. *Mar. Chem.* 57, 243–263.
488 [https://doi.org/http://dx.doi.org/10.1016/S0304-4203\(97\)00013-3](https://doi.org/http://dx.doi.org/10.1016/S0304-4203(97)00013-3)

489 Bianchi, T.S., Canuel, E.A., 2011. *Chemical Biomarkers in Aquatic Ecosystems*. Princeton
490 University Press, Princeton University.

491 Bischoff, J., Sparkes, R.B., Doğrul Selver, A., Spencer, R.G.M., Gustafsson, Ö., Semiletov, I.P.,
492 Dudarev, O. V., Wagner, D., Rivkina, E., van Dongen, B.E., Talbot, H.M., 2016. Source,
493 transport and fate of soil organic matter inferred from microbial biomarker lipids on the
494 East Siberian Arctic Shelf. *Biogeosciences* 13, 4899–4914. [https://doi.org/10.5194/bg-13-](https://doi.org/10.5194/bg-13-4899-2016)
495 [4899-2016](https://doi.org/10.5194/bg-13-4899-2016)

496 Bouloubassi, I., Lipiatou, E., Saliot, A., Tolosa, I., Bayona, J.M.M., Albaigés, J., 1997. Carbon
497 sources and cycle in the western Mediterranean—the use of molecular markers to determine
498 the origin of organic matter. *Deep Sea Res. Part II Top. Stud. Oceanogr.* 44, 781–799.
499 [https://doi.org/https://doi.org/10.1016/S0967-0645\(96\)00094-X](https://doi.org/https://doi.org/10.1016/S0967-0645(96)00094-X)

500 Briand, M.J., Bonnet, X., Goiran, C., Guillou, G., Letourneur, Y., 2015. Major Sources of
501 Organic Matter in a Complex Coral Reef Lagoon: Identification from Isotopic Signatures
502 ($\delta(13)C$ and $\delta(15)N$). PLoS One 10, e0131555.
503 <https://doi.org/10.1371/journal.pone.0131555>

504 Bull, I.D., Lockheart, M.J., Elhmmali, M.M., Roberts, D.J., Evershed, R.P., 2002. The origin of
505 faeces by means of biomarker detection. Environ. Int. 27, 647–654.

506 Carabel, S., Godínez-Domínguez, E., Verísimo, P., Fernández, L., Freire, J., 2006. An
507 assessment of sample processing methods for stable isotope analyses of marine food webs.
508 J. Exp. Mar. Bio. Ecol. 336, 254–261.
509 <https://doi.org/http://dx.doi.org/10.1016/j.jembe.2006.06.001>

510 Cawley, K.M., Ding, Y., Fourqurean, J., Jaffé, R., 2012. Characterising the sources and fate of
511 dissolved organic matter in Shark Bay, Australia: a preliminary study using optical
512 properties and stable carbon isotopes. Mar. Freshw. Res. 63, 1098–1107.
513 <https://doi.org/https://doi.org/10.1071/MF12028>

514 D’Andrilli, J., Foreman, C.M., Sigl, M., Priscu, J.C., McConnell, J.R., 2017. A 21 000-year
515 record of fluorescent organic matter markers in the WAIS Divide ice core. Clim. Past 13,
516 533–544.

517 Debieu, D., Gall, C., Gredt, M., Bach, J., Malosse, C., Leroux, P., 1992. Ergosterol biosynthesis
518 and its inhibition by fenpropimorph in Fusarium species. Phytochemistry 31, 1223–1233.
519 [https://doi.org/https://doi.org/10.1016/0031-9422\(92\)80265-G](https://doi.org/https://doi.org/10.1016/0031-9422(92)80265-G)

520 Derrien, M., Brogi, S.R., Gonçalves-Araujo, R., Retelletti Brogi, S., Gonçalves-Araujo, R.,
521 2019a. Characterization of aquatic organic matter: Assessment, perspectives and research
522 priorities. Water Res. 163, 114908. <https://doi.org/10.1016/J.WATRES.2019.114908>

523 Derrien, M., Cabrera, F.A., Tavera, N.L.V., Kantún Manzano, C.A., Vizcaino, S.C., 2015.
524 Sources and distribution of organic matter along the Ring of Cenotes, Yucatan, Mexico:
525 Sterol markers and statistical approaches. *Sci. Total Environ.* 511.
526 <https://doi.org/10.1016/j.scitotenv.2014.12.053>

527 Derrien, M., Jardé, E., Gruau, G., Pierson-Wickmann, A.-C., 2011. Extreme variability of steroid
528 profiles in cow feces and pig slurries at the regional scale: Implications for the use of
529 steroids to specify fecal pollution sources in waters. *J. Agric. Food Chem.* 59.
530 <https://doi.org/10.1021/jf201040v>

531 Derrien, M., Kim, M.S., Ock, G., Hong, S., Cho, J., Shin, K.H.K.-H.K.H., Hur, J., 2018.
532 Estimation of different source contributions to sediment organic matter in an agricultural-
533 forested watershed using end member mixing analyses based on stable isotope ratios and
534 fluorescence spectroscopy. *Sci. Total Environ.* 618, 569–578.
535 <https://doi.org/https://doi.org/10.1016/j.scitotenv.2017.11.067>

536 Derrien, M., Shin, K.-H., Hur, J., 2019b. Assessment on applicability of common source tracking
537 tools for particulate organic matter in controlled end member mixing experiments. *Sci.*
538 *Total Environ.* 666, 187–196. <https://doi.org/10.1016/j.scitotenv.2019.02.258>

539 Derrien, M., Shin, K.-H., Hur, J., 2019c. Biodegradation-induced signatures in sediment pore
540 water dissolved organic matter: Implications from artificial sediments composed of two
541 contrasting sources. *Sci. Total Environ.* 694, 133714.

542 Derrien, M., Yang, L., Hur, J., 2017. Lipid biomarkers and spectroscopic indices for identifying
543 organic matter sources in aquatic environments: A review. *Water Res.* 112.
544 <https://doi.org/10.1016/j.watres.2017.01.023>

545 Fichot, C.G., Kaiser, K., Hooker, S.B., Amon, R.M.W., Babin, M., Bélanger, S., Walker, S.A.,

546 Benner, R., 2013. Pan-Arctic distributions of continental runoff in the Arctic Ocean 3, 1053.
547 <https://doi.org/10.1038/srep01053><http://dharmasastra.live.cf.private.springer.com/articles/sr>
548 [ep01053#supplementary-information](https://doi.org/10.1038/srep01053#supplementary-information)

549 Fisher, M.M., Reddy, K.R., James, R.T., 2005. Internal nutrient loads from sediments in a
550 shallow, subtropical lake. *Lake Reserv. Manag.* 21, 338–349.
551 <https://doi.org/10.1080/07438140509354439>

