Chemical characteristics of dissolved organic matter in an oligotrophic subtropical wetland/estuarine ecosystem

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Abstract

Fluorescence properties of whole water samples and molecular characteristics of ultrafiltrated dissolved organic matter (UDOM > 1,000 D) such as lignin phenol and neutral sugar compositions and ^{13}C nuclear magnetic resonance (NMR) spectra were determined along a freshwater to marine gradient in Everglades National Park. Furthermore, UDOM samples were categorized by hierarchical cluster analysis based on their pyrolysis gas chromatography/mass spectrometry products. Fluorescence properties suggest that autochthonous DOM leached/exuded from biomass is quantitatively important in this system. ^{13}C NMR spectra showed that UDOM from the oligotrophic Taylor Slough (TS) and Florida Bay (FB) ecosystems has low aromatic C (13% ± 3% for TS; 2% ± 2% for FB) and very high *O*-alkyl C (54% ± 4% for TS; 75% ± 4% for FB) concentrations. High *O*-alkyl C concentrations in FB suggest seagrass/phytoplankton communities as dominant sources of UDOM. The amount of neutral sugars was not appreciably different between the TS and FB sites (115 ± 12 mg C g C^{-1} UDOM) but their concentrations suggest a low level of diagenesis and high production rates of this material in this oligotrophic environment. Total yield of lignin phenols (vanillyl + syringyl phenols) in TS was low (0.20–0.39 mg 100 mg C^{-1} UDOM) compared with other riverine environments and even lower in FB (0.04–0.07 mg 100 mg C^{-1} UDOM) and could be a result of photodegradation and/or dilution by other autochthonous DOM. The high *O*-alkyl and low aromatic nature of this UDOM suggests significant biogenic inputs (as compared with soils) and limited bioavailability in this ecosystem.

The Everglades, located in the southern part of the Florida Peninsula, is a unique oligotrophic ecosystem and is one of the largest subtropical wetlands in the world. The most crucial factor that makes the Southern Everglades (Everglades National Park; ENP) so unique is the limited source of nutrients (particularly phosphorus), which depends mostly on dry and wet depositions, while most other wetland and estuarine systems depend on riverine nutrient inputs (Davis 1994). In addition, because the base rock of the Florida Peninsula is calcareous, there is almost no supply of phosphorous from its weathering products (Noe et al. 2001). As a result, microbial activities in ENP are controlled by the limited availability of nutrients (Amador and Jones 1993; Fourqurean et al. 1993).

Most phosphorous (P) in the Everglades is in the organic form (Noe et al. 2001; Qualls and Richardson 2003) and, thus the dissolved organic matter (DOM) in the Everglades plays pivotal roles for nutrient cycling and fueling the microbial loop. Furthermore, DOM influences biogeochemical processes through light attenuation, pH buffering, and complexation of metals and organics. However, still very little is known about the sources, composition, bioavailability, and environmental fate of DOM in the Florida coastal Everglades and similar ecosystems in tropical and subtropical environments. To better understand the biogeochemical cycles in such systems, research efforts in the Everglades have focused on estimating the sources and dynamics of nutrients (e.g., Noe et al. 2001; Qualls and Richardson 2003). However, these studies have focused on the quantitative aspects of bulk organic nutrients, and detailed chemical characteristics of DOM in this oligotrophic wetland have mostly been ignored. Lu et al. (2003) investigated the chemical characteristics of DOM collected from a transect leading from a drainage canal throughout the freshwater portion of the Everglades Panhandle. They suggested that, in addition to canal-derived allochthonous DOM, autochthonous, freshwater marsh-derived DOM, such as carbohydrates, play an important role as a source of organic nutrients in this system.

The characterization of DOM in aquatic environments by

means other than optical methods requires the concentration/ purification of this complex mixture of organic compounds. Solid-phase extraction using adsorbents such as XAD-8 (or DAX-8) and insoluble polyvinylpyrrolidone (PVP) has been traditionally applied for the collection of the DOM fraction. However, the fractionated DOM, so-called humic substances, tends to emphasize hydrophobic aspects of DOM (Benner 2003). An alternative technique, using tangential flow (or cross-flow) ultrafiltration, enables the collection of the DOM fraction with a molecular weight >1 kDa, basically without any fractionation. Although this technique leaves the low molecular-weight DOM fraction uncharacterized, the chemical characterization of the ecologically important ultrafiltrated dissolved organic matter (UDOM) fraction has made significant contributions to the field of biogeochemistry (e.g., Hedges et al. 1994; Benner and Opsahl 2001; Engelhaupt and Bianchi 2001).

