



Characterization of particulate organic matter in a Guanabara Bay-coastal ocean transect using elemental, isotopic and molecular markers

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Abstract: Particulate organic matter (POM) from two periods (summer and fall) was characterized in a transect along the transition of the Guanabara Bay to the coastal ocean by elemental composition (C and N), isotopic ratios ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) and molecular markers (sterols, n-alcohols and fatty acids). Elemental (C/N of 5-7) and isotopic ($\delta^{13}\text{C}$ = -25.3 to -16.2 ‰ and $\delta^{15}\text{N}$ = 4.6 to 11.2 ‰) signatures were typical of planktonic organic matter. Fatty acids (totals of $12.08 \pm 7.78 \mu\text{g L}^{-1}$ in summer and $22.06 \pm 13.75 \mu\text{g L}^{-1}$ in the fall) occurred as short-chain, mono- and poly-unsaturated compounds. Total sterol concentrations ranged from 0.36 to $7.32 \mu\text{g L}^{-1}$, with predominance of cholesterol and phytosterols. Alcohols were a minor component (0.17 to $1.33 \mu\text{g L}^{-1}$). The multi-marker approach revealed an overwhelming contribution of autochthonous POM sources, reflecting the influence of the Guanabara Bay outflow on ecological processes in the transition zone considered herein. In addition, fecal material is also exported, although as a low intensity process. Moreover, the terrestrially-derived POM was not exported from the bay and accumulates in inner sediment. This supports the necessity to quantify such contributions to better constrain the carbon balance in the bay.

Keywords: lipid biomarkers, suspended particulate matter, isotopic composition, tropical bay

Resumo: Caracterização da matéria orgânica particulada em transecto Baía de Guanabara-oceano costeiro utilizando marcadores elementares, isotópicos e moleculares. A matéria orgânica particulada (MOP) de dois períodos (verão e outono) foi caracterizada ao longo de um transecto entre a Baía de Guanabara e o oceano costeiro através de composição elementar (C e N), razões isotópicas ($\delta^{13}\text{C}$ e $\delta^{15}\text{N}$) e marcadores moleculares (esteróis, n-álcoois e ácidos graxos). As assinaturas elementares (C/N = 5 a 7) e isotópicas ($\delta^{13}\text{C}$ = -25,3 a -16,2 ‰ e $\delta^{15}\text{N}$ = 4,6 a 11,2 ‰) foram típicas de matéria orgânica planctônica. Ácidos graxos (totais de $12,08 \pm 7,78 \mu\text{g L}^{-1}$ no verão e $22,06 \pm 13,75 \mu\text{g L}^{-1}$ no outono) ocorreram como compostos de cadeia curta, mono- e poli-insaturados. Concentrações de total de esteróis variaram de 0,36 a $7,32 \mu\text{g L}^{-1}$, com predominância de colesterol e fitoesteróis. Os álcoois foram um componente menos importante ($0,17$ a $1,33 \mu\text{g L}^{-1}$). A abordagem multiparâmetros revelou a contribuição predominante de fontes autóctones de MOP, o que reflete a influência da Baía de Guanabara sobre processos ecológicos na zona de transição aqui considerada. Material fecal também é exportado, embora seja um processo menos significativo. Além disso, a MOP de origem terrestre não é exportada da baía e acumula nos sedimentos internos. Isso suporta a necessidade de quantificar essa contribuição para melhor estabelecer o balanço de carbono na baía.

Palavras-chave: lipídios biomarcadores, material particulado em suspensão, composição isotópica, baía tropical.

Introduction

Rivers, estuaries/bays, tidal wetlands/mangroves and the coastal ocean represent a *continuum* of distinct but connected ecosystems that play an important role in the global carbon cycle (Hedges & Keil 1995; Bianchi & Canuel 2011; Bauer *et al.* 2013). Estuaries, in particular, are highly dynamic transitional regions where the mixture of river and sea water results in strong gradients of physico-chemical and chemical properties (Hobbie 2000). Therefore, estuaries are not only conduits of materials exchanged from land to sea reservoirs, but they also produce, transform, accumulate and/or export organic matter (Canuel *et al.* 2012). Carbon fluxes and cycling in estuaries are controlled by changes in precipitation, estuarine geomorphology and the magnitude and stoichiometry of nutrient input (Canuel *et al.* 2012; Bauer *et al.* 2013). Dissolved organic carbon (DOC) and particulate organic carbon (POC) in estuaries are derived from terrestrial, marine and estuarine primary productions. *In situ* production of OC in some estuarine waters can be significant to the coastal carbon budget because it can equal or exceed river or marine supplies. Estuaries also experience significant losses of OC owing to the combined influences of microbial degradation and photochemical oxidation, scavenging, sedimentation, and salinity-induced flocculation of DOC and POC (Canuel *et al.* 2012; Bauer *et al.* 2013).

Molecular biomarkers are organic compounds whose origin and/or chemical transformation may be related to a particular source of organic matter (OM) (Bianchi 2011). Lipids are less abundant in OM than proteins and other compounds, but they are the most common molecular markers in organic geochemistry because of their source-specificity and higher resistance to diagenesis than other organic groups (Volkman 2006; Bianchi & Canuel 2011). A benefit of using lipid biomarkers is that these compounds provide the ability to trace OM from phytoplankton, zooplankton, bacteria, and vascular plants, simultaneously. Different lipid classes, such as hydrocarbons, *n*-alkanes, fatty acids, sterols, *n*-alcohols and many others have been extensively used as molecular markers to assess the inputs of natural and anthropogenic OM to coastal environments (see reviews in Volkman 2006; Bianchi & Canuel 2011; Bianchi & Bauer 2012).

Lipid biomarkers and isotopic and elemental compositions have been considered to trace the inputs of natural and anthropogenic OM to several different aquatic systems along the Brazilian coast. These include estuaries and/or coastal bays (e.g., Martins *et al.* 2011; de Abreu-Mota *et al.* 2014; Richard *et al.* 2014; Carreira *et al.* 2016), lagoons (Carreira *et al.* 2011; Costa *et al.* 2011; Silva & Madureira 2012), mangroves (Mater *et al.* 2004) and along the continental margin (Carreira *et al.* 2012; Carreira *et al.* 2015a,b). Among them, the Guanabara Bay, in the Rio de Janeiro metropolitan region, is of special interest due to its economic, social, historic and environmental relevance.

The environmental changes in the Guanabara Bay were noticed in the early 1800's, but intensified in the last half of the 20th century. Information is available, for example, regarding the input of sewage and eutrophication (Kjerfve *et al.* 1997; Carreira *et al.* 2004; Lazzari *et al.* 2016), contamination of water, sediment and/or biota by toxic metals (Neto *et al.* 2006; Rangel *et al.* 2011; Abreu *et al.* 2016; de Carvalho Aguiar *et al.* 2016), hydrocarbons (Wagener *et al.* 2012; Mauad *et al.* 2015) and persistent organic pollutants (Dorneles *et al.* 2013), as well as changes in pelagic and benthic communities (Valentin *et al.* 1999; Eichler *et al.* 2003; Guenther *et al.* 2008; Vieira *et al.* 2008; Guenther *et al.* 2012; Fistarol *et al.* 2015).

The scientific literature dealing with the environmental quality of Guanabara Bay is vast, but little is known about the transfer of natural and anthropogenic materials between the bay and the coastal ocean. In order to amend this scenario, water samples along a transect crossing the bay's mouth were collected herein (Figure 1). The OM in the collected suspended particles was characterized considering three classes of lipid biomarkers compounds (fatty acids, sterols and *n*-alcohols), bulk parameters (elemental and isotopic compositions) and basic ancillary data.

Material and methods

Study region: The Guanabara Bay (Figure 1) is the second largest bay in the Brazilian coast, with an area of 384 km². The bay has a mean depth of 5.7 m, but its bathymetry is complex. From the entrance to the middle of the bay, the predominant feature is a channel with depths ranging from 30-40 m but can

reach over 50 m in some depressions, whereas in the north-western sector depths as low as 0.5 m are observed (Catanzaro *et al.* 2004). The hydrographic basin extending over 4,000 km² includes 45 rivers, six of which are responsible for 85% of the mean annual runoff of $100 \pm 59 \text{ m}^3 \text{ s}^{-1}$ (Kjerfve *et al.* 1997). The climate in the region is tropical humid, with a rainy summer and a dry winter; the mean annual temperature and rainfall are, respectively, 23.7 °C and 1500 mm (JICA 1994).

Tidal currents define the water circulation pattern in the bay, although gravitational currents are also important (Kjerfve *et al.* 1997). Maxima current velocities of 80-150 cm s⁻¹ have been observed at the entrance of the bay, whereas in the middle bay they vary around 30-50 cm s⁻¹, while current velocities below 30 cm s⁻¹ are observed in the inner bay (JICA 1994). Tidal amplitude averages at 0.7 m, but varies from 1.1 to 0.3 cm s⁻¹ during spring and neap tides, respectively. The estimated short mean half-water volume renewal is 11.4 days, although in some parts of the bay this time is considerably higher (Kjerfve *et al.* 1997).