552 Galletti, Y., Gonnelli, M., Retelletti Brogi, S., Vestri, S., Santinelli, C., 2019. DOM dynamics in
553 open waters of the Mediterranean Sea: New insights from optical properties. *Deep. Res.*
554 *Part I Oceanogr. Res. Pap.* 144, 95–114. <https://doi.org/10.1016/j.dsr.2019.01.007>

555 Gordon, E.S., Goni, M.A., 2003. Sources and distribution of terrigenous organic matter delivered
556 by the Atchafalaya River to sediments in the northern Gulf of Mexico. *Geochim.*
557 *Cosmochim. Acta* 67, 2359–2375. [https://doi.org/10.1016/S0016-7037\(02\)01412-6](https://doi.org/10.1016/S0016-7037(02)01412-6)

558 Graeber, D., Gelbrecht, J., Pusch, M.T., Anlanger, C., von Schiller, D., 2012. Agriculture has
559 changed the amount and composition of dissolved organic matter in Central European
560 headwater streams. *Sci. Total Environ.* 438, 435–446.
561 [https://doi.org/https://doi.org/10.1016/j.scitotenv.2012.08.087](https://doi.org/10.1016/j.scitotenv.2012.08.087)

562 Guenet, B., Danger, M., Harrault, L., Allard, B., Jauset-Alcala, M., Bardoux, G., Benest, D.,
563 Abbadie, L., Lacroix, G., 2014. Fast mineralization of land-born C in inland waters: first
564 experimental evidences of aquatic priming effect. *Hydrobiologia* 721, 35–44.
565 <https://doi.org/10.1007/s10750-013-1635-1>

566 Hansen, A.M., Kraus, T.E.C., Pellerin, B.A., Fleck, J.A., Downing, B.D., Bergamaschi, B.A.,
567 2016. Optical properties of dissolved organic matter (DOM): Effects of biological and
568 photolytic degradation. *Limnol. Oceanogr.* 61, 1015–1032.

569 <https://doi.org/10.1002/Ino.10270>

570 Harrault, L., Milek, K., Jardé, E., Jeanneau, L., Derrien, M., Anderson, D.G., 2019. Faecal
571 biomarkers can distinguish specific mammalian species in modern and past environments.
572 PLoS One 14, e0211119. <https://doi.org/10.1371/journal.pone.0211119>

573 Harvey, R.H., Tuttle, J.H., Bell, T.J., 1995. Kinetics of phytoplankton decay during simulated
574 sedimentation: Changes in biochemical composition and microbial activity under oxic and
575 anoxic conditions. *Geochim. Cosmochim. Acta* 59, 3367–3377.
576 [https://doi.org/http://dx.doi.org/10.1016/0016-7037\(95\)00217-N](https://doi.org/http://dx.doi.org/10.1016/0016-7037(95)00217-N)

577 Henrichs, S.M., 1992. Early diagenesis of organic matter in marine sediments: progress and
578 perplexity. *Mar. Chem.* 39, 119–149. [https://doi.org/10.1016/0304-4203\(92\)90098-U](https://doi.org/10.1016/0304-4203(92)90098-U)

579 Heo, Y., Kim, D.-H., Lee, H., Lee, D., Her, Namguk, J., Yoon, 2016. A new fluorescence index
580 with a fluorescence excitation-emission matrix for dissolved organic matter (DOM)
581 characterization. *Desalin. Water Treat.* 57, 20270–20282.
582 <https://doi.org/10.1080/19443994.2015.1110719>

583 Huguet, A., Vacher, L., Relexans, S., Saubusse, S., Froidefond, J.M., Parlanti, E., 2009.
584 Properties of fluorescent dissolved organic matter in the Gironde Estuary. *Org. Geochem.*
585 40, 706–719. <https://doi.org/https://doi.org/10.1016/j.orggeochem.2009.03.002>

586 Jaffé, R., Boyer, J.N., Lu, X., Maie, N., Yang, C., Scully, N.M., Mock, S., 2004. Source
587 characterization of dissolved organic matter in a subtropical mangrove-dominated estuary
588 by fluorescence analysis. *Mar. Chem.* 84, 195–210.
589 <https://doi.org/https://doi.org/10.1016/j.marchem.2003.08.001>

590 Kinsey, J.D., Corradino, G., Ziervogel, K., Schnetzer, A., Osburn, C.L., 2018. Formation of
591 Chromophoric Dissolved Organic Matter by Bacterial Degradation of Phytoplankton-

592 Derived Aggregates. *Front. Mar. Sci.* 4, 430. <https://doi.org/10.3389/fmars.2017.00430>

593 Kristensen, E., 2000. Organic matter diagenesis at the oxic/anoxic interface in coastal marine
594 sediments, with emphasis on the role of burrowing animals. *Hydrobiologia* 426, 1–24.
595 <https://doi.org/10.1023/A:1003980226194>

596 Kuznetsova, O. V, Sevastyanov, V.S., Timerbaev, A.R., 2019. What are the current analytical
597 approaches for sediment analysis related to the study of diagenesis? *Highlights from 2010*
598 *to 2018. Talanta* 191, 435–442. <https://doi.org/https://doi.org/10.1016/j.talanta.2018.08.080>

599 Leeming, R., Nichols, P.D., 1998. Determination of the sources and distribution of sewage and
600 pulp-fibre-derived pollution in the Derwent Estuary, Tasmania, using sterol biomarkers.
601 *Mar. Freshw. Res.* 49, 7–17. <https://doi.org/https://doi.org/10.1071/MF95140>

602 Lehmann, M.F., Bernasconi, S.M., Barbieri, A., McKenzie, J.A., 2002. Preservation of organic
603 matter and alteration of its carbon and nitrogen isotope composition during simulated and in
604 situ early sedimentary diagenesis. *Geochim. Cosmochim. Acta* 66, 3573–3584.
605 [https://doi.org/http://dx.doi.org/10.1016/S0016-7037\(02\)00968-7](https://doi.org/http://dx.doi.org/10.1016/S0016-7037(02)00968-7)

606 Li, P., Chen, L., Zhang, W., Huang, Q., 2015. Spatiotemporal Distribution, Sources, and
607 Photobleaching Imprint of Dissolved Organic Matter in the Yangtze Estuary and Its
608 Adjacent Sea Using Fluorescence and Parallel Factor Analysis. *PLoS One* 10, e0130852.
609 <https://doi.org/10.1371/journal.pone.0130852>

610 Meyers, P.A., 1994. Preservation of elemental and isotopic source identification of sedimentary
611 organic matter. *Chem. Geol.* 114, 289–302. [https://doi.org/http://dx.doi.org/10.1016/0009-](https://doi.org/http://dx.doi.org/10.1016/0009-2541(94)90059-0)
612 [2541\(94\)90059-0](https://doi.org/http://dx.doi.org/10.1016/0009-2541(94)90059-0)

613 Meyers, P.A., Ishiwatari, R., 1993. Lacustrine organic geochemistry—an overview of indicators
614 of organic matter sources and diagenesis in lake sediments. *Org. Geochem.* 20, 867–900.

