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Dissolved and particulate organic matter source-age characterization in the upper and lower Chesapeake Bay: A combined isotope and biochemical approach

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Abstract

In order to characterize the sources and ages of organic matter contributing to river and estuarine outflow waters, the present study investigated $\Delta^{14}C$ and $\delta^{13}C$ signatures of the major operationally defined biochemical classes of ultrafiltered dissolved organic matter (UDOM) in conjunction with lipid biomarker and elemental compositions of UDOM and suspended particulate organic matter (POM) in the Chesapeake Bay. Freshwater (Susquehanna River) UDOM was dominated by a molecularly uncharacterized (MUC) fraction, followed by total carbohydrate (TCHO), total hydrolysable amino acid (THAA) and total lipid (TLE) components. In contrast, UDOM at the bay mouth (salinity ~22–24) was comprised mainly of TCHO, followed by MUC, THAA, and TLE. The $\Delta^{14}C$ and $\delta^{13}C$ signatures of both UDOM and its major biochemical classes indicate that Susquehanna DOM is derived in part from old allochthonous terrestrial sources, whereas young marine sources dominate at the bay mouth. In contrast to the other biochemical classes, lipophilic DOM at both sites was very old (~5,000–7,000 years B.P.). In addition, factor analysis of lipid biomarker compounds revealed unique signatures for the UDOM and POM pools that imply disparate source and/or recycling properties as well as potential influences due to physical partitioning. Lipid biomarker compounds showed that although autochthonous riverine/estuarine sources dominated both the UDOM and POM pools, terrigenous lipids were elevated in the Susquehanna during high flow conditions. The presence of lipid biomarkers diagnostic of "fresh" algal material in UDOM further suggested its greater reactivity than POM. The observed biochemical and lipid biomarker compositions and isotopic signatures of UDOM and POM are consistent with previous findings suggesting that these two major organic matter pools have dissimilar reactivities and cycling times, and they derive from comparatively unique source-age materials in rivers and estuaries.

Global riverine discharge of dissolved (DOM) and particulate organic matter (POM) is on the order of ~0.4 $\times$ 10^{15} g C yr^{-1} (Meybeck 1982; Hedges 1992), making it a potentially major source of organic carbon (C) to the world's oceans. However, much of this carbon is transported through, and modified within, riverine and estuarine environments before entering the coastal ocean. Organic matter in these systems is comprised of variable amounts of identifiable biomolecules such as carbohydrates, proteins, and lipids (Mannino and Harvey 2000; Minor et al. 2001), as well as operationally defined (e.g., humic and fulvic substances in the dissolved phase) and other noncharacterizable, long-lived geomacromolecules. Radiocarbon (^{14}C) studies indicate that DOM and POM in rivers and estuaries range from modern to thousands of years in age (Hedges et al. 1986; Masiello and Druffel 2001; Raymond and Bauer 2001). Previous studies have also shown, however, that young subcomponents of estuarine DOM can be reactive and cycle on timescales of days to weeks (Guo and Santschi 1997; McCallister et al. 2004). Consequently, the short cycling time of a portion of this DOM may result in even older, more refractory forms of organic matter remaining undegraded in some rivers and estuaries (Raymond and Bauer 2001) and ultimately being exported to the coastal ocean.

Radiocarbon studies of riverine and estuarine systems have found that the average age of POM is often much older than co-occurring DOM (Masiello and Druffel 2001; Raymond and Bauer 2001). Paradoxically, the size-reactivity continuum model predicts that the bioreactivity of organic matter decreases along a continuum of size (from large to small; Amon and Benner 1996). These seemingly incongruous characteristics may be due to the complex processes governing the sources and sinks of bulk organic matter pools within riverine/estuarine systems (Hedges and Keil 1999) as well as to the unique physical and hydrological attributes of individual estuarine systems (e.g., small, mountainous river vs. coastal plain estuaries) and their associated watersheds.

Although measurement of natural ^{14}C in bulk organic matter provides an estimate of average ^{14}C age for the total pool, it does not allow for assessment of the turnover times

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and potential reactivities of organic matter subcomponents. For example, studies of molecular weight classes of DOM in estuarine environments have shown that $\Delta^{14}C$ signatures for individual size classes differ from previously published $\Delta^{14}C$ of total DOM (Guo and Santschi 1997). Furthermore, $\Delta^{14}C$ analyses of biochemical and individual molecular components of dissolved, particulate, and sedimentary organic matter often have dramatically different turnover times than bulk organic matter (Eglinton et al. 1997; Wang et al. 1998; Loh et al. 2004).

In addition to isotopic approaches, lipid biomarker compounds have been used as source-specific indicators of terrestrial and aquatic inputs and diagenetic states of DOM and POM in estuarine systems (Canuel et al. 1995; Mannino and Harvey 1999; Canuel 2001 and references therein). Because sources of estuarine organic matter are typically complex, lipid biomarker analysis can be an effective and complementary means of identifying sources of organic matter in conjunction with, for example, natural isotopic abundances, where overlapping signatures may not allow for unequivocal source resolution. In addition, the biochemical reactivity of organic matter may be assessed using lipid biomarker distributions, providing information about its potential bioavailability to heterotrophic organisms and/or heterotrophic alteration (Canuel et al. 1995; Wakeham 1995). The delineation and quantification of organic matter composition, sources, and fates in river and estuarine systems (Hedges and Keil 1999) is thus enhanced by employing multitracer approaches.

The goal of the present study was to evaluate the sources, ages, and relative reactivities of ultrafiltered DOM (UDOM) and suspended POM at two end-member sites in a large coastal plain estuary. This was achieved by measuring stable and radiocarbon isotopic signatures of bulk organic matter pools and their major dissolved biochemical classes, in conjunction with lipid biomarker and major elemental compositions of UDOM and POM. This approach further allowed for a direct comparison of the composition of the two primary physical states of organic matter along the continuum between land and the coastal ocean.

Methods

Study sites—Surface water samples (~1 m depth) were collected under high and low flow conditions from two sites in the Chesapeake Bay estuary during 2000, which was characterized by average rainfall and freshwater discharge (USGS 2002). The sites selected represent end-members within the Chesapeake system, thus allowing a comparison of the composition and potential reactivity of organic matter entering the bay at its major freshwater source relative to that exported to the Middle Atlantic Bight at the bay mouth. The freshwater station was located immediately downstream of the Conowingo Dam on the Susquehanna River (SQR, Table 1). Subsequent work on this system has indicated minimal differences in the amounts and isotopic character of organic matter collected either upstream or downstream of the dam (Bauer, unpub. data). The Susquehanna is the largest tributary of the bay, contributing about 60% of the annual freshwater input to the estuary (USGS 2002). High- and low-flow samples were collected in April and September 2000, respectively. Salinity at SQR was undetectable at both sampling times.

Samples representative of organic matter exported from the bay to the Middle Atlantic Bight were collected at the Chesapeake Bay Bridge Tunnel from the pier on Island One in the mouth of the Chesapeake Bay (CBM; Table 1). The high-flow (salinity = 22.4) and low-flow (salinity = 23.9) samples for CBM were collected in May and September 2000, respectively.