Sampling and field measurements: Samples were collected in January 2014 (summer) and May 2014 (fall) at six stations distributed along the north-south axis of Guanabara Bay (Figure 1). A large volume *in situ* McLane (model LTV-08) pump was used to collect suspended particles at 1 m water depth onto pre-combusted (450 °C overnight) glass microfiber filters (Whatman type GF/F, 142 mm, 0.7 µm particle retention). The pump operated at a flow rate of 7 L min⁻¹ and stopped when the flow decreased to 4 L min⁻¹. Between 12 and 50 L were filtered at each station. Filters were stored in pre-combusted Petri dishes and kept refrigerated on ice until arrival at the laboratory.

Total suspended particulate matter (SPM), particulate organic carbon, particulate total nitrogen (TN) and isotopic ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) compositions of the particulate organic matter (POM) were determined in the water samples sampled at three depths – surface, middle water and close to the bottom. A glass bottle of 4 L that opened only at the desired depth was used. Water samples for SPM, POC, TN, $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were kept refrigerated in the field and filtered in the laboratory using pre-combusted glass microfiber filters (Whatman type GF/F, 47 mm, 0.7 µm particle retention). All filters were stored at -20 °C until chemical analysis.

Temperature and salinity vertical profiles were performed at each station using a specific probe

(CTD Castway from Sontek, with sampling frequency of 5Hz). The measurement accuracies were 0.05 °C and 0.1, respectively. The vertical measurement intervals were about 0.3 m for all temperature and salinity profiles. The raw data were plotted and visually inspected for removal of spurious temperature and salinity values using the Matlab Software.

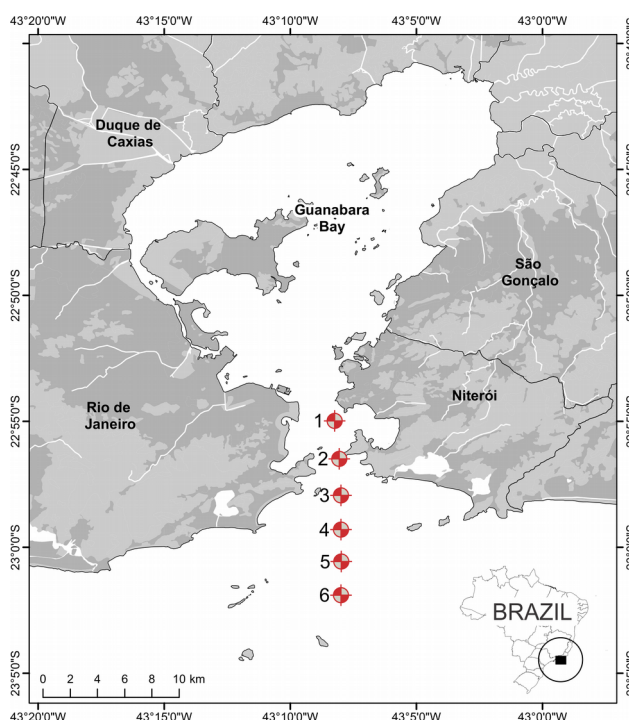


Figure 1. Sampling stations (1 to 6) in the Guanabara Bay-coastal ocean transect.

Reagents and chemicals: The 5 α -androstan-3 β -ol (98% purity), 5 α -cholestane (99% purity), 5 β -cholestan-3 β -ol (> 98% minimum purity), 5 β -cholestan-3 α -ol (> 95% minimum purity), cholest-5-en-3 β -ol (94% purity), 5 α -cholestan-3 β -ol (95% purity), 24-ethylcholest-5,22E-dien-3 β -ol (95% purity), 24-methylcholest-5-en-3 β -ol (ca. 65% purity), 24-ethylcholest-5-en-3 β -ol (98% purity), 24-methylcholesta-5,22-dien-3 β -ol, 5 β -cholestan-one, 1-octadecanol ($\geq 99.5\%$ purity), 1-nonadecanol (99% purity), 1-tetracosanol ($\geq 99\%$ purity), 1-hexacosanol ($\geq 97\%$ purity), tetracosane-d50 (98% purity), methyl nonadecanoate ($\geq 98\%$ purity) and Supelco® 37 Component FAME Mix standards were purchased from Sigma Aldrich. Hexanes (95% purity), dichloromethane (99.9% purity) and acetonitrile (99.8% purity) were supplied by Mallinckrodt; methanol (99.96% purity) and toluene (99.8% purity) by J. Baker; acetone (99.8% purity) by Qhemis and ethyl acetate (99.7% purity) by

Sigma Aldrich. Sodium sulfate anhydride (> 99% purity) was purchased from Sigma Aldrich and silica (silica gel 60, 0.063-0.200 mm) was supplied by Merck. N,O-bis(trimethylsilyl) trifluoroacetamide (BSTFA) was purchased from Supelco.

Elemental (POC, TN) and isotopic ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) compositions: Suspended particulate matter (SPM) was measured gravimetrically (± 0.01 mg) on the small (47 mm) freeze-dried filters. Sub-samples (2 cm²) of these filters were analyzed in duplicate for POC and TN on a Thermo Flash EA Elemental Analyser. Acid treatment with HCl vapor for 24 h removed the inorganic carbon prior to analysis (Hedges & Stern 1984). The final results represent the means of both analyses of each filter. Quantification was performed by the response factor relative to the cystine standard. The precision of the POC and TN analyses was always higher than 10%.

The carbon and nitrogen isotopic composition of the bulk particulate organic matter were analyzed in two additional sub-samples (2 cm²) of the small (47 mm) filters. The carbonate-free residues on the sub-samples were converted on a catalytic column in excess of pure oxygen to CO₂, N₂ and H₂O on a Thermo Flash EA Elemental Analyser. Water was removed with an anhydrous magnesium perchlorate trap. The other gases were directed to a Thermo Delta Plus stable isotope ratio mass spectrometer. Carbon and nitrogen isotopic compositions were expressed as the $\delta^N E = (R_{\text{sample}}/R_{\text{standard}} - 1)$, where N is the heavy isotope of element E and R is the abundance ratio of heavy to light isotopes of the elements ($^{13}\text{C}/^{12}\text{C}$; $^{15}\text{N}/^{14}\text{N}$). The standard USGS40 (L-glutamic acid) was used for instrument calibration and the final results were expressed relative to the Pee Dee (Belemnite) for $\delta^{13}\text{C}$ and atmospheric N₂ for $\delta^{15}\text{N}$. Precision of replicate analyses was 0.1 ‰.

Lipid biomarkers: Lipid biomarkers were determined only in the large (142 mm) filters collected at 1 m water depth, following procedures published elsewhere (Carreira *et al.* 2015b; Rada *et al.* 2016). Filters were freeze-dried and extracted with dichloromethane/methanol (9:1, v/v) using an ASE-200 Accelerated Solvent Extractor (Dionex®) at 80 °C and 1500 Psi (2 x 8 min cycles), after addition of androstanol (5 α -androstan-3 β -ol), methyl nonadecanoate (C₁₉-fatty acid methyl ester or C₁₉-FAME) and 1-nonadecanol (C₁₉-alcohol) as surrogate standards.

Bulk extracts were saponified (110° for 2h) in Reacti-Vials™ using an aqueous methanolic KOH 1

mol L⁻¹ solution. The reaction was stopped in an ice bath. Neutral lipids were extracted from the saponified extract with *n*-hexane (3 x 5 mL) at pH > 12. Sterols and *n*-alcohols in the neutral lipids were isolated by adsorptive chromatography on silica gel with dichloromethane:methanol (1:1, v/v) and finally derivatized with bis-trimethylsilyl-trifluoroacetamide (BSTFA, 70 °C for 1h). Fatty acids were recovered from the saponified extract residues at pH < 2.0 using *n*-hexane. After methylation with 3% boron trifluoride in methanol (BF₃-MeOH, 85 °C for 1h), the fatty acids were purified by adsorption chromatography with silica-gel following a published method (Canuel & Martens 1993).