615 [https://doi.org/http://dx.doi.org/10.1016/0146-6380\(93\)90100-P](https://doi.org/http://dx.doi.org/10.1016/0146-6380(93)90100-P)

616 Milliken, K.L., 2003. Late Diagenesis and Mass Transfer in Sandstone–Shale Sequences.
617 Treatise on Geochemistry 159–190. <https://doi.org/10.1016/B0-08-043751-6/07091-2>

618 Murphy, K.R., Stedmon, C.A., Graeber, D., Bro, R., 2013. Fluorescence spectroscopy and multi-
619 way techniques. PARAFAC. Anal. Methods 5, 6557–6566.
620 <https://doi.org/10.1039/C3AY41160E>

621 Navel, S., Mermillod-Blondin, F., Montuelle, B., Chauvet, E., Marmonier, P., 2012. Sedimentary
622 context controls the influence of ecosystem engineering by bioturbators on microbial
623 processes in river sediments. Oikos 121, 1134–1144. [https://doi.org/doi:10.1111/j.1600-](https://doi.org/doi:10.1111/j.1600-0706.2011.19742.x)
624 [0706.2011.19742.x](https://doi.org/doi:10.1111/j.1600-0706.2011.19742.x)

625 Ogrinc, N., Fontolan, G., Faganeli, J., Covelli, S., 2005. Carbon and nitrogen isotope
626 compositions of organic matter in coastal marine sediments (the Gulf of Trieste, N Adriatic
627 Sea): indicators of sources and preservation. Mar. Chem. 95, 163–181.
628 <https://doi.org/http://dx.doi.org/10.1016/j.marchem.2004.09.003>

629 Osburn, C.L., Boyd, T.J., Montgomery, M.T., Bianchi, T.S., Coffin, R.B., Paerl, H.W., 2016a.
630 Optical Proxies for Terrestrial Dissolved Organic Matter in Estuaries and Coastal Waters.
631 Front. Mar. Sci. 2. <https://doi.org/10.3389/fmars.2015.00127>

632 Osburn, C.L., Handsel, L.T., Peierls, B.L., Paerl, H.W., 2016b. Predicting Sources of Dissolved
633 Organic Nitrogen to an Estuary from an Agro-Urban Coastal Watershed. Environ. Sci.
634 Technol. 50, 8473–8484. <https://doi.org/10.1021/acs.est.6b00053>

635 Pardue, J.W., Scalán, R.S., Van Baalen, C., Parker, P.L., 1976. Maximum carbon isotope
636 fractionation in photosynthesis by blue-green algae and a green alga. Geochim. Cosmochim.
637 Acta 40, 309–312. [https://doi.org/https://doi.org/10.1016/0016-7037\(76\)90208-8](https://doi.org/https://doi.org/10.1016/0016-7037(76)90208-8)

638 Park, M., Snyder, S.A., 2018. Sample handling and data processing for fluorescent excitation-
639 emission matrix (EEM) of dissolved organic matter (DOM). *Chemosphere* 193, 530–537.
640 <https://doi.org/https://doi.org/10.1016/j.chemosphere.2017.11.069>

641 Pedrosa-Pàmies, R., Parinos, C., Sanchez-Vidal, A., Gogou, A., Calafat, A., Canals, M.,
642 Bouloubassi, I., Lampadariou, N., 2015. Composition and sources of sedimentary organic
643 matter in the deep eastern Mediterranean Sea. *Biogeosciences* 12, 7379–7402.
644 <https://doi.org/10.5194/bg-12-7379-2015>

645 Retelletti Brogi, S., Derrien, M., Hur, J., 2019a. In-Depth Assessment of the Effect of Sodium
646 Azide on the Optical Properties of Dissolved Organic Matter. *J. Fluoresc.* 1–9.
647 <https://doi.org/10.1007/s10895-019-02398-w>

648 Retelletti Brogi, S., Kim, J.-H., Ryu, J.-S., Jin, Y.K., Lee, Y.K., Hur, J., 2019b. Exploring
649 sediment porewater dissolved organic matter (DOM) in a mud volcano: Clues of a
650 thermogenic DOM source from fluorescence spectroscopy. *Mar. Chem.*
651 <https://doi.org/10.1016/J.MARCHEM.2019.03.009>

652 Shah, V.G., Dunstan, R.H., Geary, P.M., Coombes, P., Roberts, T.K., Von Nagy-Felsobuki,
653 E., 2007. Evaluating potential applications of faecal sterols in distinguishing sources of
654 faecal contamination from mixed faecal samples. *Water Res.* 41, 3691–3700.
655 <https://doi.org/10.1016/j.watres.2007.04.006>

656 Sillanpää, M., 2015. *Natural Organic Matter in Water: Characterization and Treatment Methods*,
657 *Natural Organic Matter in Water: Characterization and Treatment Methods*. Elsevier Inc.
658 <https://doi.org/10.1016/C2013-0-19213-6>

659 Southwell, M.W., Kieber, R.J., Mead, R.N., Avery, G.B., Skrabal, S.A., 2010. Effects of sunlight
660 on the production of dissolved organic and inorganic nutrients from resuspended sediments.

661 Biogeochemistry 98, 115–126. <https://doi.org/10.1007/s10533-009-9380-2>

662 Toming, K., Tuvikene, L., Vilbaste, S., Agasild, H., Viik, M., Kisand, A., Feldmann, T., Martma,
663 T., Jones, R.I., Nõges, T., 2013. Contributions of autochthonous and allochthonous sources
664 to dissolved organic matter in a large, shallow, eutrophic lake with a highly calcareous
665 catchment. *Limnol. Oceanogr.* 58, 1259–1270. <https://doi.org/10.4319/lo.2013.58.4.1259>

666 van der Meij, W.M., Temme, A.J.A.M., Lin, H.S., Gerke, H.H., Sommer, M., 2018. On the role
667 of hydrologic processes in soil and landscape evolution modeling: concepts, complications
668 and partial solutions. *Earth-Science Rev.* <https://doi.org/10.1016/j.earscirev.2018.09.001>

669 Volkman, J.K., 2005. Sterols and other triterpenoids: source specificity and evolution of
670 biosynthetic pathways. *Org. Geochem.* 36, 139–159.
671 <https://doi.org/10.1016/J.ORGGEOCHEM.2004.06.013>

672 Volkman, J.K., Tanoue, E., 2002. Chemical and Biological Studies of Particulate Organic Matter
673 in the Ocean. *J. Oceanogr.* 58, 265–279. <https://doi.org/10.1023/a:1015809708632>

674 Vonk, J.E., Tank, S.E., Mann, P.J., Spencer, R.G.M., Treat, C.C., Striegl, R.G., Abbott, B.W.,
675 Wickland, K.P., 2015. Biodegradability of dissolved organic carbon in permafrost soils and
676 aquatic systems: a meta-analysis. *Biogeosciences* 12, 6915–6930.
677 <https://doi.org/10.5194/bg-12-6915-2015>

678 Wakeham, S.G., Canuel, E.A., 1990. Fatty acids and sterols of particulate matter in a brackish
679 and seasonally anoxic coastal salt pond. *Org. Geochem.* 16, 703–713.
680 [https://doi.org/http://dx.doi.org/10.1016/0146-6380\(90\)90111-C](https://doi.org/http://dx.doi.org/10.1016/0146-6380(90)90111-C)

681 Waterson, E.J., Canuel, E.A., 2008. Sources of sedimentary organic matter in the Mississippi
682 River and adjacent Gulf of Mexico as revealed by lipid biomarker and $\delta^{13}\text{C}_{\text{TOC}}$ analyses.
683 *Org. Geochem.* 39, 422–439.