Ultrafiltered DOM—Large volume water samples (~100–150 liter) were collected using a submersible centrifugal pump (Geotech Geosquirt) with precleaned (10% HCl, Nanopure water) polyurethane tubing. Samples were filtered sequentially through precombusted (525°C for 4 h) 0.7-μm Whatman GF/F glass-fiber filters and precleaned (10% HCl, Nanopure water) 0.2-μm MSI polypropylene or Gelman Criticap capsule filters into precleaned (10% HCl, Nanopure water) 20-liter polycarbonate carboys. The GF/F filters were subsequently used for POM analyses (see below). Filtrates were transferred to an Amicon DC-10L tangential flow ultrafiltration system equipped with 3,000 d molecular-weight cutoff spiral wound cellulose filter cartridges (Amicon model S10Y3). The proportions of bulk DOM collected as UDOM using these cartridges for the various samples are given in Table 1. Samples were reduced in volume to ~1 liter and frozen until processing. For analyses, samples were thawed, diafiltered to remove salts, and lyophilized. Ultrafiltered dissolved organic carbon (C), nitrogen (N), and phosphorus (P) were measured prior to biochemical class extractions using methods as described for POM elemental analyses below. The overall analytical standard errors for the various analyses ($n = 20$ each) were 90 μg C L^{-1}, 5.6 μg N L^{-1}, and 0.31 μg P L^{-1} for ultrafiltered dissolved organic C, N, and P analyses, respectively. Detection limits were 12 μg C L^{-1} for C, 0.14 μg N L^{-1} for N, and 0.31 μg P L^{-1} for P.

UDOM biochemical class extractions and carbon isotope analyses—Lyophilized samples were extracted sequentially for TLE, THAA, and TCHO contents, and each fraction was analyzed for $\Delta^{14}C$ and $\delta^{13}C$ isotopic signatures (Loh et al. 2004). Briefly, TLE was extracted from the lyophilized sample using a modified Bligh-Dyer extraction with dichloromethane : methanol (2 : 1 v/v) aided by an accelerated solvent extractor (Dionex ASE 200). Residue from the lipid extraction was divided by weight into two portions for THAA and TCHO extraction and isotopic analyses. One portion was hydrolyzed (6 mol L^{-1} HCl at 100°C for 19 h) and eluted with 1.5 mol L^{-1} NH_4 OH through a cation exchange column to collect the THAA fraction. The other portion was hydrolyzed for the TCHO fraction (72% H_2SO_4 for 2 h, then 0.6 mol L^{-1} H_2SO_4 at 100°C for 2 h). The solution was neutralized with $Ba(OH)_2 \cdot 8H_2O$, adjusted to pH 6–7 with 1.5 mol L^{-1} NH_4 OH, and eluted with Nanopure water through a cation/anion exchange column to trap the TCHO fraction. Samples were converted to graphite (Vogel

Table 1. Hydrographic information, elemental ratios, and lipid class concentrations of UDOM and POM for Chesapeake Bay.

	UDOM				POM			
Station	SQR		CBM		SQR		CBM	
Latitude	39°39′N		36°57′N		39°39′N		36°57′N	
Longitude	76°11′W		76°02′W		76°11′W		76°02′W	
Sample*	SQR-HF	SQR-LF	CBM-HF	CBM-LF	SQR-HF	SQR-LF	CBM-HF	CBM-LF
Temperature (°C)	12.4	25.9	12.6	23.5	12.4	25.9	12.6	23.5
Salinity	0.0	0.0	22.4	23.9	0.0	0.0	22.4	23.9
Discharge ($m^3\ s^{-1}$)†	2,679	303	3,370	1,036	2,679	303	3,370	1,036
Sample volume (liter)	68	103	121	123	15.5	40.0	30.5	42.5
% bulk DOC‡	56	17	11	23				
Organic carbon ($\mu g\ C\ L^{-1}$)	1,417	279	291	367	1,336	264	682	323
N ($\mu g\ N\ L^{-1}$) §	81.4	26.8	35.7	44.0	bd	51.8	140	67.2
Organic phosphorus ($\mu g\ P\ L^{-1}$)	5.58	1.29	2.65	3.49	bd	4.21	bd	2.46
Suspended particulate inorganic P ($\mu g\ P\ L^{-1}$)					54.3	9.44	15.0	4.47
C : N‖	20	12	10	10	—	6	6	6
C : P‖	656	558	283	271	—	162	—	339
N : P‖	32	46	30	28	—	27	—	61
(ng mg organic $carbon^{-1}$)								
Total lipid	410	867	1,275	654	9,962	47,854	35,155	36,064
Fatty acids	179	453	551	450	8,213	35,604	29,052	28,387
Sterols	121	347	442	164	1,321	6,730	3,833	4,354
Alcohols	107	56	267	36	186	369	280	411
Phytol	1.98	10.7	14.9	2.86	242	5,152	1,990	2,911

* HF are samples collected during high-flow conditions, LF are samples collected during low-flow conditions.
† Discharge data for Chesapeake Bay sites were obtained from U.S. Geological Survey (2002).
‡ UDOM as percentage of bulk dissolved organic carbon (DOC) based on C recoveries after ultrafiltration, diafiltration, and lyophilization, measured after combustion to CO_2.
§ Organic nitrogen (for UDOM) and particulate nitrogen (for POM).
‖ Organic elemental ratios could not be calculated for the SQR-HF and CBM-HF samples because particulate N and organic P were both below detection limits (for CBM-HF, only organic P was not detected); bd, below detection.

et al. 1987) and $\Delta^{14}C$ analyses were performed by accelerator mass spectrometry (AMS) at the National Ocean Sciences AMS (NOSAMS) facility in Woods Hole, Massachusetts. $\delta^{13}C$ measurements were made on separate splits of the sample CO_2 using a Finnigan Delta S isotope ratio mass spectrometer. Errors ($\pm 1\sigma$) are reported as replicate analyses of either standard compounds or samples (n = 2–3), and were ±9‰ for UDOM, ±49‰ for THAA, and ±75‰ for TCHO for $\Delta^{14}C$ measurements, and ±0.9‰ for all $\delta^{13}C$ measurements. Due to sample size limitations for TLE, replicate sample extractions could not be performed. Errors for TLE $\Delta^{14}C$ measurements were thus based on AMS analytical uncertainties alone (±4–7‰).

Suspended POM—Suspended particulate organic C, particulate N, and particulate organic P (defined as material >0.7 μm nominal pore size of the GF/F filters) were measured prior to extraction for total lipid biomarker analyses (see below). For suspended particulate organic C and particulate N contents, duplicate subsamples of 14.6-mm diameter were cored from the glass fiber filters and fumed with concentrated HCl in a desiccator to remove carbonates prior to analysis using a Fisons EA1500 elemental analyzer. The suspended particulate organic P samples were oxidized using the high-temperature ashing oxidation method of Aspila et al. (1976). Overall analytical standard errors for this entire data set were 96 $\mu g\ C\ L^{-1}$ (n = 20), 11.2 $\mu g\ N\ L^{-1}$ (n = 18), and 0.31 $\mu g\ P\ L^{-1}$ (n = 16) for suspended particulate organic C, particulate N, and particulate organic P, respectively, and 2.8 $\mu g\ P\ L^{-1}$ (n = 19) for suspended particulate inorganic P analyses. Detection limits were 0.96 $\mu g\ C\ L^{-1}$ for C, 0.14 $\mu g\ N\ L^{-1}$ for N, and 0.31 $\mu g\ P\ L^{-1}$ for P.