Derivatized sterols and *n*-alcohols were determined by gas chromatography coupled to mass spectrometry (Trace GC Ultra, MS: ISQ with automatic sampler). An Agilent J&W DB-5 type column (5% methyl-phenyl siloxane, 30 m x 0.32 mm x 0.25 μm film) was used with helium as the carrier gas (maintained at a constant flow rate of 1.4 mL min⁻¹) and the following oven temperature program: 60 °C for 1 min, from 60 to 220 °C at 20 °C min⁻¹, up to 280 °C at 1 °C min⁻¹ and, finally, to 300 °C at 20 °C min⁻¹ with an isothermal hold of 5 min. The GC/MS system operated in electron ionization (EI) (70 eV) and full scan (m/z 50-550) modes. GC/MS quantification was performed using a calibration curve (100 ng mL⁻¹ to 5,000 ng mL⁻¹) with commercial standards (1-octadecanol, 1-nonadecanol, 1-tetracosanol, 1-hexacosanol, 5 β -cholestan-3 β -ol, cholest-5-en-3 β -ol, 5 α -cholestan-3 β -ol, 24-ethylcholesta-5,22-dien-3 β -ol, 24-ethylcholest-5-en-3 β -ol, 24-methylcholesta-5,22-dien-3 β -ol, 24-methylcholest-5-en-3 β -ol, 5 β -cholestan-one and androstan-3 β -ol) and by considering the peak areas of key ions (m/z 69.3, 83.4, 129.3, 215.5, 217, 231.5, 333.5 or 370.5 for sterols and m/z 103.3 for *n*-alcohols and phytol) and relative response factors vs. the internal standard 5 α -cholestane (m/z 217). Similar response factors of key ions were assumed for structurally related compounds for which standards were unavailable. GC/MS component identification was based on full scan spectra obtained from the available standards (see above) or by comparison with spectra reported in the literature for the other compounds.

Fatty acids as methyl-esters derivatives were determined by gas chromatography/flame ionization detection. An Agilent J&W DB-23 type column (50%-Cyanopropyl)-methylpolysiloxane, 60 m x 0.25 mm x 0.25 μm film) was used with helium as

the carrier gas (maintained at a constant flow rate of 1.4 mL min^{-1}) and the following oven temperature program: 170°C at $30^\circ\text{C min}^{-1}$, up to 170 to 215°C at 2°C min^{-1} , and, finally, to 240°C at $20^\circ\text{C min}^{-1}$. GC/FID quantification was performed using a calibration curve (500 ng mL^{-1} to $50,000 \text{ ng mL}^{-1}$) with commercial standards (Supelco® 37 Components FAME Mix) and methyl nonadecanoate and relative response factors vs. the internal standard tetracosane-d50. Other fatty acids not included in the standard mixture were quantified using response factors to compounds with similar structures.

Results

Water temperature and salinity profiles: The mean water column temperatures in the summer (Jan/2014) and fall (May/2014) were $17.2 \pm 2.7^\circ\text{C}$ and $20.4 \pm 2.3^\circ\text{C}$. The salinity had a mean value of 35.0 ± 0.8 for both campaigns. The complete dataset obtained by the CTD can be provided upon request. An overall increase in temperature and a decrease in salinity toward lower depths for all stations in both campaigns was observed (Fig. 2). Particularly, for the deepest stations (5 and 6), located outside the bay, this vertical decrease in salinity toward lower depths was due to the larger offshore excursion of the estuarine outflow plume during ebb tides, as was

the case for both campaigns. In these outside stations, the plume mixes with oceanic waters, mainly on surface levels, reducing the salinity differences from top to bottom. The stations 5 and 6 also display a larger temperature range from top to bottom, compared to the other stations, due to the stronger presence of cold oceanic subsurface waters as discussed later.

Bulk properties of suspended particles and organic matter: The data for suspended particulate matter (SPM), particulate organic carbon (POC) and total nitrogen (TN), C/N molar ratio and isotopic composition of organic matter ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) are displayed in Tables I and II. The SPM mean concentrations and range in January/2014 ($14.1 \pm 5.14 \text{ mg L}^{-1}$ / 7.51 to 26.8 mg L^{-1}) were lower than in May/2014 ($21.6 \pm 7.26 \text{ mg L}^{-1}$ / 11.9 to 39.1 mg L^{-1}). In both samplings, the highest SPM values were observed at stations inside the bay (i.e., 1, 2 and 3) and at depths of 1 m and 5 m in most cases.

The mean and range of POC content in January/2014 samples ($0.33 \pm 0.17 \text{ mg L}^{-1}$, range from 0.16 to 0.92 mg L^{-1}) were similar to the values observed in May/2014 ($0.36 \pm 0.15 \text{ mg L}^{-1}$, range from 0.17 to 0.77 mg L^{-1}). As observed for SPM, samples at 1 m and 5 m from stations 1, 2, 3 and 4 had relatively higher POC concentrations.

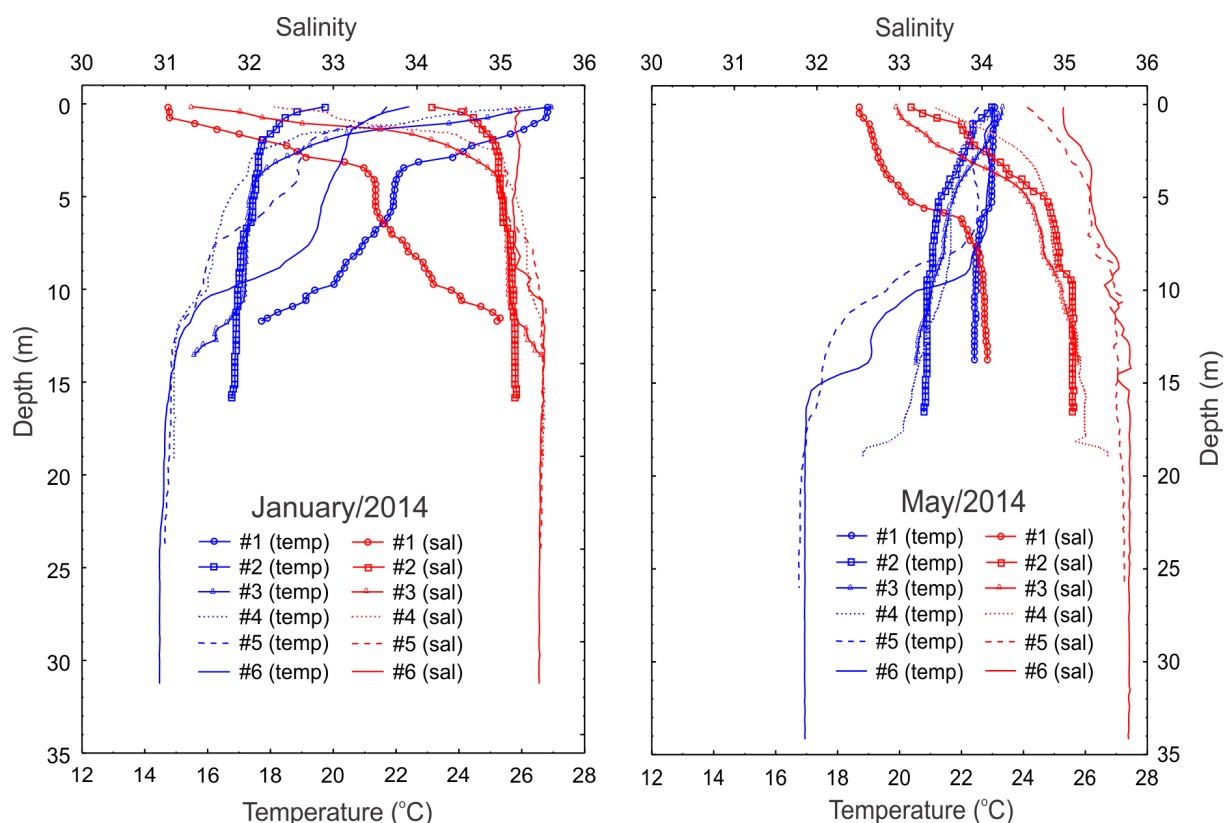


Figure 2. Vertical temperature and salinity during the January/2014 and May/2014 samplings.

Table I. January/2014. Suspended particulate matter (SPM), particulate organic carbon (POC; mg L⁻¹ and % of SPM), total nitrogen (TN), C/N molar ratio and carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) isotopic composition of the particulate organic matter in the Guanabara bay transect.

Station	Sampling depth (m)	SPM (mg L ⁻¹)	POC (mg L ⁻¹)	POC (%)	TN (mg L ⁻¹)	C/N (molar)	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)
1	1	26.8	0.27	1.00	0.06	5.32	-25.3	7.7
	5	22.9	0.54	2.34	0.09	6.70	-21.0	7.0
	8	10.3	0.30	2.87	0.05	6.37	-21.1	7.2
2	1	16.2	0.35	2.17	0.06	6.55	-21.1	6.5
	5	12.1	0.29	2.42	0.05	6.56	-21.4	7.1
	15	12.1	0.19	1.54	0.04	5.49	-23.1	5.0
3	1	19.4	0.92	4.73	0.17	6.19	-20.0	7.5
	5	15.7	0.33	2.10	0.06	6.24	-22.2	6.6
	13	17.1	0.29	1.72	0.05	6.48	-22.3	6.7
4	1	16.8	0.42	2.53	0.08	5.88	-20.2	7.5
	5	11.5	0.25	2.18	0.05	6.00	-21.6	6.8
	18	13.1	0.23	1.79	0.04	6.42	-23.6	5.1
5	1	12.5	0.24	1.95	0.05	5.49	-22.8	6.0
	5	12.9	0.24	1.87	0.05	5.95	-22.2	4.9
	20	7.51	0.40	5.39	0.08	5.95	-20.3	8.9
6	1	8.29	0.19	2.26	0.04	6.17	-20.9	5.9
	5	10.8	0.33	3.05	0.05	7.45	-20.5	4.6
	30	8.02	0.16	1.96	0.03	5.42	-24.9	4.6

Table II. May/2014. Suspended particulate matter (SPM), particulate organic carbon (POC; mg L⁻¹ and % of SPM), total nitrogen (TN), C/N molar ratio and carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) isotopic composition of the particulate organic matter in the Guanabara bay transect (nd = not detected).