Among the most commonly encountered coastal ecosystems in tropical and subtropical areas lay the coastal wetland, fringe mangrove, and seagrass estuarine ecotones, similar in many aspects to the aquatic environments of South Florida, and information regarding UDOM characteristics in such systems is scarce. Additionally, the oligotrophic nature of the Everglades proper adds to the interest in UDOM cycling due to the organic nature of essential nutrients that is often observed in such systems. To better understand the biogeochemical role of UDOM in aquatic ecosystems, detailed surveys of the chemical characteristics of UDOM in different aquatic subenvironments are necessary because the molecular distribution of UDOM components will reflect the origin and biogeochemical processes taking place in the environment. Therefore, the objective of this study is to investigate in detail the molecular characteristics of DOM in a subtropical, oligotrophic aquatic environment, namely, the Florida coastal Everglades, along a freshwater wetland, mangrove fringe to estuarine transect. For this purpose, surface waters from five sites along Taylor Slough (TS), ranging from freshwater marsh to mangrove ecotone environments, as well as three sites within the Florida Bay (FB) estuary were collected for analyses. Fluorescence properties of DOM were measured for a quick assessment of DOM quality. UDOM samples (<0.7 μm, >1,000 Da) were concentrated and freeze dried for the determination of bulk C composition, lignin-phenol concentration, and neutral sugar composition. These molecular characteristics were investigated by ^{13}C-nuclear magnetic resonance (NMR) spectroscopy, TMAH thermochemolysis (Hatcher et al. 1995), and hydrolysis with trifluoroacetic acid (TFA; Amelung et al. 1996), respectively. Furthermore, UDOM samples were classified by cluster analysis based on their pyrolysis products obtained via flash pyrolysis-gas chromatography/mass spectroscopy (Py-GC/ MS).

Site description

TS is the shallow-water, marl soil-based drainage system of southeast ENP (from $25°25'26''$N, $-80°35'25''$W to $25°11'27''$N, $-80°38'21''$W). It is the second largest drainage system in ENP and the primary source of freshwater dis-
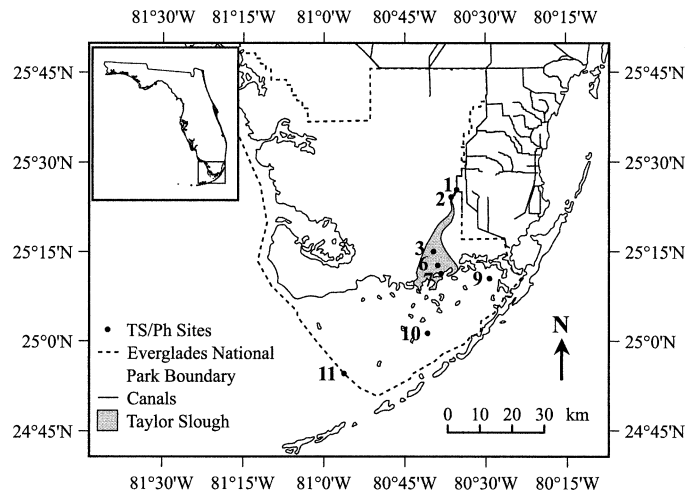

Fig. 1. Map of sampling sites showing the studied Florida Coastal Everglade Taylor Slough/Panhandle (TS/Ph) sampling stations.