Lipid biomarker analyses—A portion of the TLE from UDOM or POM samples was dried and saponified under a nitrogen headspace with 1 mol L^{-1} KOH (in aqueous methanol) at 110°C for 2 h. Neutral lipids were recovered by extraction into hexane at basic pH and separated into constituent fractions using 5% deactivated silica columns (Canuel and Martens 1993). The saponification residue was acidified to pH 2 and the acids extracted into hexane. The acid fraction was collected and methylated using BF_3-methanol to yield fatty-acid methyl esters. All alcohols/sterols and fatty-acid methyl esters were analyzed by gas chromatography using a flame ionization detector and/or gas chromatography–mass spectrometry as outlined in Canuel (2001). Relative analytical standard errors based on triplicate analyses of a standard fatty acid and sterol mixture were 8.2% ± 1.5% for fatty acids and 2.3% ± 1.3% for sterols. Based on previous work on a similar instrument, detection limits by flame ionization was linear

over a range of 10^7 with a minimum detection limit of 1 pg s^{-1} (Canuel et al. 1995).

Each class of lipid yielded several compounds. A subset of fatty acid and sterol biomarker compounds, selected for their source specificity and to represent potential sources, was analyzed using factor analysis (SPSS version 11) in order to examine the characteristics of the UDOM and POM samples. As this method requires that the number of dependent variables be less than the number of independent variables, fatty acid and sterol observations were analyzed separately. For each compound class, data were analyzed using relative abundances (percentage of total fatty acids or sterols). This approach also provided an opportunity to examine whether the dominant factors (i.e., sources of variance) for each of the lipid compound classes corroborated one another.

Results

Bulk elemental properties—Concentrations of C, N, and P in the DOM and POM pools generally covaried, there were no clear differences due to flow regime (Table 1). Particulate N was below detection for the high-flow SQR sample, but was measurable in all other samples (Table 1). The exclusive form of particulate P in both SQR and CBM high-flow samples was particulate inorganic P, which also comprised the majority (65–69%) of the total particulate P in low-flow samples (Table 1). Elemental ratios (C : N, C : P, and N : P) for both UDOM and POM generally exceeded Redfield, with the exception of suspended particulate organic C : N, which was similar to Redfield (Table 1).

UDOM biochemical classes and isotopic signatures—Organic C contents of the TLE, THAA, and TCHO fractions were <1%, 13–24%, and 23–50% of the ultrafiltered dissolved organic C, respectively (Table 2). The molecularly uncharacterized (MUC) fraction was determined by difference and comprised 31–64% ultrafiltered dissolved organic C (Table 2). Overall, the MUC fraction of the UDOM was 1.5- to 2-fold greater for samples collected at SQR than for the CBM samples. The biochemical composition of UDOM was similar during both high- and low-flow conditions at CBM (Table 2).

The $\delta^{13}C$ signatures of bulk UDOM, as well as the THAA and TCHO fractions, were more depleted at SQR than CBM (Table 2). The TLE fraction was the isotopically lightest of the three biochemical classes measured at CBM (Fig. 1). Comparisons of $\delta^{13}C$ signatures of the TLE fraction between the CBM and the SQR site were not possible because the entire sample was required for $\Delta^{14}C$ analysis. With the exception of the CBM low-flow sample, THAA and TCHO $\delta^{13}C$ signatures were enriched relative to bulk UDOM, whereas TLE and MUC $\delta^{13}C$ signatures were depleted (Table 2; Fig. 1). Isotopic signatures for the MUC fraction were estimated by isotopic mass balance as

$$X_{\mathrm{MUC}} = \frac{X_{\mathrm{UDOM}} - f_{\mathrm{TLE}}X_{\mathrm{TLE}} - f_{\mathrm{THAA}}X_{\mathrm{THAA}} - f_{\mathrm{TCHO}}X_{\mathrm{TCHO}}}{f_{\mathrm{MUC}}} \quad (1)$$

where X is the $\Delta^{14}C$ or $\delta^{13}C$ signature of each biochemical fraction, f is the relative contribution of each to the total UDOM pool, and $f_{\mathrm{TLE}} + f_{\mathrm{THAA}} + f_{\mathrm{TCHO}} + f_{\mathrm{MUC}} = 1.0$. Based on this calculation, the $\delta^{13}C$ signature of the MUC fraction was depleted by approximately 6‰ at SQR compared with CBM (Table 2). Errors associated with the $\delta^{13}C$ estimate for the MUC fraction will be dependent upon the $\delta^{13}C$ measurement errors of the bulk UDOM and the other fractions. The error for these measurements is given in the Methods section above.

Bulk UDOM $\Delta^{14}C$ signatures ranged from 4‰ to 108‰ (Table 2), indicating that bulk DOM contained bomb ^{14}C and was, on average, decadal in age. $\Delta^{14}C$ values for THAA were also modern, whereas values for TCHO indicated ^{14}C ages of ~800–1500 yr B.P. (Table 2). The TLE fraction was older than the corresponding THAA and TCHO fractions by about 5,000–7,000 yr (Table 2, Fig. 1). In contrast, $\Delta^{14}C$ signatures for the MUC fraction in the SQR high-flow and CBM low-flow samples were calculated by isotopic mass balance to be modern in age (Table 2). With the exception of the MUC fractions, $\Delta^{14}C$ signatures from all other biochemical classes were depleted relative to bulk UDOM.

Lipid compositions—At SQR, TLE concentrations were twofold lower for UDOM and fivefold lower for POM during high-flow compared with low-flow conditions (Table 1). Conversely, at the CBM site TLE concentrations in UDOM were twofold higher during high-flow conditions compared with low-flow conditions, and there were no flow-related differences for the POM samples (Table 1).

Total fatty acid (FA) abundances were directly related to TLE concentrations, with abundances in the POM pool generally higher than those in the UDOM pool (Table 1). Total FA comprised ~43–69% of the total lipid classes analyzed in UDOM and ~74–83% in POM. Combined total even-numbered, medium-chained saturated FA (14:0, 16:0, and 18:0) dominated both the POM and UDOM samples (Table 3). Total FA concentrations were lower in the SQR samples collected during high flow, whereas concentrations were similar regardless of flow at the CBM site (Table 1). Long-chained FA with carbon chain lengths $\geq C_{24}$ were found in all POM samples, but only in the high-flow UDOM sample from SQR (Table 3).

Polyunsaturated FA were detected in all POM and UDOM samples except for low-flow UDOM from SQR (Table 3). Polyunsaturated FA present were 18:2, 18:3, 16:2, 16:3, 20:5ω3 and 22:6ω3 (Table 3). The proportion of polyunsaturated FA associated with POM was higher during low-flow conditions at both sites (19% ± 2% vs. 6.6% ± 1.4%). With the exception of UDOM samples from SQR, branched FA were found in all UDOM and POM samples (Table 3), but their relative abundances (percentage branched FA) were greater in the POM pool (5.1% ± 2.4% vs. 0.6% ± 0.6%). Odd- (iso and anteiso C_{13}, C_{15}, C_{17}) and even-numbered (iso C_{14}, C_{16}) branched FA were found in all POM samples, whereas only odd-numbered branched FA were found in UDOM samples from CBM (Table 3).