Station	Sampling depth (m)	SPM (mg L ⁻¹)	POC (mg L ⁻¹)	POC (%)	TN (mg L ⁻¹)	C/N (molar)	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)
1	1	26.4	0.37	1.40	0.09	5.07	-18.6	10.8
	5	26.3	0.46	1.77	0.10	5.70	-18.5	10.2
	8	23.4	0.40	1.71	0.09	5.40	-17.7	11.2
2	1	26.3	0.42	1.60	0.10	4.94	-16.6	9.20
	5	23.0	0.46	1.99	0.08	6.35	-16.2	8.80
	15	22.7	0.39	1.71	0.08	5.76	-16.5	10.2
3	1	28.1	0.43	1.52	0.08	6.24	-16.6	9.80
	5	28.6	0.77	2.70	0.15	5.99	-16.8	10.5
	13	nd	Nd	nd	nd	nd	nd	Nd
4	1	18.8	0.36	1.91	0.07	6.04	-17.1	10.3
	5	39.1	0.47	1.21	0.10	5.72	-16.5	9.10
	18	20.5	0.32	1.58	0.07	5.46	-17.2	10.3
5	1	12.6	0.20	1.61	0.04	5.57	-18.4	10.4
	5	14.3	0.28	1.92	0.06	5.31	-18.4	10.1
	20	13.1	0.22	1.71	0.05	5.56	-18.7	10.2
6	1	17.5	0.17	0.97	0.04	5.65	-19.7	10.3
	5	14.8	0.19	1.30	0.04	6.32	-19.7	10.6
	30	11.9	0.18	1.53	0.03	6.93	-19.3	9.50

When POC is expressed as a percentage of SPM, slightly enriched OM particles were collected in January/2014 (2.4 ± 1.1 %) in comparison to May/2014 (1.6 ± 0.38 %). A high and positive correlation between POC and TN was observed in

both samplings (Figure 3), suggesting a common origin for both elements in the suspended particulate matter. The C/N molar ratio was very similar in both samplings (6.1 ± 0.50 in Jan/2014 and 5.8 ± 0.50 in May/2014), with no trend regarding position along

the transect in the water column. These values are typical of planktonic OM and were similar to the C/N means of 6.7 ± 1.3 observed in samples collected inside the bay during 2009/2010 (Kalas *et al.* 2009).

The carbon isotopic composition ($\delta^{13}\text{C}$) of POC showed distinct values in the two samplings (Tables I and II). In January/2014, $\delta^{13}\text{C}$ averaged -21.9 ± 1.5 (-20.0 to -25.3 ‰). In May/2014, on the other hand, enriched values of ^{13}C were measured, with $\delta^{13}\text{C}$ of -17.8 ± 1.2 ‰ (-16.2 to 19.7 ‰). The $\delta^{15}\text{N}$ values were also different between the two samplings: 6.4 ± 1.2 ‰ (4.6 to 8.9 ‰) in January/2014 and 10.1 ± 0.6 ‰ (8.8 to 11.2 ‰) in May/2014. No clear trend for $\delta^{15}\text{N}$ with respect to stations in the transect of the sampling depth was observed.

Composition and distribution of lipid biomarkers: Lipid biomarkers in the present study included 35 fatty acids (saturated, mono-unsaturated, polyunsaturated and branched compounds), 12 sterols (unsaturated and saturated compounds) and 11 alcohols (saturated straight chain compounds). The results for the two samplings are listed in Table III. Detailed compositions of fatty acids and n-alcohols are provided as Supplementary Material (Tables S1 and S2).

Fatty acids, as methyl esters or FAMES, were the most abundant class of compounds, with means of 12.1 ± 7.78 $\mu\text{g L}^{-1}$ (75.6% of total quantified lipids) in January/2014 and 22.1 ± 13.7 $\mu\text{g L}^{-1}$ (92.1 % of total quantified lipids) in May/2014. The second most abundant class was composed of sterols, with 2.61 ± 2.34 $\mu\text{g L}^{-1}$ (17% of total) and 1.32 ± 1.12 $\mu\text{g L}^{-1}$ (5.85% of total) in January/2014

and May/2014, respectively. Finally, n-alcohols varied between 0.87 ± 0.30 $\mu\text{g L}^{-1}$ (7.36% of total) and 0.62 ± 0.54 $\mu\text{g L}^{-1}$ (2.0% of total) in the two samplings. Total quantified lipid concentrations varied from 6.02 to 46.25 $\mu\text{g L}^{-1}$ considering all samples (Table III), with no clear trend among the stations. The higher values of total quantified lipids at stations 5 and 6 in January/2014 and 3 and 4 in May/2014 are noteworthy.

Fatty acids (FAs) occurred as compounds with a predominance of even-numbered chains ranging from 14 to 26 C atoms ($\text{C}_{14} - \text{C}_{26}$). Short-chain and saturated compounds (SCFA, $\text{C}_{14} - \text{C}_{18}$) represented on average $30.2 \pm 7.5\%$ and $56.0 \pm 10.6\%$ of total FAs in each sampling. Higher SCFA concentrations were measured in the second sampling, particularly at station 3 (Table 3). Monounsaturated FAs with C_{16} , C_{17} , C_{18} , C_{20} and C_{22} (MUFAs) contributed with similar proportions in the two samplings, averaging $17.2 \pm 6.0\%$ and $14.0 \pm 3.9\%$ of total FAs in January/2014 and May/2014. The most abundant MUFA was $\text{C}_{16:1}$. Long-chain fatty acids (LCFA), saturated compounds with chain-length $\geq \text{C}_{24}$ were virtually absent in the first sampling and represented less than 0.5 % of total FAs in the second sampling. On the other hand, polyunsaturated FAs (PUFAs), with 18, 20 and 22 carbon atoms and 2 to 6 unsaturations, were abundant, representing circa 20 % of total FAs in the two samplings. Finally, branched FAs (*iso*- and *anteiso*- C_{15} , C_{17} and 10-methyl- C_{16}) were more abundant in the first sampling ($6.2 \pm 7.5\%$ of total FAs) when compared to the second one ($2.6 \pm 0.8\%$ of total FAs).

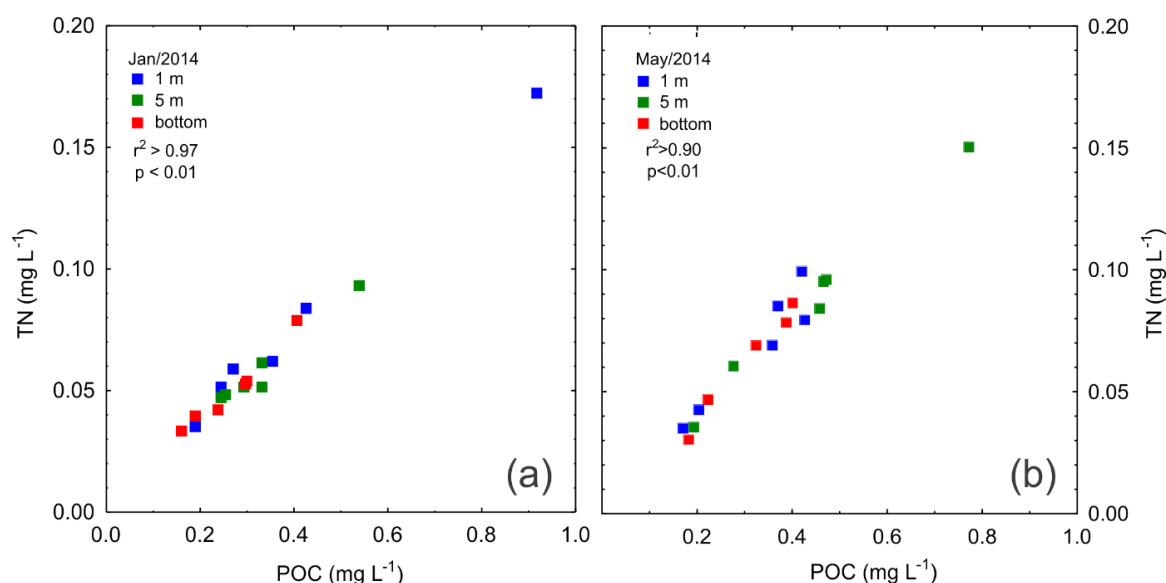


Figure 3. Linear correlation between POC and TN in (a) January/2014 and (b) May/2014 considering samples collected at 1 m, 5 m and close to the bottom

Table III. Lipid biomarkers (fatty acids, n-alcohols and sterols) in the suspended organic matter in the two samplings (January and May of 2014) in the Guanabara bay transect (na = not analyzed).