684 <https://doi.org/https://doi.org/10.1016/j.orggeochem.2008.01.011>

685 Wild, B., Andersson, A., Bröder, L., Vonk, J., Hugelius, G., McClelland, J.W., Song, W.,
686 Raymond, P.A., Gustafsson, Ö., 2019. Rivers across the Siberian Arctic unearth the patterns
687 of carbon release from thawing permafrost. *Proc. Natl. Acad. Sci. U. S. A.* 116, 10280–
688 10285. <https://doi.org/10.1073/pnas.1811797116>

689 Wunsch, U.J., Murphy, K.R., Stedmon, C.A., 2017. The One-Sample PARAFAC Approach
690 Reveals Molecular Size Distributions of Fluorescent Components in Dissolved Organic
691 Matter. *Environ. Sci. Technol.* <https://doi.org/10.1021/acs.est.7b03260>

692 Xiao, H.-Y., Liu, C.-Q., 2010. Identifying organic matter provenance in sediments using isotopic
693 ratios in an urban river. *Geochem. J.* 44, 181–187. <https://doi.org/10.2343/geochemj.1.0059>

694 Yamashita, Y., Boyer, J.N., Jaffé, R., 2013. Evaluating the distribution of terrestrial dissolved
695 organic matter in a complex coastal ecosystem using fluorescence spectroscopy. *Cont. Shelf*
696 *Res.* 66, 136–144. <https://doi.org/http://dx.doi.org/10.1016/j.csr.2013.06.010>

697 Yamashita, Y., Maie, N., Briceño, H., Jaffé, R., 2010. Optical characterization of dissolved
698 organic matter in tropical rivers of the Guayana Shield, Venezuela. *J. Geophys. Res.*
699 *Biogeosciences* 115, n/a-n/a. <https://doi.org/10.1029/2009JG000987>

700 Yu, Z.T., Wang, X.J., Zhang, E.L., Zhao, C.Y., Liu, X.Q., 2015. Spatial distribution and sources
701 of organic carbon in the surface sediment of Bosten Lake, China. *Biogeosciences* 12, 6605–
702 6615. <https://doi.org/10.5194/bg-12-6605-2015>

703 Zhang, L., Yin, K., Wang, L., Chen, F., Zhang, D., Yang, Y., 2009. The sources and
704 accumulation rate of sedimentary organic matter in the Pearl River Estuary and adjacent
705 coastal area, Southern China. *Estuar. Coast. Shelf Sci.* 85, 190–196.
706 <https://doi.org/10.1016/j.ecss.2009.07.035>

707 Zhang, Y., Su, Y., Liu, Z., Yu, J., Jin, M., 2017. Lipid biomarker evidence for determining the
708 origin and distribution of organic matter in surface sediments of Lake Taihu, Eastern China.
709 *Ecol. Indic.* 77, 397–408. <https://doi.org/https://doi.org/10.1016/j.ecolind.2017.02.031>

710 Zimmerman, A.R., Canuel, E.A., 2001. Bulk Organic Matter and Lipid Biomarker Composition
711 of Chesapeake Bay Surficial Sediments as Indicators of Environmental Processes. *Estuar.
712 Coast. Shelf Sci.* 53, 319–341. <https://doi.org/http://dx.doi.org/10.1006/ecss.2001.0815>

713 Zsolnay, A., Baigar, E., Jimenez, M., Steinweg, B., Saccomandi, F., 1999. Differentiating with
714 fluorescence spectroscopy the sources of dissolved organic matter in soils subjected to
715 drying. *Chemosphere* 38, 45–50. [https://doi.org/http://dx.doi.org/10.1016/S0045-](https://doi.org/http://dx.doi.org/10.1016/S0045-6535(98)00166-0)
716 [6535\(98\)00166-0](https://doi.org/http://dx.doi.org/10.1016/S0045-6535(98)00166-0)

717

Figure 1. Relative abundances (%) of the four different fluorescent components (C1–C4) in the artificial sediments at five different mixing ratios of two end-members (i.e., soil and algae) under oxic and anoxic conditions at the incubation time of 0 and 60 days. The lines and dashed lines represent the evolution of the parameters with varying mixing ratios of the two end-members.