Table 2. Isotopic signatures of dissolved and particulate organic matter pools and biochemical fractions in UDOM from the Chesapeake Bay.

Sample*	Bulk DOM†		LMW DOM‡		UDOM (HMW)		POM§		TLE			THAA			TCHO			MUC		
	$\Delta^{14}C$	$\delta^{13}C$	$\Delta^{14}C$	$\delta^{13}C$	$\Delta^{14}C$	$\delta^{13}C$	$\Delta^{14}C$	$\delta^{13}C$	‖%	$\Delta^{14}C$	$\delta^{13}C$	‖%	$\Delta^{14}C$	$\delta^{13}C$	‖%	$\Delta^{14}C$	$\delta^{13}C$	¶%	$\Delta^{14}C$#	$\delta^{13}C$#
SQR-HF	–81 (679)††	–27.0	–326 (3,172)	–26.4	108 (Modern)	–27.5	–61 (506)	–25.6	0.19	–487 (5,354)	–28**	13.4	16 (Modern)	–25.0	22.5	–91 (762)	–23.9	63.9	199 (Modern)	–29.3
SQR-LF	–81 (679)	–27.0	–91 (764)	–27.1	4 (Modern)	–26.0	na	–28.0	0.46	Lost	Lost	16.5	Lost	–25.1	35.0	–167 (1,468)	–24.3	48.0	—	–27.4
CBM-HF	–77 (644)	–23.7	–103 (877)	–24.1	41 (Modern)	–21.8	60 (Modern)	–23.0	0.68	Lost	–27.7	24.0	25 (Modern)	–20.8	44.8	3 (Modern)	–21.0	30.6	—	–23.4
CBM-LF	–29 (236)	–23.7	–48 (398)	–24.3	37 (Modern)	–21.5	18 (Modern)	–21.4	0.44	–575 (6,866)	–28.3	18.8	39 (Modern)	–21.2	50.0	17 (Modern)	–21.8	30.7	78 (Modern)	–21.1

* HF, samples collected during high flow; LF, samples collected during low-flow conditions.
† Bulk DOM $\Delta^{14}C$ values from Raymond and Bauer (2001, for SQR) and Bauer et al. (2002, for CBM). $\delta^{13}C$ values from Bauer (unpub. data). All isotope values are reported in ‰.
‡ Isotopic signatures of low molecular weight (LMW) DOM, estimated by isotopic mass balance as $X_{LMW} = (X_{DOM} - f_{UDOM}X_{UDOM})/(1 - f_{UDOM})$, where X is the $\Delta^{14}C$ or $\delta^{13}C$ value of each molecular weight fraction, f is the relative contribution of UDOM (high molecular weight [HMW]) and LMW material to DOM, and $f_{UDOM} + f_{LMW} = 1.0$.
§ POM $\Delta^{14}C$ values from Bauer (unpub. data) and Bauer et al. (2002, for CBM). POM $\delta^{13}C$ values from Canuel (unpub. data).
‖ % biochemical fraction in UDOM, based on carbon equivalents measured after combustion to CO_2.
¶ %MUC = 100 – (%TLE + %THAA + %TCHO).
# Isotopic signatures of MUC were estimated by isotopic mass balance (see text).
** Assumed from average of all observed values for purposes of inclusion of $\Delta^{14}C$-$\delta^{13}C$ pair in Fig. 1 due to inadequate mass of sample for isotopic analysis.
†† Values in parentheses are ^{14}C ages in years B.P., calculated from $\Delta^{14}C$ values as Age (in years B.P.) = –8033 ln $(1 + \Delta^{14}C/1000)$, where years B.P. indicate years before 1950, prior to thermonuclear weapons testing (Stuiver and Polach 1977). Since $\Delta^{14}C$ values were not reservoir-corrected, all ages are not true calendar ages.

Fig. 1. $\Delta^{14}C$ versus $\delta^{13}C$ signatures for bulk UDOM and biochemical classes in Susquehanna River (SQR) and Chesapeake Bay mouth (CBM) waters during low flow (LF) and high flow (HF) conditions (samples from the same site are grouped in circles). TLE, total lipid extract; THAA, total hydrolyzable amino acids; TCHO, total carbohydrates; MUC, molecularly uncharacterized component. Ages are expressed as ^{14}C years B.P. and calculated as in Table 2.

Sterols comprised the majority of the neutral lipids analyzed in all Chesapeake Bay UDOM and POM. At SQR, total sterol concentrations associated with POM were fivefold lower during high-flow, whereas there were little or no flow-related differences at the CBM site or for the UDOM pool at both sites (Table 1). Sterol distributions in both UDOM and POM samples (Table 4) were dominated by cholest-5-en-3β-ol (cholesterol; $C_{27}\Delta^5$), and 24-ethyl-cholest-5-en-3β-ol ($C_{29}\Delta^5$). Small amounts of 5α(H)-stanols such as cholestanol [5α(H)-cholestan-3β-ol] and stigmastanol [24-ethyl-5α(H)-cholestan-3β-ol] and trace amounts of 4-methyl stanols [4α,23S,24R-trimethyl-5α(H)-cholestan-3β-ol and 4α,23R,24R-trimethyl-5α(H)-cholestan-3β-ol] were also detected (Table 4). Phytol abundances associated with POM increased 1.5- to 20-fold from high-flow periods to low-flow periods. In contrast, only trace amounts of phytol were found in UDOM samples (Table 1).

Using factor analysis, the lipid biomarker data set was described by two factors, which together explained 80–83.5% of the variance. Factor 1 accounted for 60–62.5% of the variability in the data set, whereas Factor 2 accounted for 20–21% of the variability. For fatty acids, loadings for Factor 1 were most positive for odd-numbered branched FA (iso and anteiso C_{13}, C_{15}, C_{17}) and most negative for odd-numbered (C_{13}, C_{15}, C_{17}) saturated FA, 18:0, and 16:0 fatty acids (Fig. 2a). All POM samples had positive scores for Factor 1, whereas all UDOM samples had negative scores (Fig. 2b). Factor 2 loadings were most positive for long-chained FA and most negative for 22:6ω3 (Fig. 2a). With the exception of the low-flow POM sample from SQR, all SQR samples had positive scores for Factor 2, whereas all CBM samples had negative scores (Fig. 2b).

For sterols, Factor 1 loadings were most positive for $C_{27}\Delta^5$, $C_{29}\Delta^5$, and $C_{29}\Delta^{5,22}$, and most negative for $C_{28}\Delta^{5,22}$, $C_{27}\Delta^{5,22}$, and $C_{28}\Delta^{5,24(28)}$ (Fig. 3a). Similar to Factor 2 for the fatty acid data set (see above), with the exception of the low-flow POM sample from SQR, all SQR samples had

Table 3. Relative fatty acid distributions and fatty acid class concentrations for UDOM and POM from Chesapeake Bay.