Station	C1 (Jan/2014)						C2 (May/2014)					
	1	2	3	4	5	6	1	2	3	4	5	6
^A Fatty acids ($\mu\text{g L}^{-1}$)												
Total	13.89	4.38	8.08	5.01	24.45	16.66	na	27.91	41.47	22.75	11.09	7.09
SCFA	4.54	1.69	2.64	2.06	12.00	7.39	na	18.37	20.31	11.65	5.53	5.45
LCFA	nd	nd	nd	nd	0.05	nd	na	0.00	0.19	0.08	0.03	0.03
MUFA	3.88	1.07	2.11	1.28	1.52	5.10	na	4.62	5.67	3.26	2.18	0.68
PUFA	4.61	1.35	2.73	1.39	4.85	3.42	na	3.19	14.03	4.59	2.83	0.66
Branched	0.69	0.19	0.46	0.19	5.67	0.41	na	1.05	0.75	2.95	0.34	0.17
^B Alcohols ($\mu\text{g L}^{-1}$)												
Total	0.99	0.56	0.94	1.05	0.44	1.20	1.33	na	1.07	0.17	0.22	0.31
SCOH	0.06	0.23	0.39	0.44	0.15	0.24	0.75	na	0.12	0.01	0.03	0.14
LCOH	0.25	0.11	0.18	0.19	0.12	0.33	0.04	na	0.05	< lq	0.01	0.01
Phytol	0.61	0.18	0.31	0.34	0.13	0.54	0.49	na	0.76	0.13	0.14	0.11
^C Sterols ($\mu\text{g L}^{-1}$)												
Total	2.24	1.08	1.60	1.56	1.75	7.29	2.38	na	3.70	0.75	0.63	0.80
26 $\Delta^{5,22}$	0.01	0.03	0.02	0.02	0.05	0.29	0.06	na	0.06	0.02	0.02	0.02
27 $\Delta^{0,5b}$	0.07	0.06	0.05	0.05	0.05	0.08	0.14	na	0.08	0.02	0.03	0.01
27 Δ^5	0.51	0.28	0.37	0.36	0.51	1.62	0.32	na	0.21	0.06	0.07	0.08
27 $\Delta^{5,22}$	0.11	0.07	0.09	0.08	0.15	0.58	0.12	na	0.10	0.03	0.04	0.04
28 Δ^5	0.03	0.02	0.02	0.02	0.04	1.39	0.07	na	0.07	0.02	0.01	0.02
28 $\Delta^{5,22}$	0.30	0.16	0.21	0.23	0.28	1.08	0.23	na	0.23	0.06	0.07	0.09
28 $\Delta^{5,24(28)}$	0.28	0.16	0.18	0.20	0.38	1.48	0.25	na	0.28	0.12	0.16	0.19
29 Δ^5	0.30	0.10	0.19	0.16	0.11	0.35	0.13	na	0.11	0.03	0.03	0.03
29 $\Delta^{5,22}$	0.09	0.03	0.06	0.06	0.06	0.16	0.08	na	0.08	0.02	0.02	0.02
30 Δ^{22}	0.40	0.13	0.29	0.28	0.06	0.13	0.91	na	2.47	0.36	0.17	0.29
27 Δ^0	0.08	0.03	0.06	0.06	0.03	0.08	0.04	na	0.02	0.01	0.01	0.01
29 Δ^0	0.06	0.02	0.05	0.04	0.02	0.05	0.01	na	< lq	< lq	< lq	< lq
Total lipids ($\mu\text{g L}^{-1}$)	17.12	6.02	10.62	7.61	26.64	25.15	na	na	46.25	23.67	11.94	8.20

nd = not detected; < lq = below limit of detection ($0.01 \mu\text{g L}^{-1}$);

^A Short-chain (SCFAs < C₂₂), long-chain (LCFA $\geq$ C₂₄), monounsaturated (MUFAs: C₁₆, C₁₇, C₁₈, C₂₀ and C₂₂), polyunsaturated (PUFAs: C_{20:2}, C_{20:3}, C_{22:2}, C_{22:6}) and branched fatty acids (*iso* and *anteiso*- C₁₅ and C₁₇ and 10-methyl-C₁₇);

^B Short-chain (SCOH $\leq$ C₂₂) and long-chain (LCOH $\geq$ C₂₄) alcohols

^C The symbol a $\Delta^{b,c}$ (a = number of C atoms; b, c = position of the unsaturation in the C structure) stands for: 24-norcholesta-5,22-dien-3 β -ol (26 $\Delta^{5,22}$), 5 β -cholestan-3 β -ol (27 $\Delta^{0,5b}$, coprostanol), cholest-5-en-3 β -ol (27 Δ^5 ; cholesterol), cholesta-5,22E-dien-3 β -ol (27 $\Delta^{5,22}$), 24-methylcholesta-5-en-3 β -ol (28 Δ^5), 24-methylcholesta-5,22E-dien-3 β -ol (28 $\Delta^{5,22}$), 24-methylcholesta-5,24(28)-dien-3 β -ol (28 $\Delta^{5,24(28)}$), 24-ethylcholesta-5-en-3 β -ol (29 Δ^5), 24-ethylcholesta-5,22E-dien-3 β -ol (29 $\Delta^{5,22}$), 4 α -23,24-trimethyl-5 α -cholest-22(E)-en-3 β -ol (30 Δ^{22}), 5 α -cholestan-3 β -ol (27 Δ^0 ; cholesterol) and 24-ethyl-5 α -cholestan-3 β -ol (29 Δ^0)

Twelve sterols were found containing between C₂₆ and C₃₀ at total concentrations averaging $2.59 \pm 2.34 \mu\text{g L}^{-1}$ in January/2014 and $1.41 \pm 1.35 \mu\text{g L}^{-1}$ in May/2014 (Table 3). The large standard deviations of the means were caused by very high concentrations at station 6 in the first sampling ($7.30 \mu\text{g L}^{-1}$ of total sterols) and at stations 3 and 4 (2.38 and $3.70 \mu\text{g L}^{-1}$ of total sterols, respectively). In the first sampling, cholest-5-en-3 β -ol (cholesterol; 27 Δ^5) was the most abundant sterol (24.5 ± 2.7 % of total sterols), followed by 24-methylcholesta-5,24(28)-dien-3 β -ol (28 $\Delta^{5,24(28)}$; 15.5 ± 4.33 %), 24-methylcholesta-5,22E-dien-3 β -ol (28 $\Delta^{5,22}$; 14.5 ± 1.1 %) and 4 α ,23,24-trimethylcholesta-22-en-3 β -ol (dinosterol, 30 Δ^{22} ; 11.9 ± 7.4 %). However, during the second sampling, a high prevalence of dinosterol was observed, contributing to 43.4 ± 15.2 % of the total sterols, whereas the other abundant sterols in the first sampling represented between 9.5 and 16.4 % of the total sterols, on average.

Other usually abundant sterols in aquatic systems, such as 24-ethylcholesta-5-en-3 β -ol (sitosterol; 29 Δ^5) and 24-ethylcholesta-5,22-dien-3 β -ol (29 $\Delta^{5,22}$, stigmasterol) (Bianchi & Canuel 2011), were relatively less important, with an overall contribution of 3-6 % of the total sterols. These results are consistent with those observed by Kalas *et al.* (2009), who showed a higher contribution of 29 Δ^5 and 29 $\Delta^{5,22}$ to the SPM only at stations in the northern portions of the bay, but not at its mouth. Saturated sterols (5 α -cholestan-3 β -ol and 5 α -24-ethylcholesta-3 β -ol) were found in minor concentrations. Lastly, the fecal sterol 5 β -cholesta-3 β -ol (Bianchi & Canuel 2011) were detected in all stations, at concentrations typically below $0.08 \mu\text{g L}^{-1}$, but reaching $0.138 \mu\text{g L}^{-1}$ at station 1 in the second sampling (Table 3).

The last class of quantified lipid biomarkers (alcohols) was marked by the large predominance of phytol (3,7,11,15-tetramethyl-2-hexadecene-1-ol)

and the C₁₆-alcohol, both summing up circa 70 % of total alcohols. Regarding long-chain alcohols (LCOH; $\geq C_{22}$), they were only detected in appreciable amounts in January/2014, particularly C₂₄OH (ca 10 % of total alcohols) and C₃₀OH (4.5 – 9.0 % of total alcohols).