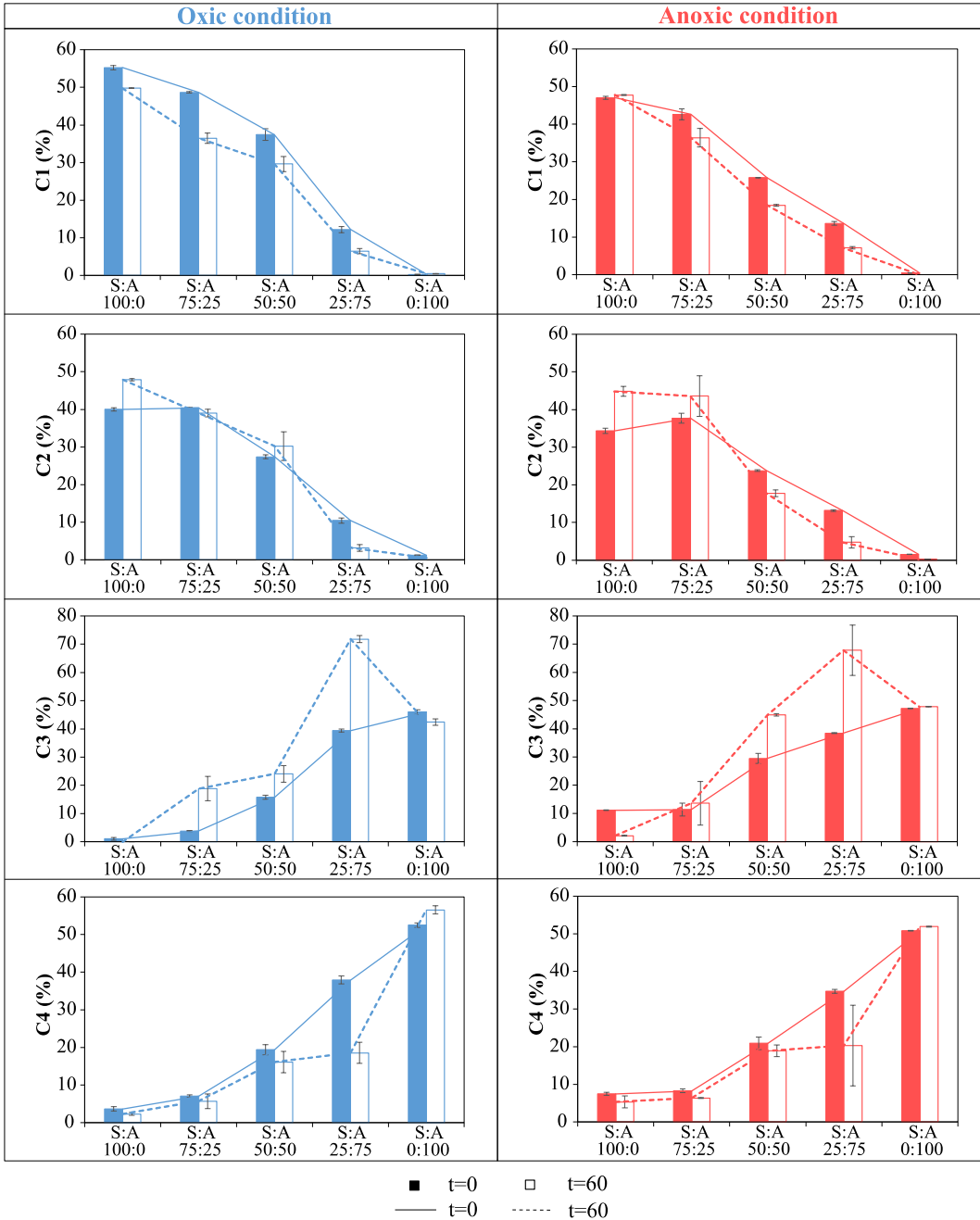


Figure 2. Values of the absorbance and fluorescent indices SUVA (a), FI (b), YFI (c), HIX (d), and BIX (e) in the artificial sediments at five different mixing ratios of two end-members (i.e., soil and algae) under oxic and anoxic conditions at the incubation time of 0 and 60 days. The blue color is used for the data of oxic conditions while pink color for those of anoxic conditions.

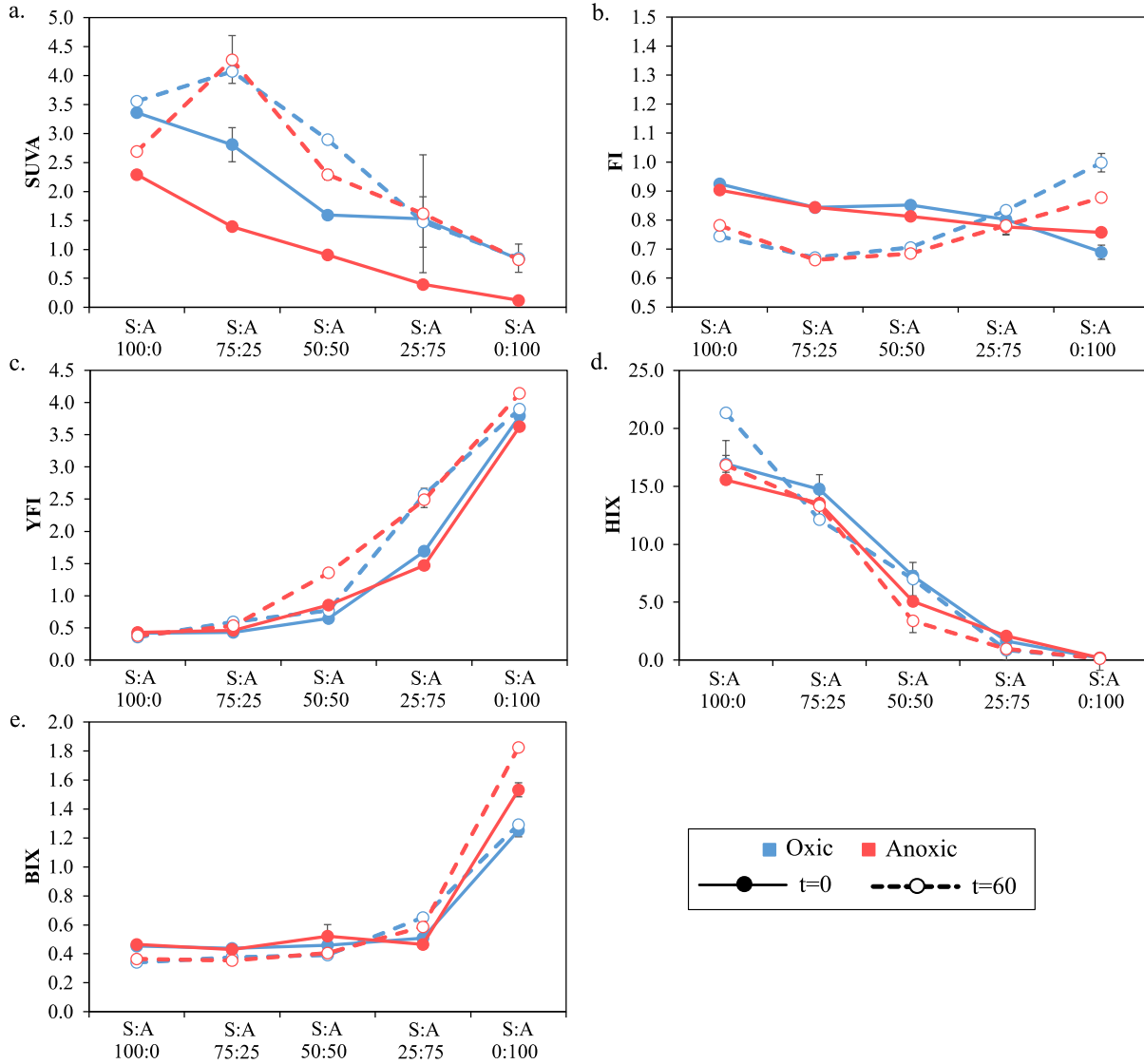


Figure 3. Values of the carbon stable isotopes in the artificial sediments at five different mixing ratios of two end-members (i.e., soil and algae) under oxic and anoxic conditions at the incubation time of 0 and 60 days. The blue color is used for the data of oxic conditions while pink color for those of anoxic conditions.