	UDOM				POM			
Sample*	SQR-HF	SQR-LF	CBM-HF	CBM-LF	SQR-HF	SQR-LF	CBM-HF	CBM-LF
(% total FA)								
12:0	Bd	bd	bd	bd	0.3	0.1	0.3	0.4
br13,15,17†	Bd	bd	1.0	1.2	7.4	3.6	2.6	4.3
br14,16†	Bd	bd	bd	bd	1.1	0.5	0.3	0.6
n-13,15,17‡	4.4	5.2	4.5	4.1	2.2	1.4	1.8	2.2
14:0	2.3	2.3	6.1	4.3	4.3	6.3	14.9	10.1
16:2 + 16:3	Bd	bd	bd	bd	1.3	7.6	1.6	1.5
16:1ω7	7.0	5.2	15.6	11.5	19.6	25.3	20.9	23.9
16:0	25.7	36.7	34.5	31.0	19.3	22.3	30.1	21.9
18:2 + 18:3	bd	bd	3.9	2.2	3.2	2.7	2.6	3.1
18:1ω9	15.8	14.6	9.5	13.7	15.2	11.7	14.9	13.7
18:0	27.4	27.1	17.7	24.1	9.2	2.4	2.8	2.5
20:5ω3	10.1	bd	bd	bd	0.6	7.7	2.0	6.6
22:6ω3	bd	bd	bd	0.6	bd	1.9	1.3	4.8
22:1	4.2	9.0	4.4	3.5	bd	0.5	0.5	0.4
LCFA§	1.5	bd	bd	bd	10.3	2.1	1.2	0.9
(ng mg OC^{-1})								
n-FA‡	113	323	346	286	4,006	12,536	14,947	10,928
MUFA‖	48.4	130	174	139	3,087	14,118	10,922	11,312
PUFA¶	18.1	bd	25.5	19.7	423	7,504	2,334	4,775
brFA†	bd	bd	5.72	5.36	697	1,446	850	1,371
TAR_{FA}#	0.052	0.000	0.000	0.000	0.327	0.067	0.024	0.022

* HF, samples collected during high flow; LF, samples collected during low-flow conditions.
† Branched saturated fatty acids include iso- and anteiso-methyl branched carboxylic acids.
‡ *n*-fatty acids are all straight-chained, saturated carboxylic acids.
§ Long-chained fatty acids (LCFA) include all saturated carboxylic acids with 24 or greater carbon chain lengths.
‖ Monounsaturated fatty acids.
¶ Polyunsaturated fatty acids include all carboxylic acids with two or more double bonds.
# Terrestrial to aquatic ratio for straight-chained saturated fatty acids (see text for equation); bd, below detection.

Table 4. Relative sterol distributions and total sterol concentrations for UDOM and POM from Chesapeake Bay.

	UDOM				POM			
Sample*	SQR-HF	SQR-LF	CBM-HF	CBM-LF	SQR-HF	SQR-LF	CBM-HF	CBM-LF
(% total sterols)								
$C_{26}\Delta^{5,22}$†	bd	bd	7.2	0.7	5.1	1.0	7.0	9.1
$C_{27}\Delta^{5,22}$	3.2	8.9	8.0	11.2	3.7	7.2	10.4	10.6
$C_{27}\Delta^{5}$	34.6	28.9	14.1	19.6	26.7	22.4	17.6	15.6
$C_{28}\Delta^{5,22}$	5.2	12.5	20.9	22.0	5.4	18.4	16.8	16.9
$C_{28}\Delta^{5,24(28)}$	bd	6.9	9.2	15.1	5.4	20.1	5.7	12.9
$C_{28}\Delta^{5}$	10.7	5.1	1.3	bd	5.8	6.3	13.1	1.5
$C_{29}\Delta^{5,22}$	9.2	8.4	8.1	7.1	6.2	4.3	4.5	3.1
$C_{29}\Delta^{5}$	28.3	25.4	28.0	17.0	24.5	15.8	8.9	9.0
4Me sterols‡	bd	bd	1.9	4.9	bd	0.7	3.8	2.3
5α(H)-stanols§	8.9	3.9	1.2	2.4	8.6	2.3	9.4	14.0
(ng mg OC^{-1})								
Autochthonous‖	52.1	199	271	121	540	3,462	1,703	2,199
Allochthonous¶	58.6	135	166	39.5	483	1,777	1,017	596
5α(H)-stanols	10.8	13.5	5.50	4.02	113	154	360	610
Alloch : autoch ratio#	1.12	0.681	0.612	0.327	0.894	0.513	0.597	0.271

* HF, samples collected during high flow; LF, samples collected during low-flow conditions.
† The nomenclature is $C_x\Delta^y$, where x is the total number of carbon atoms and y the positions of the double bonds. See text for common names used.
‡ 4-methyl sterols include 4α,23,24-trimethylcholest-22-en-3β-ol (dinosterol), 4α,23S,24R-trimethyl-5α(H)-cholestan-3β-ol, and 4α,23R,24R-trimethyl-5α(H)-cholestan-3β-ol.
§ 5α(H)-stanols include 5α-cholestan-3β-ol (cholestanol), and 24-ethyl-5α(H)-cholestan-3β-ol (stigmastanol).
‖ Autochthonous sterols include $C_{26}\Delta^{5,22}$, $C_{27}\Delta^{5,22}$, $C_{27}\Delta^{5}$ (cholesterol), $C_{28}\Delta^{5,22}$ (brassicasterol), $C_{28}\Delta^{5,22(28)}$ (24-Methylenecholesterol), dinosterol, 4α,23S,24R-trimethyl-5α(H)-cholestan-3β-ol, and 4α,23R,24R-trimethyl-5α(H)-cholestan-3β-ol.
¶ Allochthonous sterols include $C_{28}\Delta^{5}$ (campesterol), $C_{29}\Delta^{5,22}$ (stigmasterol), and $C_{29}\Delta^{5}$.
# Ratio of allochthonous to autochthonous sterols (see text for explanation); bd, below detection.

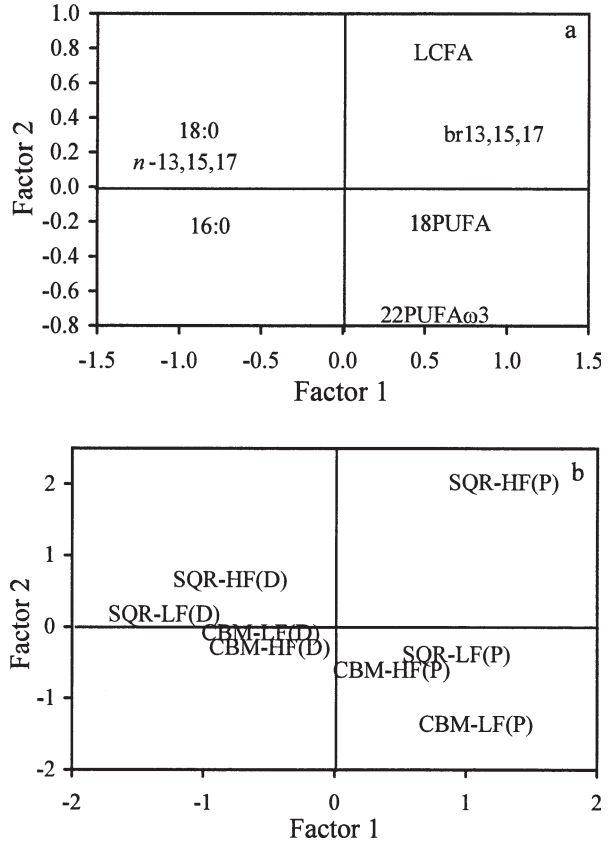

Fig. 2. Factor analysis (a) loadings and (b) score plots for Factors 1 and 2 using fatty acid abundances of UDOM and POM from the Susquehanna River (SQR) and Chesapeake Bay mouth (CBM) during low flow (LF) and high flow (HF) conditions. See text and Table 3 for fatty acid identifications in panel (a). Samples with "(D)" and "(P)" at the end of the label indicate UDOM and POM samples, respectively. Factor 1 accounted for 62.5% of the variability in the data set, whereas Factor 2 accounted for 20.4% of the variability.