Discussion

Water types in the studied region: Temperature-Salinity (TS) diagrams displayed, on both campaigns, the presence of two water masses in the studied region, as indicated by the relatively linear shape of the TS curves (Figure 4). The Coastal Water (CW) is indicated by salinities lower than 33 (Castro *et al.* 2006), whereas the presence of South Atlantic Central Water (SACW) is indicated by temperatures above 6 °C and below 20 °C and salinities between 34.6 and 36.2 (Miranda 1985). Mixing of these two water masses are seen to occur at the subsurface. The Tropical Water (TW), usually found at the surface, outside the bay mouth (Bergamo 2006), was not detected because the larger estuarine outflow, during the ebb tide, pushes TW offshore. This larger outflow intensifies the mixing of CW and TW then reducing the surface salinity values below 36.2 as verified at stations 5 and 6.

On January/2014, lower salinity values occur at surface on the bay as the result of larger river discharges during the wet season, from November to April, than during the dry season, from May to October. Also, easterly winds that blow over the continental shelf, predominantly during the austral spring and summer (Castro *et al.* 2006) causes surface Ekman transport towards the ocean. This contributes to a stronger presence of SACW at shallow depths along the inner shelf with eventual intrusions in the Guanabara Bay (Melo 2004), where it was outstandingly observed very near the surface on station 2 (Figure 4). As a result, on Fig. 4, the TS diagram displays a larger presence of SACW on January in comparison to May of 2014.

On January/2014, lower salinity values occur at surface on the bay as the result of larger river discharges during the wet season, from November to April, than during the dry season, from May to October. Also in January, the prevailing easterly winds that blow over the continental shelf contributed to a stronger presence of SACW at shallow depths along the inner shelf and also within the Guanabara Bay, where it was outstandingly observed very near the surface on station 2 (Figure 4).

Bulk geochemical properties of particulate organic

carbon: Mean C/N molar ratio values of suspended particles were similar in both samplings (6.1 ± 0.50 in Jan/2014 and 5.8 ± 0.51 in May/2014). These values suggest the predominance of autochthonous rather than allochthonous contribution to the pool of particulate organic matter (Meyers 1997; Maksymowska *et al.* 2000; Bianchi & Canuel 2011; Wakeham & McNichol 2014). The carbon and nitrogen isotopic compositions of the particulate organic matter (POM) also ranged within values usually ascribed to autochthonous organic matter ($\delta^{13}C = -19.9 \pm 2.5$ ‰ and $\delta^{15}N = 8.2 \pm 2.1$ ‰). However, both $\delta^{13}C$ and $\delta^{15}N$ showed significantly ($p < 0.001$) different values between samplings (Figure 5). All POM samples in January/2014 had depleted ^{13}C ($\delta^{13}C < -20.0$ ‰) and ^{15}N ($\delta^{15}N < 8.0$, except for one sample), but in May/2014 the opposite trend was observed, of enriched ^{13}C ($-20.0 < \delta^{13}C < -16.0$ ‰) and ^{15}N ($8.8 < \delta^{15}N < 11.2$ ‰).

The results of the isotopic composition between samplings described above may be explained by changes in the autotrophic activity of each sampling. Lower fractionation towards the heavy isotope occurs when photosynthetic activity is high, resulting in an isotopically-enriched phytoplanktonic organic matter (Fogel & Cifuentes 1993). This was the case of samples collected in May/2014 (Figure 5). For carbon, there was incorporation of ^{13}C -enriched HCO_3^- resulting in less negative $\delta^{13}C$, whereas for nitrogen the heavy isotope enrichment is probably caused by the incorporation ammonium produced by the mineralization of organic matter (Middelburg & Nieuwenhuize 2001).

It is noteworthy that Kalas *et al.* (2009) also found isotopically-enriched POM in stations inside the bay. Therefore, our results indicate events of higher phytoplanktonic activity occurring in the transition area from the Guanabara Bay to the coastal ocean. One last point to be addressed is the depleted ^{13}C ($\delta^{13}C < -23.0$ ‰) values for bottom samples at stations 2, 4 and 6 in January/2014 (Figure 5), which may be related to some influence of resuspension of more refractory OM deposited in the sediment.

Characterization of suspended organic matter using lipid biomarkers: The lipid biomarker composition of surface samples was considered in order to characterize in further details the source of particulate organic matter in the bay-coastal ocean transect. Due to the relatively limited dataset, the traditional multivariate statistical analysis approach (e.g., Canuel 2001; Yunker *et al.* 2005) was not

considered herein. Instead, lipids were grouped according to their assignment to specific sources of organic matter (McCallister *et al.* 2006; Yoshinaga *et al.* 2008; Schmidt *et al.* 2010).

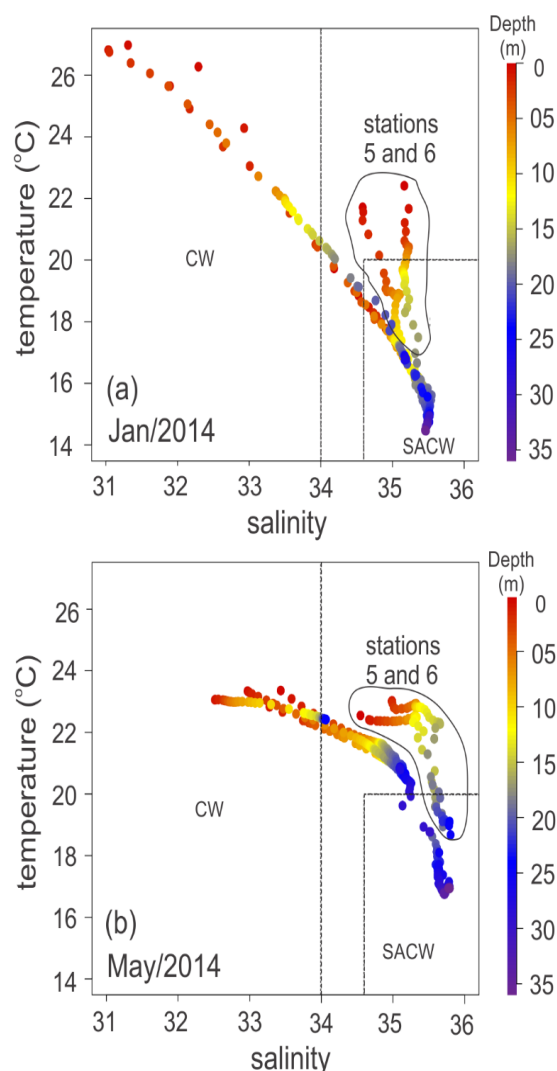


Figure 4. Temperature-salinity diagrams of waters collected in (a) January/2014 and (b) May/2014 at six stations at the transect along the Guanabara Bay-coastal ocean transition. Data for stations 5 and 6 are highlighted. Two types of water masses are marked by dashed-lines: CW = Coastal Water and SACW = South Atlantic Central Waters. See text for further information about water masses constraints.

Our lipid biomarker data allowed for the definition of four groups, as follows: (i) primary production or autochthonous sources: sum of cholest-5-en-3 β -ol ($27\Delta^5$), cholesta-5,22-dien-3 β -ol ($27\Delta^{5,22}$), 24-methylcholesta-5,22-dien-3 β -ol ($28\Delta^{5,22}$), 4 α ,23,24-trimethylcholest-22-en-3 β -ol ($30\Delta^{22}$), 24-

methylcholesta-5,24(28)-dien-3 β -ol ($28\Delta^{5,24(28)}$) and 24-norcholesta-5,22-dien-3 β -ol ($26\Delta^{5,22}$); (ii) terrestrial/riverine: sum of long-chain fatty acids (LCFA) and long-chain n-alcohols (LCOH); (iii) zooplankton: sum of cholesterol ($27\Delta^5$) and mono-unsaturated fatty acid $C_{18:1w9}$; (iv) bacteria: sum of iso- and anteiso- branched fatty acids C_{15} and C_{17} .

Figure 6 displays the resulting lipid biomarker-based groups of organic matter, including representations as a percentage of each group (Fig. 6-a) and as POC-normalized concentrations (Fig. 6-b). An overwhelming contribution of OM produced by primary production in both samplings is observed, followed by an important contribution of secondary production (i.e., zooplankton). This reveals enhanced eutrophication caused by an excess of nutrients and consequent high rates of accumulation of organic matter in sediment not only inside the bay (Carreira *et al.* 2002; Cotovicz Jr *et al.* 2015), but elevated phytoplankton activity is also observed in the bay-coastal ocean transition zone. This finding is consistent with recent estimates of a 0.01 % contribution by Guanabara Bay to the total global carbon influx to the ocean associated with nutrients exported by the bay (Lazzari *et al.* 2016).