charge and transport of terrigenous materials to Florida Bay (Boyer and Jones 1999). Along TS, dominant freshwater marsh vegetation changes from a mixture of sawgrass (*Cladium jamaicense*), spikerush (*Eleocharis cellulosa*), and periphyton assemblages to a red mangrove (*Rhizophora mangle*) -dominated fringe and into the seagrass-dominated FB. The coastal zone and northeast FB are not significantly affected by tides because FB is a large (2,200 km^2), shallow, enclosed estuarine system (Enos and Perkins 1979). The system receives freshwater inflow from the Everglades and opens to the Gulf of Mexico along its western margin. The sampling sites in this study (Fig. 1) are located in the Taylor Slough/Everglades Panhandle (TS/Ph) portion of ENP and they are identical to those established for the Florida Coastal Everglades Long Term Ecological Research (FCE-LTER) program. The TS/Ph sites are composed of TS sites (1, 2, 3, 6, and 7), Ph sites (4, 5, and 8), and FB (9, 10, and 11), which will, from here on be referred to by their corresponding station numbers. In this study, the entire transect of Taylor Slough and Florida Bay was investigated. The sampling sites begin at a canal overflow point and extend south to the Florida Bay estuary. The dominant vegetation of sites 1 and 2 is a mixture of sawgrass, spikerush, and assemblages of periphyton. Site 3 is almost equivalent to sites 1 and 2 but coexists with dwarf mangrove forests, while sites 6 and 7 are in the mangrove fringe. Sites 9–11 are located in Florida Bay, where the main vegetation is seagrass (mainly turtle grass; *Thalassia testudinum*; Zieman et al. 1989).

Sampling and methods

Sample collection—Surface-water samples were collected in 25-L white, low-density polyethylene Carboy bottles (Nalge Nunc International) during the early part of the dry season (from 5 Dec 2001 to 28 Jan 2002). The bottles were cleaned by soaking in 0.5 mol L^{-1} HCl followed by 0.1 mol L^{-1} NaOH for 24 h each. Water samples were filtered through precombusted (470°C for 4 h) 0.7-μm GF/F glass

fiber filters (Whatman International), followed by concentration using a Pellicon 2 Mini tangential flow ultrafiltration system equipped with a nominal 1,000 Da molecular-weight cut-off regenerated cellulose membrane (Millipore) (Dai et al. 1998). The water samples were concentrated to 100 ml at an inlet pressure of 10 psi and an outlet pressure of 8 psi. For water samples collected from FB (sites 9–11), diafiltration was conducted as follows: 1 L of Milli-Q® water (Millipore) was added to the concentrated sample and then reconcentrated to 100 ml. This process was repeated a total of three times. The concentrated samples were freeze dried and powdered with an agate mortar. While the percentage of UDOC to total DOC was not determined in this experiment, % UDOC to total DOC collected from the same sites in September–October 2002 was 14% ± 4% and 24% ± 11% for TS and FB, respectively.

Water samples for fluorescence analysis were collected separately in 30-ml brown polyethylene bottles, stored on ice, and transported to the laboratory. The water samples were filtered through precombusted (470°C for 4 h) Whatman GF/F glass fiber filters prior to analysis.

Dissolved organic carbon concentration and fluorescence analysis—Dissolved organic carbon (DOC) concentrations were analyzed using a high-temperature catalytic combustion method on a Shimadzu TOC-5000 total organic carbon analyzer. Samples (4 ml) were acidified with 10 μl of concentrated HCl and sparged for 5 min with nitrogen to remove inorganic carbon. The mean of 3–6 injections (coefficient of variation [C.V.] < 2%) was reported for each sample. Fluorescence spectra were recorded on a Perkin Elmer LS 50B spectrometer equipped with a 150-W xenon arc lamp as the light source. The emission monochromator was scanned from 250 to 550 nm with excitation at 313 and 370 nm (Donard et al. 1989; De Souza Sierra et al. 1994). Further, synchronous excitation-emission fluorescence spectra at a constant offset value ($\delta\lambda$) between emission and excitation wavelength of 30 nm were measured from 250 to 550 nm (Lu et al. 2003; Jaffé et al. 2004). Both excitation and emission slits were set at 10 nm. Absorbance of the DOM solution was scanned from 250 to 550 nm for the correction of inner-filter effects on a Shimadzu UV-2101PC ultraviolet-visible spectrophotometer. The inner-filter effects were corrected for all the spectra following the procedure described by McKnight et al. (2001): $F_{cor} = F_{obs} \times 10^{0.5 \times (A_{em} + A_{ex})}$, where F_{cor} and F_{obs} are the corrected and observed fluorescence intensity, respectively; A_{em} and A_{ex} are the absorbances at emission and excitation wavelength (1-cm path length), respectively. Spectra were not corrected for instrumental response. Milli-Q water was used as a blank to background subtract water Raman scatter peaks. The fluorescence intensities were expressed in quinine sulfate units (QSU): 1 QSU = 1 μg L^{-1} of quinine sulfate monohydrate in a 0.05 mol L^{-1} H$_2$SO$_4$ solution at excitation/emission (Ex/Em) = 350/450 nm (Wu and Tanoue 2001). Four indices were used in this study (Lu et al. 2003; Jaffé et al. 2004): (1) maximum intensity (Int$_{max}$), maximum fluorescence emission intensity with an excitation of 313 nm (Donard et al. 1989); (2) maximum wavelength (λ_{max}), the wavelength that gives the Int$_{max}$ (Donard et al. 1989); (3) fluorescence index (F. I.), the ratio