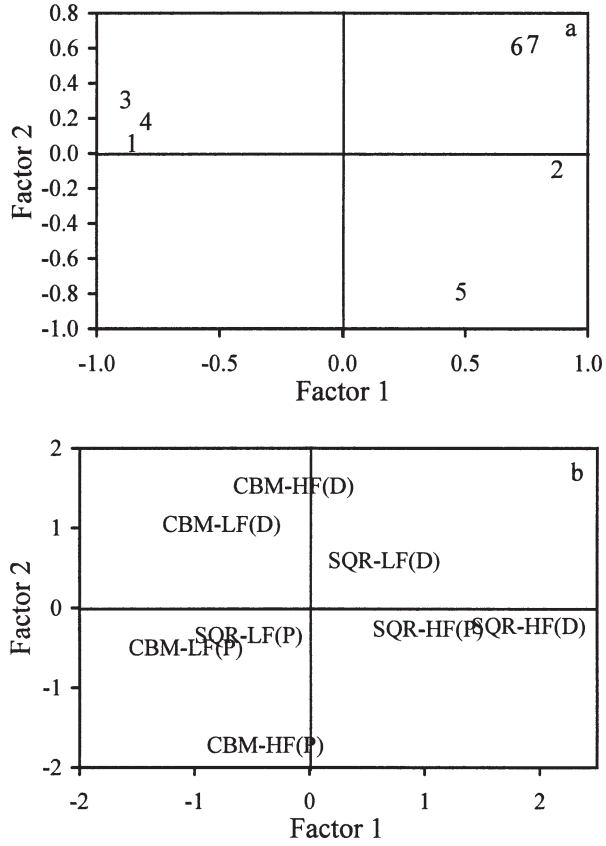

Fig. 3. Factor analysis (a) loadings and (b) score plots for Factors 1 and 2 using sterol abundances of UDOM and POM from the Susquehanna River (SQR) and Chesapeake Bay mouth (CBM) during low flow (LF) and high flow (HF) conditions. Sterol compound identifications in panel (a) are 1-$C_{27}\Delta^{5,22}$, 2-$C_{27}\Delta^{5}$, 3-$C_{28}\Delta^{5,22}$, 4-$C_{28}\Delta^{5,24(28)}$, 5-$C_{28}\Delta^{5}$, 6-$C_{29}\Delta^{5,22}$, 7-$C_{29}\Delta^{5}$. Samples with "(D)" and "(P)" at the end of the label indicate UDOM and POM samples, respectively. Factor 1 accounted for 60.1% of the variability in the data set, whereas Factor 2 accounted for 21% of the variability.

positive scores for Factor 1, whereas all CBM samples had negative scores (Fig. 3b). Factor 2 loadings were most positive for $C_{29}\Delta^{5}$ and $C_{29}\Delta^{5,22}$ and most negative for $C_{28}\Delta^{5}$ (Fig. 3a). Factor 2 scores were most positive for the UDOM samples from CBM and most negative for the high-flow POM sample from CBM (Fig. 3b). As a whole, factor analysis results indicate there are distinct compositional differences in the lipid biomarker compounds associated with the UDOM and POM samples. In addition, the magnitude of these differences varies as a function of location in the estuary and flow regime.

Discussion

Isotopic character of UDOM and dissolved biochemical classes—In addition to the influence of starting inorganic carbon source ($\delta^{13}C_{DIC}$) on δ^{13}C-organic matter signatures, the observed deviations in δ^{13}C of the TLE, THAA, and TCHO fractions from bulk UDOM δ^{13}C signatures (Fig. 1) are in part determined by kinetic fractionations at the cellular level during production of these biochemical classes (Schouten et al. 1998). As a result, the isotopic compositions of compound classes, or even specific compounds, may not be indicative of a discrete precursor "source" to each of the individual biochemical classes. Lipids, for example, are generally depleted in ^{13}C relative to protein and carbohydrate fractions (Schouten et al. 1998). Consistent with these predictions, ^{13}C signatures for the total lipid fractions were depleted. We also found that δ^{13}C signatures of TCHO at SQR were not as depleted as bulk UDOM (Fig. 1), suggesting that this fraction may have a relatively greater contribution from phytoplankton or riverine algae (del Giorgio and France 1996).

Using Δ^{14}C in conjunction with δ^{13}C may allow allochthonous and autochthonous sources of organic matter to rivers and estuaries to be more fully differentiated as a result of the much greater dynamic range of natural Δ^{14}C (–1000 to ~200‰) versus δ^{13}C (approximately –32 to –10‰). At SQR, bulk UDOM was depleted in ^{13}C but enriched in ^{14}C (Table 2; Fig. 1) and had C : N ratios of 12–20 (Table 1). These signatures are consistent with DOM

from soil leachates that have been found in previous studies to be depleted in ^{13}C but enriched in ^{14}C (Trumbore et al. 1992) and to have C : N ratios of ~15 (Schlesinger 1997). In addition, during high-flow conditions, increased water column turbidity may limit the growth of microalgae in this region of the bay, thereby reducing the potential contribution of freshwater phytoplankton as a primary source of organic matter. However, although average isotopic signatures for the bulk organic matter suggest that allochthonous sources are important at SQR, certain subcomponents of the bulk pool (e.g., TCHO) may be influenced to a relatively greater extent by algal contributions.

The $\Delta^{14}C$ values of various biochemical classes in this study also showed a greater range than the corresponding $\delta^{13}C$ signatures (Fig. 1), especially for the Susquehanna River samples. In addition, with the exception of the TLE fractions, river organic matter fractions displayed a much broader overall range in both $\Delta^{14}C$ and $\delta^{13}C$ than bay mouth fractions. Thus, the factors responsible for the large isotopic ranges both within and among biochemical classes in the river samples appear to be much more complex and variable than those near the point of estuarine discharge where values display greater coherence (Fig. 1).

Lipid biomarker indicators of UDOM and POM sources and reactivity—Lipid biomarker analyses have been shown to be an effective tool for examining complex sources of organic matter characteristic of estuarine and coastal systems (Canuel et al. 1995; Mannino and Harvey 1999; Canuel 2001). Similar to findings from previous studies, autochthonous (algal/plankton) sources dominate both DOM and POM to the Chesapeake Bay (Harvey and Johnston 1995; Canuel 2001). Fresh planktonic organic matter is an important source of material to the river-bay system as indicated by the presence of polyunsaturated FA (comprising as much as 19% ± 3% of the total FA in low-flow POM samples; Table 3) and by the most negative loadings in Factor 2 for 22:6ω3 (Fig. 2a).