Another interesting feature observed in Figure 6-a is the very low contribution of terrestrial-riverine OM in suspended particles. This suggests retention in of OM delivered by the drainage basin in the bay sediment. Only in January/2014 were at most stations (except station 5) a relatively higher contribution of 4.7 ± 1.4 % from terrestrial-riverine OM observed, which can be derived from a higher influence of runoff during the rainy summer season. In fact, the relatively low salinities of surface water during the January/2014 samplings (values between 31-33; Figure 2), which was not observed in May/2014, are consistent with a higher influence of continental runoff in the sampling region. The contribution of bacterially-derived OM in suspended particles represented a relatively important fraction of 7.0 ± 2.0 %, if sample 5 collected in January/2014 is excluded. The very high contribution (44.1%) of bacteria to POC at station 5 in January/2014 was caused by the branched iso- C_{17} fatty acid. Our data is consistent with previous observations that bacterioplankton metabolism in surface waters at the entrance to the Guanabara Bay is responsible, alongside phytoplankton, for producing particulate organic carbon (Guenther *et al.* 2008).

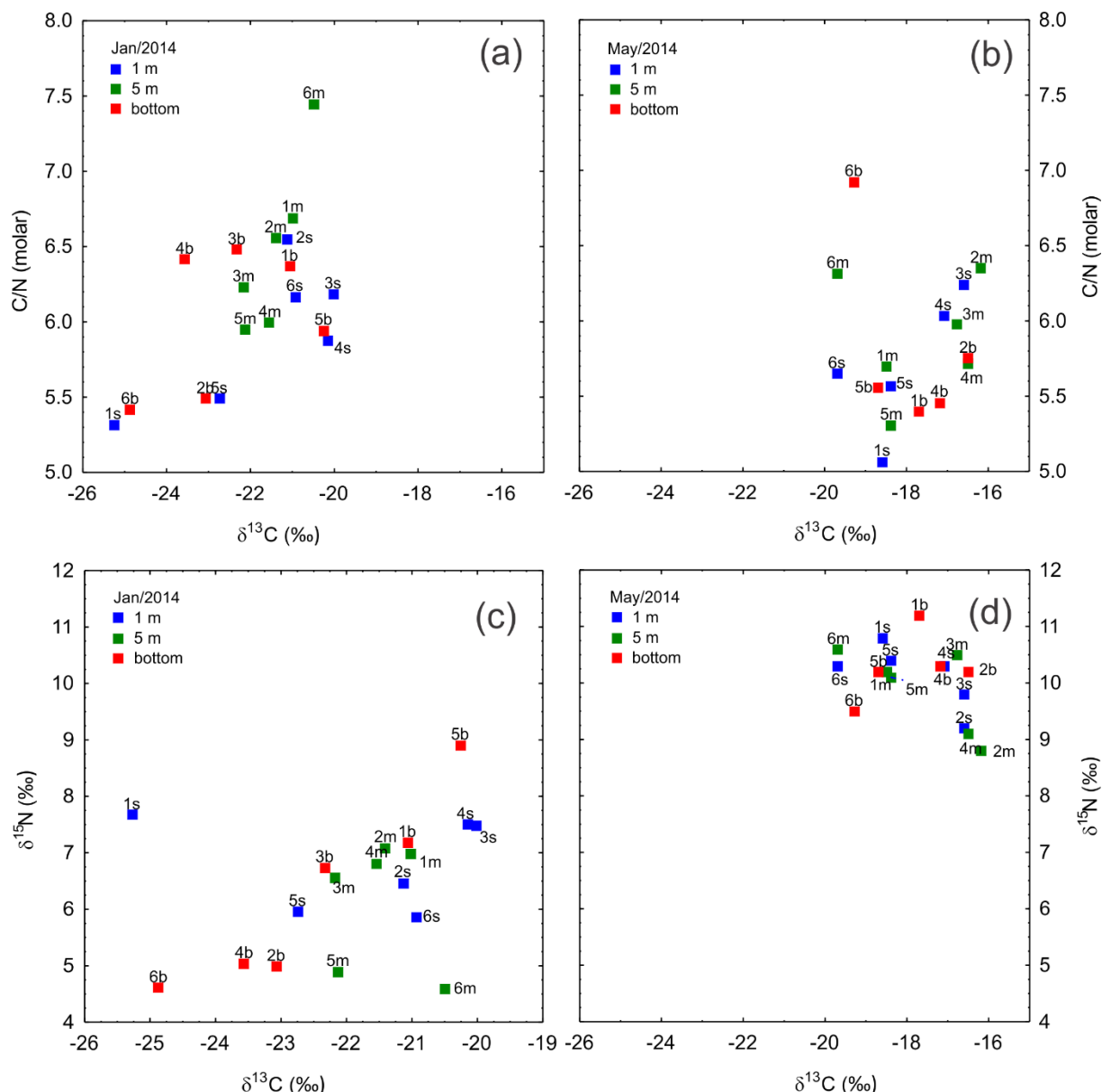


Figure 5. Plots of the C/N and $\delta^{13}\text{C}$ (a,b) and $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ (c,d) results for January and May/2014

An additional way to evaluate the contribution of groups to the pool of suspended organic matter is by evaluating POC-normalized concentrations of lipid biomarkers (Figure 6-b). Unfortunately, stations 1 and 2 in May/2014 could not be displayed because part of the data was lost. The higher contribution of lipids to POC in May/2014 compared to January/2014 is consistent with the isotopic composition data discussed previously, which suggests an enhanced primary production activity during the second sampling. In addition, during the fieldwork in January/2014, the water at stations 5 and 6 was clearly altered by a massive phytoplankton bloom. This event caused an increase in POC-normalized lipid concentrations at stations 5 and 6 when compared to the other stations in the

same sampling. Similarly, this algal bloom might be responsible for the high contribution of bacterial-derived OM observed at station 5 in January/2014, although we cannot explain why a similar influence on station 6 was not observed (Figure 6-b).

No similar previous information of fatty acids, sterols and alcohols for suspended particulate matter outside the Guanabara Bay is available for comparison. However, Kalas *et al.* (2009) found 1 to 4 $\mu\text{g mg POC}^{-1}$ of sterols in samples collected inside the bay in 2009/2010, whereas our samples collected in 2014 at the mouth and outside Guanabara Bay showed higher values, ranging between 2 and 8.7 $\mu\text{g mg POC}^{-1}$. Considering that most sterols in both studies are derived from plankton, this difference in POC-normalized sterols might be ascribed to an increase

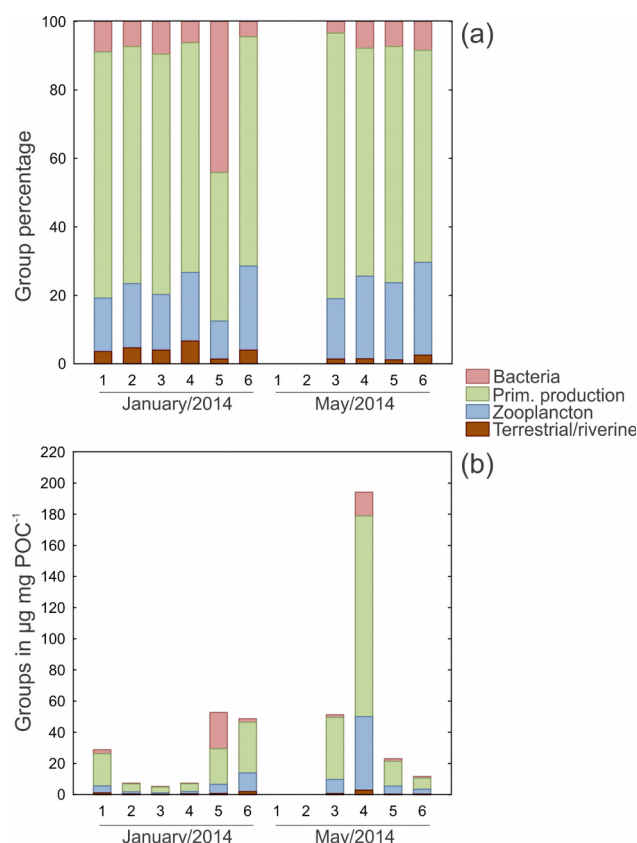


Figure 6. Lipid grouping according to sources of particulate organic matter (bacteria, primary production, zooplankton and terrestrial/riverine) in (a) percentage and (b) normalized concentrations. See text for details regarding the group-definition criteria.

in the contribution of primary production to the POM pool in the studied area over time.

Contribution of sewage-derived OM: Coprostanol (5 β -cholestan-3 β -ol) is a traditional tracer of fecal material in aquatic systems (Takada & Eganhouse 1998). Many studies regarding sewage contamination in Guanabara Bay waters and sediments using coprostanol and other fecal sterols are available (Carreira *et al.* 2004; Costa & Carreira 2005; Cordeiro *et al.* 2008; Santos *et al.* 2008; Kalas *et al.* 2009), but this is the first report of coprostanol in suspended particles of waters outside the bay (Table III). The mean concentration of coprostanol was $0.06 \pm 0.03 \mu\text{g L}^{-1}$, considering the two samplings. Lazzari *et al.* (2016) found higher values of coprostanol in SPM ($0.45 \pm 0.20 \mu\text{g L}^{-1}$ to $0.65 \pm 0.30 \mu\text{g L}^{-1}$) in the vicinity of our station 1, in the inner bay. Therefore, the sewage particles are efficiently 'diluted' by other types of OM and/or coprostanol is degraded during transport in the oxygenated water column (Bianchi & Canuel 2011).