of emission intensities at 450 and 500 nm with an excitation of 370 nm (f$_{450}$/f$_{500}$) (Battin 1998; McKnight et al. 2001); (4) % 285, calculated from a synchronous spectrum. % 285 = Ex$_{285}$/(Ex$_{285}$ + Ex$_{350}$ + Ex$_{385}$ + Ex$_{460}$) × 100, where Ex$_{285}$, Ex$_{350}$, Ex$_{385}$, and Ex$_{460}$ are the emission intensities at the respective excitation wavelengths (nm) noted in the subscripts (Lu et al. 2003). The reproducibility of the optical measurements described above was found to be below ±5% for Int$_{max}$, ±2.8 nm for λ_{max}, ±0.02 for F. I., and ±0.4% for % 285.

Carbon content—Total organic C content was measured on a Carlo Erba NA 1500 nitrogen/carbon analyzer at 1050°C, hippuric acid as a standard. To remove carbonate, 2–5 mg of powered sample was weighed into a silver capsule and exposed to hydrochloric acid vapor for 4 h, followed by drying under vacuum to eliminate any remaining hydrochloric acid (Hedges and Stern 1984). Then, the capsules were closed for analysis. The measurement was conducted in duplicate (average C.V.; 0.2% for C, 0.9% for N).

Solid-state ^{13}C NMR spectroscopy—Solid-state ^{13}C NMR spectra were obtained at a ^{13}C resonance frequency of 50.3 MHz on a Bruker ASX200 NMR spectrometer equipped with a commercial 7-mm cross-polarization magic angle spinning (CPMAS) probe using a standard CPMAS pulse sequence. ^{13}C chemical shifts are expressed with respect to tetramethylsilane by using the carbonyl carbon of glycine (176.48 ppm) as an external reference. Other analytical conditions were as follows: rotation frequency, 4.5 kHz; contact time, 1 ms; recycle delay, 2 s; scans accumulated, 3,000–20,000; spectral width, 25 kHz; filter frequency, 32 kHz; Lorentzian line broadening, 120 Hz. NMR spectra were divided into four regions according to chemical shifts as follows: 0–45 ppm (alkyl C), 45–110 ppm (*O*-alkyl C), 110–160 ppm (aromatic C), 160–210 ppm (carbonyl C) (Kögel-Knabner 1997). The relative abundance of these regions was reported as percentage of total spectral area, at reproducibility within ±2%, determined by repacking the sample in triplicate. The first-order spinning sidebands of aromatic and carbonyl signals (220 and 260 ppm, respectively) were corrected if necessary, according to Knicker and Skjemstad (2000).

Analysis of neutral sugars—Sugar composition analysis was performed according to Amelung et al. (1996). Briefly, about 10 mg of powdered UDOM sample was mixed with an internal standard (50 μg myo-inositol; Sigma-Aldrich) and 10 ml of 4 mol L^{-1} trifluoroacetic acid (TFA; Sigma-Aldrich) and hydrolyzed at 105°C for 4 h. Following filtration with a pre-combusted GF/F glass fiber filter, the solution was rotary-evaporated to remove TFA. The sample was then reconstituted with 2 ml water and passed through Amberlite® XAD-4 (Sigma-Aldrich), and Dowex® 50 W × 8 (Sigma-Aldrich) columns, successively. The sample was freeze dried and derivatized with 400 μl bis-(trimethylsilyl)-trifluoroacetamide (Aldrich) at 75°C for 5 min. Analysis was performed using a Hewlett Packard 6890 GC-MS series gas chromatograph coupled to a 5973 quadrupole mass selective detector (Hewlett-Packard). One μl of the solution was injected into a J&W DB1MS capillary column (100% dimethylpolysilox-

Table 1. Concentration and fluorescence properties of DOM in Taylor Slough and Florida Bay.