Factor analysis reveals unique signatures for the UDOM and POM fractions (Figs. 2 and 3). Algal biomarkers such as C_{16} and C_{20} fatty acids (16:1ω7, 16:2, 16:3, and 20:5ω3) and $C_{28}\Delta^{5,22}$ and $C_{28}\Delta^{5,24(28)}$ sterols (Canuel and Martens 1993;Volkman et al. 1998 and references therein) were most abundant in the low-flow POM sample from SQR (Tables 3 and 4), suggesting that riverine sources of algal material may assume greater importance during low-flow regimes, with diatoms likely the predominant group (Table 4). Although diatoms generally dominate during spring phytoplankton blooms in the bay, a shift from diatoms to cyanobacteria and microflagellates occurs as the season progresses (Glibert et al. 1995). Other biomarkers generally attributed to phytoplankton, bacteria, and cyanobacteria such as 16:1ω7 and 18:1ω9 (Table 3), and to crustacean zooplankton such as cholesterol (Table 4), were also found in all bay UDOM and POM samples. In addition, phytol, a product of the alkaline hydrolysis of chlorophyll, comprised a substantial portion of the neutral lipids associated with POM (Table 1). Phytol abundances suggest that algal material is relatively more important as a direct source of POM compared with UDOM.

Long-chained FA are important constituents of higher plants and have been found previously in river and estuarine organic matter (Mannino and Harvey 1999; Canuel 2001). These compounds are the dominant FA in terrigenous vascular plants, aquatic plants such as seagrasses, and can occur at trace levels in some algae (Canuel and Martens 1993 and references therein). In this study, long-chained FA had the most positive loading on Factor 2 (Fig. 2a) and were found in all SPOM samples, but only in the high-flow UDOM sample from SQR (Table 3). These long-chain FA impart a unique terrigenous signature to the SQR POM samples (Fig. 2b), consistent with their depleted $\delta^{13}C$ signatures. Additionally, up to 100% of the P associated with high-flow SQR particles (Table 1) is inorganic, further supporting the dominance of terrigenous delivery to this site during high flow. Higher abundances of long-chained FA in the POM samples may result from the dominance of vascular plant sources to the particulate phase as well as physical factors. The relative insolubility of these compounds likely contributes to their absence (or low concentration) in the UDOM.

The terrestrial to aquatic ratio for saturated FA (TAR_{FA}) has been commonly used to indicate the relative importance of terrigenous (higher plant) to aquatic (algae and bacteria) sources (Meyers 1997). This ratio is calculated as

$$TAR_{FA} = \frac{C_{24} + C_{26} + C_{28}}{C_{12} + C_{14} + C_{16}} \quad (2)$$

where C_{12}–C_{28} are saturated fatty acids with carbon chain lengths of 12–28. In all instances the ratio was well below 1, indicating the predominance of aquatic-derived organic matter throughout the system (Table 3). The highest TAR_{FA} ratios (i.e., greater relative terrestrial plant contributions) were found at SQR during high flow in the POM pool (Table 3).

Similar to TAR_{FA}, ratios of allochthonous (i.e., plant) to autochthonous (i.e., algal/plankton) sterols (Alloch : Autoch ratio) were also greatest in high-flow SQR samples (Table 4), indicating the greater relative importance of allochthonous sources of organic matter at the SQR site during high flow. This is further supported by the factor analysis results showing unique signatures for high-flow SQR samples and SQR UDOM collected during low flow (Fig. 3b). However, with the exception of the high-flow UDOM sample from SQR, the Alloch : Autoch ratios indicate that autochthonous sources of DOM and POM predominate under high-flow conditions (Alloch : Autoch <1; Table 4). In addition, the consistently greater percentage of allochthonous higher plant sterols (e.g., campesterol, stigmasterol, and $C_{29}\Delta^5$; Table 4) in the UDOM versus the corresponding POM (high-flow UDOM = 42.9 [± 8] vs. high-flow POM = 31.5 [± 7] and low-flow UDOM = 31.5 [± 11] vs. POM = 20 [± 9]) may be due to partitioning of terrigenous organic matter on the basis of physical (e.g., dissolved vs. sorbed particulate) state. Thus, source and physical factors (e.g., solubility) must be

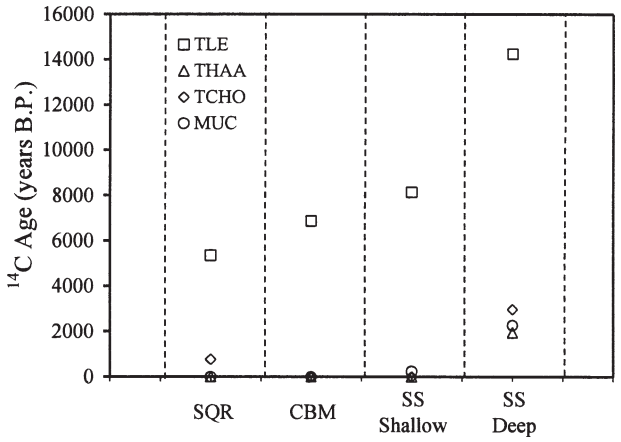

Fig. 4. Radiocarbon age distributions of major biochemical compound classes in UDOM in Susquehanna River (SQR), Chesapeake Bay mouth (CBM), and Sargasso Sea (SS) surface (~3 m depth) and deep (~1,500 m depth) waters. Multiple points for the same symbol at a given site represent the minimum and maximum ages observed for that component at that location. All four classes (TLE, THAA, TCHO, and MUC) were measured at each site, but may not be visible because of overlap along abscissa where multiple classes have co-occurring modern ages. Sargasso Sea data from Loh et al. (2004).

considered when interpreting the composition of dissolved and particulate fractions (Aufdenkampe et al. 2001).

In addition to providing information on aquatic vs. terrigenous sources, lipid biomarkers also serve as a tool for assessing heterotrophic transformations of organic matter (Wakeham 1995). Contributions of bacterial biomass, and by inference the degree to which samples may have been modified by heterotrophic processes, are reflected in the observed enrichments of branched FA (Table 3). Both odd- and even-numbered branched FA were detected in the POM (Table 3), whereas only odd-branched FA were detected in the UDOM, suggesting that unique bacterial communities may be associated with each of the two major organic matter pools (Kaneda 1991). The Δ^{14}C signatures at CBM indicate that the majority of bulk UDOM is modern, and thus recently produced (Fig. 1), similar to earlier findings of total DOM (Raymond and Bauer 2001) and UDOM ^{14}C ages in the bay (Guo and Santschi 1997). However, the presence of bacterial fatty acids (Table 3) and C : N : P ratios of bulk UDOM greater than Redfield (Table 1) suggest that the presumably reactive (i.e., due to its young mean age) UDOM at this site is reworked to some degree.