Notwithstanding the low concentrations of coprostanol in the bay-coastal water transect, the

consideration of diagnostic indexes can aid in confirming the relevance of sewage contamination. One such index is formed by the coprostanol to coprostanol plus cholesterol ratio (Grimalt *et al.* 1990): values above 0.7 confirm the fecal origin of coprostanol, while below 0.3 fecal contamination is not confirmed. Herein, the overall means of this index was 0.62 ± 0.13 , very similar to the threshold proposed in the literature, confirming contamination of suspended particles by fecal material. In fact, in the second sampling the values of this ratio were higher than the overall means, ranging between 0.62 and 0.78, indicating a high influence of fecal material on suspended particles in May/2014.

Conclusion

The isotopic and lipid biomarker approach successfully allowed for the characterization of the suspended particulate organic matter in the Guanabara Bay-coastal ocean transition zone. The isotopic composition ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) indicated that the bulk of the organic matter is strongly influenced by autotrophic processes, which can occur both by *in situ* production and the contribution of material originated from the bay. This scenario was confirmed through lipid biomarkers, and allows for the conclusion that the Guanabara Bay is directly (by the export of autochthonous OM) and/or indirectly (by the export of nutrients) responsible for the increment in phytoplanktonic production in the water column in the coastal waters adjacent to the bay. Importantly, the autotrophic processes varied widely between the two sampling periods, indicating that the study area is strongly influenced by prevailing oceanographic conditions. It was also possible to identify direct export of fecal material in particulate form to the coastal ocean, although this seems to be a low intensity process. Another important result is the finding that the organic matter of terrestrial and/or water origin is retained in the bay and probably accumulates in the sediments. The quantification of this allochthonous source contribution needs to be identified to better understand the processes that lead to the increase in OM storage in the bay's sediment.

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Characterization of particulate organic matter in a Guanabara Bay-coastal ocean transect using elemental, isotopic and molecular markers

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Supplementary Material

Table S1. Distribution of individual fatty acids in the suspended particles in the two namplings. Values in $\mu\text{g L}^{-1}$.

Station	January/2014						May/2014					
	1	2	3	4	5	6	1	2	3	4	5	6
Fatty acids*												
14:0	1.20	0.53	0.72	0.56	4.68	3.12	na	1.78	1.64	1.20	0.84	0.82
14:1	0.16	0.02	0.08	0.03	0.07	0.03	na	0.08	0.05	0.27	0.02	0.01
i15	0.46	0.11	0.27	0.13	0.26	0.18	na	0.53	0.37	2.66	0.16	0.08
a15	0.20	0.06	0.14	0.07	0.15	0.12	na	0.40	0.29	0.22	0.14	0.07
15:0	0.11	0.05	0.08	0.06	0.25	0.18	na	0.27	0.16	0.14	0.08	0.05
15:1	<lq	<lq	0.04	<lq	<lq	<lq	na	<lq	0.07	<lq	<lq	0.02
16:0	3.21	1.06	1.83	1.50	6.93	4.08	na	15.08	17.20	9.60	4.37	4.22
16:1 ω 7	2.59	0.82	1.51	0.96	0.07	4.34	na	1.77	1.69	1.20	1.00	0.14
i17	<lq	<lq	0.04	<lq	5.10	0.11	na	0.05	0.06	0.04	0.03	0.01
a17	0.03	0.01	0.02	<lq	0.16	<lq	na	0.06	0.03	0.03	0.02	0.01
17:0	0.04	0.03	0.03	0.02	0.01	0.10	na	0.18	0.11	0.09	0.04	0.03
17:1	0.39	0.02	0.03	0.02	0.43	0.08	na	0.29	0.10	0.11	0.08	0.04
18:0	0.12	0.09	0.08	<lq	0.40	0.18	na	1.51	1.39	0.80	0.32	0.41
18:1 ω 9t	0.04	0.02	<lq	<lq	0.09	0.10	na	<lq	0.02	<lq	<lq	0.03
18:1 ω 9c	0.65	0.19	0.42	0.27	0.82	0.53	na	2.48	3.63	1.69	0.98	0.42
18:2 ω 6t	0.03	<lq	0.03	<lq	0.01	<lq	na	0.14	<lq	0.05	0.03	0.02
18:2 ω 6c	0.55	0.13	0.30	0.17	0.61	0.42	na	1.40	3.13	1.16	0.54	0.30
18:3 ω 6	0.07	0.02	0.05	0.03	0.11	0.09	na	0.27	0.58	0.16	0.05	0.01
18:3 ω 3	1.66	0.31	0.99	0.49	1.15	0.51	na	0.49	0.83	0.64	0.70	0.27
20:0	<lq	<lq	<lq	<lq	0.04	<lq	na	<lq	<lq	<lq	0.03	0.03
20:1	0.03	<lq	0.02	<lq	0.03	0.02	na	<lq	0.05	<lq	0.08	0.01
20:2	<lq	<lq	<lq	<lq	0.01	0.03	na	<lq	<lq	<lq	<lq	<lq
20:3 ω 6	<lq	<lq	0.02	<lq	0.03	0.03	na	<lq	<lq	<lq	<lq	<lq
20:3 ω 3	0.14	0.05	0.09	0.04	0.10	0.12	na	0.02	<lq	0.03	0.03	0.02
20:4 ω 6	0.03	<lq	<lq	<lq	0.02	<lq	na	<lq	<lq	<lq	0.01	0.03
20:5 ω 3	1.13	0.54	0.74	0.43	1.82	1.63	na	0.65	4.90	1.46	0.78	0.01
22:0	<lq	<lq	<lq	<lq	<lq	<lq	na	<lq	0.08	0.05	<lq	<lq
22:1 ω 9	0.02	<lq	0.02	<lq	0.02	<lq	na	<lq	0.06	<lq	0.02	<lq
22:2	<lq	<lq	<lq	<lq	<lq	<lq	na	0.06	0.09	0.05	<lq	<lq
23:0	0.02	<lq	0.03	<lq	0.05	0.05	na	0.23	0.25	<lq	0.05	<lq
24:0	<lq	<lq	<lq	<lq	0.05	<lq	na	<lq	0.14	0.08	0.03	0.03
22:6 ω 3	1.00	0.30	0.50	0.25	0.97	0.61	na	0.16	4.50	1.03	0.67	<lq
24:1	<lq	<lq	<lq	<lq	<lq	<lq	na	<lq	<lq	<lq	<lq	<lq
26:0	<lq	<lq	<lq	<lq	<lq	<lq	na	<lq	0.05	<lq	<lq	<lq
28:0	<lq	<lq	<lq	<lq	<lq	<lq	na	<lq	<lq	<lq	<lq	<lq

* nomenclature of fatty acids: a:b ω c, where a = number of carbons in the chain, b = number of unsaturation; ω c = position of the first unsaturation in the carbon chain; "i" refers to "iso" and "a" refers to "ante-iso" homologs of a particular fatty acid.
na = not analyzed; < lq = below limit of quantification ($0.01 \mu\text{g L}^{-1}$)

Table S2. Distribution of individual n-alcohols. Values in $\mu\text{g L}^{-1}$.

Station n-alcohols*	January/2014						May/2014					
	E1	E2	E3	E4	E5	E6	E1	E2	E3	E4	E5	E6
C ₁₄ -OH	<lq	<lq	<lq	<lq	<lq	0.03	0.09	na	<lq	<lq	<lq	<lq
C ₁₆ -OH	0.06	0.04	0.06	0.08	0.02	0.21	0.17	na	0.12	0.02	0.03	0.04
C ₁₈ -OH	<lq	0.19	0.33	0.37	0.13	<lq	0.49	na	<lq	<lq	<lq	0.11
C ₂₀ -OH	0.05	0.03	0.05	0.06	0.03	0.07	0.04	na	0.08	0.02	0.03	0.04
C ₂₂ -OH	0.02	0.01	0.02	0.02	0.01	0.03	0.02	na	0.08	<lq	0.02	<lq
C ₂₄ -OH	0.10	0.05	0.10	0.10	0.05	0.13	0.01	na	0.02	<lq	<lq	<lq
C ₂₆ -OH	0.02	0.01	0.02	0.02	0.01	0.04	0.01	na	0.02	<lq	<lq	<lq
C ₂₈ -OH	0.01	0.01	0.01	0.01	0.01	0.01	<lq	na	<lq	<lq	<lq	<lq
C ₃₀ -OH	0.09	0.03	0.04	0.05	0.04	0.11	0.01	na	0.01	<lq	<lq	<lq
C ₃₂ -OH	0.03	0.01	0.01	0.02	0.01	0.04	<lq	na	<lq	<lq	<lq	<lq

*nomenclature: C_n-OH, where n is the number of C in the linear chain