Site number	DOC (μmol L^{-1})	Fluorescence emission index (F. I.)	Maximum fluorescence wavelength (nm)	Maximum fluorescence intensity (QSU)	%285	C:N ratio of UDOM (mol basis)	Salinity	Sampling date
Taylor Slough (TS)								
1	962	1.56	418.2	40.8	28.6	25.8	0	06 Dec 01
2	778	1.61	413.9	32.9	30.3	23.0	0	06 Dec 01
3	699	1.53	423.1	30.7	32.7	23.1	0	05 Dec 01
6	775	1.55	423.0	31.9	37.1	21.4	0.7	10 Dec 01
7	903	1.49	429.6	36.4	30.7	21.7	2.1	10 Dec 01
Florida Bay (FB)								
9	511	1.61	396.3	18.8	47.8	15.7	28.2	20 Jan 02
10	557	1.70	390.9	19.5	66.1	15.7	35.0	28 Jan 02
11	395	1.62	396.2	15.9	52.5	17.5	37.0	28 Jan 02

ane; 30-m length $\times$ 0.25-mm inner diameter $\times$ 0.25-μm film thickness; J&W Scientific). Quantification was based on the comparison of the peak area on the total ion chromatogram with known concentrations of standard materials (arabinose, ribose, xylose, rhamnose, fucose, mannose, galactose, and glucose; Sigma-Aldrich) that were processed in the same way as the samples. Initial oven temperature was set at 160°C, held for 0.5 min, ramped at 8°C min^{-1} to 185°C, followed by 3°C min^{-1} to 191°C, by 0.5°C min^{-1} to 195°C, and thereafter by 10°C min^{-1} to 250°C and held for 5 min. Mass spectra were recorded under electron impact ionization conditions (70 eV) at 1 scan s^{-1} in the m/z = 50–500 mass range. Detection limit was ~100 μg g^{-1} in UDOM.

Tetramethylammonium hydroxide thermochemolysis— The tetramethylammonium hydroxide (TMAH) thermochemolysis was performed according to Hatcher et al. (1995). Briefly, 4–10 mg powdered UDOM sample was placed in a 5-ml glass ampoule (Chemglass) and 200 μl of solution consisting of 25% TMAH in methanol (Sigma-Aldrich) and 200 μl of an internal standard, n-eicosane (50 μg ml^{-1} in methanol) were added. The methanol was evaporated under vacuum, and the ampoule was flame sealed and placed in a gas chromatographic oven at 250°C for 30 min. After cooling, the ampoule was cracked open and the inner glass surface was washed with 1 ml of methylene chloride three times and concentrated to approximately 200 μl under a gentle stream of nitrogen. Analysis of this extract was performed on a Hewlett Packard 6890 GC-MS series gas chromatograph (GC) coupled to a 5973 mass selective detector. One microliter of the solution was injected into a DB5MS (5% phenyl, 95% methyl polysiloxane; 30-m length, 0.25-mm inner diameter, 0.25-μm film thickness; J&W Scientific) capillary column. Helium served as the carrier gas. The column temperature was programmed as follows: initial temperature at 40°C, ramped at 10°C min^{-1} to 120°C, followed by 3°C min^{-1} to 200°C, and thereafter by 4°C min^{-1} to 300°C (Mannino and Harvey 2000). Mass spectra were recorded under electron impact ionization conditions (70 eV) at 1 scan s^{-1} in the m/z = 50–500 mass range. The detection limit was 1 ng for the eicosane standard. The assignment of peaks was based on the comparison of mass spectra with the spectral

library (NIST 98) and/or mass spectral interpretation. A response factor for phenolic compounds to eicosane was calculated by averaging the relative response of methylation products of vanillin (Aldrich), vanillic acid (Aldrich), and acetovanilone (Aldrich) to that of the internal standard. The concentration of phenolic compounds was estimated by comparing the area with that of the eicosane standard. The analysis of each sample was conducted in triplicate. The C.V. was 20% $\pm$ 8% and 15% $\pm$ 10% for sum of V and S compounds (mg 100 mg C^{-1} UDOM) and DV and DS compounds, respectively (see Table 4 for nomenclature).