River-estuarine-oceanic age distributions of dissolved biochemical classes—The currently available information on radiocarbon ages of the major biochemical classes (TLE, TCHO, THAA, and MUC) in UDOM across the river-estuary-oceanic continuum reveals several compelling features (Fig. 4). First, the lipid component of UDOM is by far the most highly aged biochemical class in each system, which is consistent with the presumed hydrophobic, surface-active nature of this material. Similar δ^{13}C (~−28‰) for the TLE fractions measured at both sites (Loh et al. 2004; Table 2 this study) may suggest a possible common source (e.g., highly aged natural organic matter or anthropogenic petroleum hydrocarbons) for this long-lived material, or similar processes acting to preserve the TLE component (e.g., sorptive preservation of the hydrophobic fraction), across different environments. A maximum potential contribution of petroleum carbon may be calculated by assuming that the TLE is comprised exclusively of modern and ^{14}C-dead (i.e., petroleum) fractions. This simplistic isotope mass balance calculation indicates that the contribution from petroleum sources to the TLE fraction is at most ~60%, which translates to a contribution of <1% to the bulk UDOM. Realistically, the petroleum contribution is probably much lower considering that it is highly unlikely that all of the aged C in the TLE fraction is (1) comprised entirely of petroleum and (2) completely ^{14}C-dead (i.e., there is likely to be a spectrum of ages across different organic components). Indeed, our lipid biomarker analyses support this conjecture by showing a spectrum of compounds reflecting algal, microbial, and terrigenous sources, which are likely to have different associated ages. It is also noteworthy that in both river/estuarine UDOM (Tables 3 and 4) as well as in oceanic UDOM (Loh 2002; Loh et al., unpub. data), the TLE fraction contains a number of compounds that are diagnostic of inputs of relatively labile cellular components. Thus, the highly aged lipophilic fraction of UDOM likely contains a mixture of individual compounds having a broad spectrum of ages (Eglinton et al. 1997). An alternative interpretation is that lipid biomarker compounds traditionally used for distinguishing organic matter reactivity and/or diagenetic state may paradoxically be preserved for much longer times in dissolved compared with other (e.g., sedimentary or particulate) phases. This seems unlikely given the hydrophobic nature of lipids and the susceptibility of UDOM to photochemical degradation (Mopper and Kieber 2002 and references therein).

Along the river-oceanic continuum, there are only two other instances aside from the TLE in which any of the compound classes show significant (i.e., non-zero) radiocarbon ages: (1) the TCHO component of SQR UDOM, and (2) all of the biochemically definable components (i.e., TCHO, THAA, and MUC) in deep Sargasso Sea UDOM (Fig. 4). With respect to the TCHO component, SQR TCHO is more highly aged than either SQR THAA or MUC, and only somewhat younger than deep Sargasso Sea TCHO. This may be interpreted to signify the greater aging and concomitant slower recycling of TCHO (as determined from the ^{14}C age) compared with either THAA or MUC in the Susquehanna River watershed. Also, paradoxically, the TCHO component has an overall modern age in both of the more seaward environments (i.e., in estuarine [CBM] and surface ocean waters [Fig. 4]), suggesting potential contributions from autochthonous production.

Results from this study also contribute possible insights regarding between-system differences in the MUC fraction. It is important to note that MUC is operationally defined and its composition is often determined by difference. The dissimilarity in both TLE versus MUC and TCHO versus MUC ages in SQR UDOM suggests that MUC is more likely to have a proteinaceous precursor in SQR. This

contrasts with oceanic waters where neither THAA nor TCHO contributions to MUC can be excluded (Fig. 4; Loh et al. 2004), although TLE contributions to oceanic MUC are similarly minimal. However, the MUC fraction in riverine and estuarine environments may have unique sources (and therefore potentially different ^{14}C ages) at different locations and times (e.g., high- vs. low-flow periods, seasons, etc.). Nevertheless, this study gives us a first approximation of what this uncharacterized fraction may be comprised of in a river-estuary system. These data collectively suggest potential differences in the diagenetic processes responsible for aging and preservation of dissolved versus particulate biochemicals that further complicate interpretation of such data solely on the basis of general organic matter sources (e.g., terrestrial vs. marine).

Comparison of DOM and POM chemical and isotopic characteristics—Consistent with our findings, differences in the composition of the DOM and POM pools of river and estuarine waters have also been documented in previously published studies (Mannino and Harvey 1999; Minor et al. 2001; Aufdenkampe et al. 2001). One of the major differences identified in this study lies in the lipid biomarker signatures of the DOM and POM pools as revealed by factor analysis of the individual fatty acid (Fig. 2) and sterol (Fig. 3) compounds. Results indicate that Factor 1 in the fatty acid factor analysis (Fig. 2b) differentiates between DOM and POM samples. This is similarly shown in the sterol data set, where Factor 2 also differentiates between DOM and POM samples (Fig. 3b). These results indicate there are compositional differences in the lipid biomarker compounds associated with the DOM and POM samples. These compositional differences may reflect distinct sources and/or differences in the degradative state of these pools. Isotopic signatures ($\delta^{13}C$) for UDOM and POM are similar, but distinct (~2‰ difference), at SQR, and under high-flow conditions at CBM (1.2‰ difference), further supporting source/compositional differences between UDOM and POM. Additionally, during high-flow regimes, POM in the Susquehanna River exhibited a greater influence of terrigenous material as indicated by the dominance of inorganic P in suspended particles (Table 1); presence of long-chained FA and both odd- and even-numbered branched FA (Table 3); and comparatively higher TAR_{FA} (Table 3) and Alloch : Autoch sterol ratios (Table 4).

In addition, diagenetic differences also exist between the DOM and POM pools. The particulate pool appears to have undergone a lower degree of recycling than the dissolved pool, as suggested by lower C : N, C : P, and N : P ratios (Table 1), and higher proportions of both polyunsaturated and branched FA (Table 3). Finally, the isotopic composition of dissolved biochemical classes suggests that the precursor of the MUC fraction in the DOM pool in this system is likely to be proteinaceous in nature (see above) compared with open ocean where both TCHO and THAA contributions cannot be discounted due to similar ^{14}C ages between these fractions and the MUC fraction (Loh et al. 2004). This, however, contrasts with findings where the MUC fraction of the oceanic POM pool has been interpreted as having a lipid origin due to the similar ^{14}C ages of these two fractions (Hwang and Druffel 2003). By extension, it may be hypothesized that similar fundamental diagenetic processes responsible for aging and preservation of dissolved versus particulate biochemicals may be operative in both river-estuarine and oceanic settings.

The simultaneous application of isotopic and biochemical techniques represents a powerful and evolving approach for delineating the sources, source-ages, and potential reactivities of different components of the DOM and POM pools in marine and freshwater systems. The ability to better characterize and more accurately quantify the sources and sinks of a given organic pool increases as a function of the number of parameters employed, provided the primary end-member sources and sinks can be identified and adequately constrained. The present study illustrates one such multiparameter approach that facilitates greater insight into the fate of river and estuarine organic matter across a spectrum ranging from bulk materials to general biochemical classes to highly specific organic biomarker compounds. Future studies should benefit from evolving analytical capabilities in natural abundance isotopic and organic separation chemistry in order to more fully describe the cycling of autochthonous and allochthonous forms of organic matter and their role in aquatic carbon cycles.

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