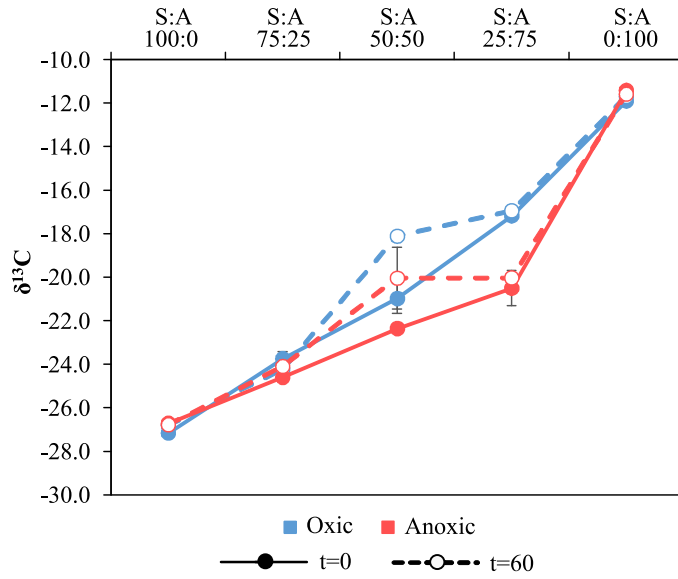
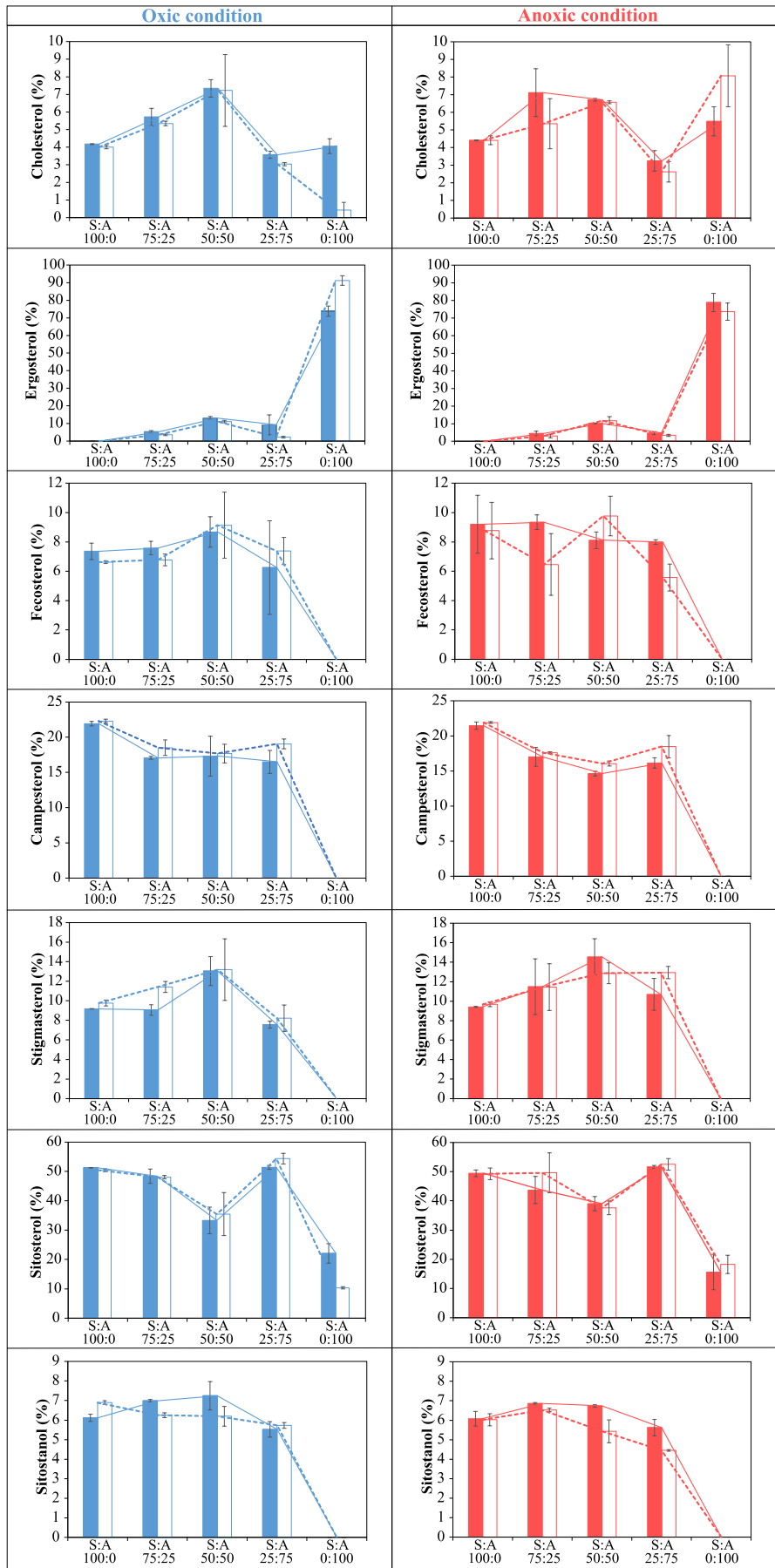


Figure 4. Relative abundances (%) of the seven sterols/stanols in the artificial sediments at five different mixing ratios of two end-members (i.e., soil and algae) under oxic and anoxic conditions at the incubation time of 0 and 60 days. The lines and dashed lines represent the evolution of the parameters with varying mixing ratios of the two end-members.



■ t=0 □ t=60
 — t=0 - - - t=60

Table 1. The relative changes of the 4 identified components (%) and the spectroscopic indices, the carbon stable isotopic ratios (‰) and the distribution in the sterols/stanols in the artificial sediment of the two end-members before versus after incubation under oxic and anoxic conditions (average values \pm standard deviation).

		Soil end-member				Algae end-member			
		Oxic		Anoxic		Oxic		Anoxic	
		0 day- incubation	60 days- incubation	0 day- incubation	60 days- incubation	0 day- incubation	60 days- incubation	0 day- incubation	60 days- incubation
Absorbance and fluorescence proxies	C1 (%)	55.2 \pm 0.6	49.8 \pm 0.1	47.0 \pm 0.4	47.7 \pm 0.1	0.3 \pm 0.1	0.5 \pm 0.0	0.5 \pm 0.0	0.0 \pm 0.0
	C2 (%)	40.0 \pm 0.4	47.9 \pm 0.3	34.4 \pm 0.7	44.8 \pm 1.3	1.2 \pm 0.1	0.6 \pm 0.0	1.5 \pm 0.0	0.2 \pm 0.0
	C3 (%)	1.1 \pm 0.5	0.0 \pm 0.0	11.2 \pm 0.1	2.1 \pm 0.1	46.0 \pm 0.8	42.4 \pm 1.1	47.2 \pm 0.1	47.8 \pm 0.1
	C4 (%)	3.7 \pm 0.5	2.3 \pm 0.3	7.5 \pm 0.4	5.3 \pm 1.6	52.5 \pm 0.6	56.5 \pm 1.1	50.8 \pm 0.1	51.9 \pm 0.1
	SUVA ₂₅₄ (L.mg ⁻¹ .m ⁻¹)	3.4 \pm 0.1	3.6 \pm 0.1	2.3 \pm 0.0	2.7 \pm 0.1	0.8 \pm 0.1	0.8 \pm 0.2	0.1 \pm 0.0	0.8 \pm 0.0
	FI	0.9 \pm 0.0	0.7 \pm 0.0	0.9 \pm 0.0	0.8 \pm 0.0	0.7 \pm 0.0	1.0 \pm 0.0	0.8 \pm 0.0	0.9 \pm 0.0
	YFI	0.4 \pm 0.0	0.4 \pm 0.0	0.4 \pm 0.0	0.4 \pm 0.0	3.8 \pm 0.0	3.9 \pm 0.0	3.6 \pm 0.0	4.1 \pm 0.0
	HIX	16.9 \pm 0.7	21.3 \pm 0.2	15.6 \pm 0.4	16.8 \pm 2.1	0.2 \pm 0.0	0.2 \pm 0.0	0.2 \pm 0.0	0.1 \pm 0.0
	BIX	0.5 \pm 0.0	0.3 \pm 0.0	0.5 \pm 0.0	0.4 \pm 0.0	1.3 \pm 0.0	1.3 \pm 0.0	1.5 \pm 0.0	1.8 \pm 0.0
Isotopes	$\delta^{13}\text{C}$ (‰)	-27.2 \pm 0.1	-26.8 \pm 0.1	-26.7 \pm 0.1	-26.8 \pm 0.1	-11.9 \pm 0.0	-11.7 \pm 0.0	-11.4 \pm 0.1	-11.6 \pm 0.1
Sterols/stanols	Cholesterol (%)	4.2 \pm 0.0	4.0 \pm 0.1	4.4 \pm 0.0	4.4 \pm 0.2	4.1 \pm 0.4	0.4 \pm 0.4	5.5 \pm 0.8	8.1 \pm 1.8
	Ergosterol (%)	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	73.9 \pm 2.9	91.1 \pm 2.7	78.9 \pm 5.2	73.6 \pm 4.9
	Fecosterol (%)	7.4 \pm 0.6	6.6 \pm 0.1	9.2 \pm 2.0	8.8 \pm 1.9	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0
	Campesterol (%)	21.9 \pm 0.3	22.3 \pm 0.3	21.5 \pm 0.5	21.9 \pm 0.1	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0
	Stigmasterol (%)	9.2 \pm 0.0	9.8 \pm 0.3	9.4 \pm 0.0	9.7 \pm 0.2	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0
	Sitosterol (%)	51.3 \pm 0.1	50.4 \pm 0.5	49.1 \pm 1.1	49.2 \pm 2.0	22.1 \pm 3.3	10.3 \pm 0.4	15.6 \pm 6.0	18.3 \pm 3.1
	Sitostanol (%)	6.1 \pm 0.2	6.9 \pm 0.1	6.1 \pm 0.4	6.0 \pm 0.3	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0

Table 2. The evaluation of selected source discrimination indices in the relationships with increasing algal carbon fraction (% Algae) of the artificial sediments at 0-day incubation.

Parameters	Oxygen condition	Tendency	Relationship type ^a	R ² (<i>p</i> -value)	S _m ^b	S _m /SD ^c	S _p ^d	% difference ^e	Applicability ^f
Extractable OM									
C1 (%)	Oxic	↓	Linear	0.958 (<0.01)	0.585	0.905	0.550	6.5	000
	Anoxic	↓	Linear	0.979 (<0.01)	0.488	0.992	0.465	4.9	000
C2 (%)	Oxic	↓	Linear	0.933 (<0.01)	0.430	1.159	0.388	10.9	00X
	Anoxic	↓	Linear	0.904 (<0.01)	0.361	0.752	0.328	9.9	000
C3 (%)	Oxic	↑	Linear	0.935 (<0.01)	0.501	0.917	0.449	11.7	00X
	Anoxic	↑	Linear	0.946 (<0.01)	0.397	0.443	0.360	10.1	OXX
C4 (%)	Oxic	↑	Linear	0.956 (<0.01)	0.514	0.668	0.488	5.3	000
	Anoxic	↑	Linear	0.939 (<0.01)	0.452	0.713	0.433	4.4	000
SUVA ₂₅₄ (L.mg ⁻¹ .m ⁻¹)	Oxic	↓	Linear	0.944 (<0.01)	0.025	0.258	0.025	0.3	OXX
	Anoxic	↓	Linear	0.961 (<0.01)	0.021	1.433	0.022	1.6	000
FI	Oxic	↓	Linear	0.877 (0.019)	0.002	0.290	0.002	13.0	OXX
	Anoxic	↓	Linear	0.964 (<0.01)	0.001	0.137	0.001	1.5	0X0
YFI	Oxic	↑	Nonlinear	0.775 (0.049)	0.032	1.419	0.034	5.0	X00
	Anoxic	↑	Nonlinear	0.774 (0.049)	0.030	3.006	0.032	7.4	X00
HIX	Oxic	↓	Linear	0.956 (<0.01)	0.186	0.551	0.168	11.3	00X
	Anoxic	↓	Linear	0.937 (<0.01)	0.169	0.471	0.154	9.9	0X0
BIX	Oxic	↑	Nonlinear	0.558 (0.147)	0.007	0.361	0.008	16.5	XXX
	Anoxic	↑	Nonlinear	0.519 (0.170)	0.009	0.262	0.011	18.7	XXX
δ ¹³ C (‰)	Oxic	↑	Linear	0.985 (<0.01)	0.148	0.660	0.153	2.8	000
	Anoxic	↑	Linear	0.863 (0.023)	0.139	0.481	0.153	9.3	0X0
Cholesterol (%)	Oxic	↓	Nonlinear	0.059 (0.694)	0.010	0.029	0.001	726.5	XXX
	Anoxic	↓	Nonlinear	0.028 (0.787)	0.007	0.012	0.011	163.2	XXX
Ergosterol (%)	Oxic	↑	Nonlinear	0.625 (0.111)	0.607	0.299	0.739	17.8	XXX
	Anoxic	↑	Nonlinear	0.562 (0.144)	0.632	0.418	0.789	19.9	XXX
Fecosterol (%)	Oxic	↓	Nonlinear	0.541 (0.157)	0.064	0.061	0.074	12.8	XXX
	Anoxic	↓	Nonlinear	0.635 (0.107)	0.079	0.123	0.092	14.1	XXX
Campesterol (%)	Oxic	↓	Nonlinear	0.695 (0.079)	0.178	0.179	0.219	19.0	XXX
	Anoxic	↓	Nonlinear	0.722 (0.068)	0.175	0.300	0.215	18.4	XXX

Stigmasterol (%)	Oxic	↓	Nonlinear	0.429 (0.230)	0.079	0.167	0.092	13.5	XXX
	Anoxic	↓	Nonlinear	0.319 (0.321)	0.079	0.061	0.094	16.6	XXX
Sitosterol (%)	Oxic	↓	Nonlinear	0.447 (0.217)	0.221	0.101	0.292	24.1	XXX
	Anoxic	↓	Nonlinear	0.428 (0.231)	0.239	0.080	0.338	29.4	XXX
Sitostanol (%)	Oxic	↓	Nonlinear	0.530 (0.163)	0.055	0.200	0.061	10.4	XXX
	Anoxic	↓	Nonlinear	0.544 (0.155)	0.054	0.296	0.061	11.8	XXX

^a: The relationship is considered linear for $R^2 > 0.8$ with a p -value < 0.01 .

^b: The absolute value of the regression slope for the measured values.

^c: The absolute value of the regression slope for the measured values divided by the average standard deviation of the measurement. A value of the S_m/SD superior to 1.0 was considered as an acceptable sensitivity in the source discrimination relative to the measurement uncertainty

^d: The absolute value of the regression slope for the predicted values (e.g., data presented in Table S2).

^e: The difference (%) between the slope of the measured and predicted values. A difference of $< 10\%$ is considered an acceptable deviation. The value of 10% was arbitrarily chosen by the authors.

^f: The applicability is evaluated via the criteria based on R^2 , S_m/SD , and % difference. O means the value of the standard respect the critical value while X the indices failed (e.g., Critical values for R^2 and an associated p -value of R^2 , S_m/SD and % difference are > 0.8 and < 0.01 , > 0.5 , and $< 10\%$, respectively).

Table 3. The evaluation of selected source discrimination indices in the relationships with increasing algal carbon fraction (% Algae) of the artificial sediments at 60-days incubation.

Parameters	Oxygen condition	Tendency	Relationship type ^a	R ² (<i>p</i> -value)	S _m ^b	S _m /SD ^c	S _p ^d	% difference ^e	Applicability ^f
Extractable OM									
C1 (%)	Oxic	↓	Linear	0.967 (< 0.01)	0.514	0.614	0.493	4.4	000
	Anoxic	↓	Linear	0.980 (< 0.01)	0.499	0.795	0.477	4.5	000
C2 (%)	Oxic	↓	Linear	0.937 (< 0.01)	0.522	0.439	0.473	10.3	OXX
	Anoxic	↓	Linear	0.922 (< 0.01)	0.512	0.281	0.446	14.9	OXX
C3 (%)	Oxic	↑	Linear	0.643 (0.103)	0.551	0.284	0.424	30.0	XXX
	Anoxic	↑	Linear	0.737 (0.063)	0.582	0.168	0.457	27.5	XXX
C4 (%)	Oxic	↑	Linear	0.787 (0.045)	0.485	0.268	0.542	10.5	XXX
	Anoxic	↑	Linear	0.809 (0.038)	0.429	0.152	0.466	8.0	OXO
SUVA ₂₅₄ (L.mg ⁻¹ .m ⁻¹)	Oxic	↓	Linear	0.856 (0.024)	0.032	0.192	0.027	18.4	OXX
	Anoxic	↓	Nonlinear	0.611 (0.118)	0.026	0.080	0.019	36.9	XXX
FI	Oxic	↑	Nonlinear	0.654 (0.097)	0.003	0.200	0.003	5.7	XXO
	Anoxic	↑	Nonlinear	0.321 (0.319)	0.001	0.112	0.001	29.8	XXX
YFI	Oxic	↑	Linear	0.867 (0.021)	0.036	0.682	0.035	2.3	000
	Anoxic	↑	Linear	0.919 (< 0.01)	0.038	0.688	0.038	0.8	000
HIX	Oxic	↓	Linear	0.934 (< 0.01)	0.214	0.542	0.212	1.3	000
	Anoxic	↓	Linear	0.898 (0.014)	0.183	0.169	0.167	9.5	OXO
BIX	Oxic	↑	Nonlinear	0.739 (0.062)	0.009	0.534	0.010	8.5	XOX
	Anoxic	↑	Nonlinear	0.622 (0.113)	0.013	1.202	0.015	13.6	XOX
δ ¹³ C (‰)	Oxic	↑	Linear	0.969 (< 0.01)	0.150	1.431	0.151	0.7	000
	Anoxic	↑	Nonlinear	0.898 (0.014)	0.138	0.274	0.152	9.3	OXO
Cholesterol (%)	Oxic	↓	Nonlinear	0.346 (0.297)	0.038	0.068	0.036	5.8	XXO
	Anoxic	↓	Nonlinear	0.123 (0.563)	0.018	0.022	0.037	49.8	XXX
Ergosterol (%)	Oxic	↑	Nonlinear	0.536 (0.160)	0.724	0.835	0.911	20.6	XOX
	Anoxic	↑	Nonlinear	0.560 (0.146)	0.591	0.335	0.736	19.8	XXX
Fecosterol (%)	Oxic	↓	Nonlinear	0.328 (0.313)	0.051	0.069	0.066	23.7	XXX
	Anoxic	↓	Nonlinear	0.591 (0.129)	0.074	0.059	0.088	16.0	XXX
Campesterol (%)	Oxic	↓	Nonlinear	0.620 (0.114)	0.176	0.262	0.223	21.0	XXX
	Anoxic	↓	Nonlinear	0.630 (0.109)	0.172	0.392	0.219	21.6	XXX

Stigmasterol (%)	Oxic	↓	Nonlinear	0.495 (0.185)	0.091	0.084	0.098	6.9	XXO
	Anoxic	↓	Nonlinear	0.272 (0.367)	0.071	0.082	0.097	26.2	XXX
Sitosterol (%)	Oxic	↓	Nonlinear	0.427 (0.232)	0.296	0.140	0.401	26.3	XXX
	Anoxic	↓	Nonlinear	0.434 (0.227)	0.236	0.073	0.309	23.8	XXX
Sitostanol (%)	Oxic	↓	Nonlinear	0.638 (0.105)	0.057	0.328	0.069	17.0	XXX
	Anoxic	↓	Nonlinear	0.723 (0.068)	0.056	0.278	0.060	6.2	XXO

^a: The relationship is considered linear for $R^2 > 0.8$ with a p -value < 0.01 .

^b: The absolute value of the regression slope for the measured values.

^c: The absolute value of the regression slope for the measured values divided by the average standard deviation of the measurement. A value of the S_m/SD superior to 1.0 was considered as an acceptable sensitivity in the source discrimination relative to the measurement uncertainty

^d: The absolute value of the regression slope for the predicted values (e.g., data presented in Table S2).

^e: The difference (%) between the slope of the measured and predicted values. A difference of $< 10\%$ is considered an acceptable deviation.

^f: The applicability is evaluated via the criteria based on R^2 , S_m/SD , and % difference. O means the value of the standard respect the critical value while X the indices failed (e.g., Critical values for R^2 and an associated p -value of R^2 , S_m/SD and % difference are > 0.8 and < 0.01 , > 0.5 , and $< 10\%$, respectively).