Pyrolysis gas chromatography/mass spectrometry—Pyrolysis gas chromatography/mass spectrometry (Py-GC/MS) analyses were performed as previously described (Lu et al. 2003) except that, in this study, UDOM instead of total freeze-dried DOM samples were used. Briefly, UDOM samples (about 5 mg) were pyrolyzed at 650°C for 20 s in a helium atmosphere using a Pyroprobe 1500 pyrolyzer (Chemical Data Systems). Separation of pyrolysis products was carried out on a DB5MS fused-silica column (30-m length $\times$ 0.25-mm inner diameter, 0.25-μm film thickness; J&W Scientific) at a split ratio of 1:75 under helium atmosphere. The oven was connected to the split/splitless injection port of a Hewlett Packard 6890 GC coupled to a HP 5973 mass spectrometer. The oven temperature program was as follows: initial temperature was held at 40°C for 2 min, ramped at 7°C min^{-1} to 300°C, where it was held for 15 min. The assignment of peaks was based on the comparison of mass spectra with the spectral library (NIST 98) and/or mass spectral interpretation. Other analytical conditions were identical with those of TMAH thermochemolysis described above.

Hierarchical cluster analysis of pyrolysis products— Based on the relative abundance of individually identified pyrolysis products (approximately 100 compounds; peak area of individual compound to total peak area of identified compounds in pyrogram) a hierarchical cluster analysis (HCA) was performed using an agglomerative method (Ward method) with SPSS version 11.0.1 software (SPSS) for the interpretation of the multivariate pyrolysis data set.

Table 2. Carbon composition of Taylor Slough and Florida Bay UDOM based on ^{13}C CPMAS NMR spectra.

Site number	% alkyl C (0–45 ppm)	% O-alkyl C (45–110 ppm)	% aromatic C (110–160 ppm)	% carbonyl C (160–210 ppm)	alkyl C/O-alkyl C	Aromaticity*
Taylor Slough (TS)						
1	18 (29)†	35 (24)	25	22	0.52	32
2	19 (30)	50 (39)	16	15	0.38	18
3	19 (30)	53 (42)	14	14	0.35	16
6	22 (35)	59 (46)	8	11	0.37	9
7	21 (33)	55 (43)	13	11	0.37	14
Florida Bay (FB)						
9	13 (22)	76 (67)	2	9	0.17	2
10	13 (21)	77 (69)	1	9	0.16	1
11	15 (23)	69 (61)	5	11	0.21	5
Average (2–7)	20±1 (32±2)	54±4 (42±3)	13±3	13±2	0.37±0.01	14±4
Average (9–11)	13±1 (22±1)	75±4 (66±4)	2±2	10±1	0.18±0.03	3±2

* Aromaticity is calculated by % aromatic C/(% alkyl C + % O-alkyl C. + % aromatic C) × 100.
† Numbers in partentheses are C compositions calculated according to Engelhaupt and Bianchi (2001) and Bianchi et al. (2004) (alkyl C 0–60 ppm; O-alkyl C, 60–110 ppm).

The objective of this study was to use this statistical approach to classify the UDOM composition of different samples using a molecular fingerprinting approach.

Results and discussion

Bulk DOM characteristics—The DOC concentrations at the TS sites (1–7) and the FB sites (9–11) were 823 ± 107 and 488 ± 83 μmol L^{-1}, respectively. The concentration was slightly higher at site 1 and decreased along the slough to site 3, and then increased to site 7, probably due to mangrove-derived DOM inputs (Table 1). The DOC concentration of surface water in the southern Everglades (or ENP) was reported to be lower than the northern Everglades (Qualls and Richardson 2003), which may reflect lower primary productivity and lower soil organic C concentration of marl soil compared with peat soil. However, the DOC concentration at the FB sites was high compared with other estuaries (Boyer et al. 1999; Del Giorgio and Davis 2003), which can be attributed to (1) FB being a semiclosed system, which results in a poor water exchange rate with the open ocean (Enos and Perkins 1979); (2) concentration of DOM due to high water-evaporation rates (Fourqurean et al. 1993); and (3) DOM enrichment due to seagrass/phytoplankton primary productivity in addition to land-derived DOM inputs to FB.

The F. I. values ranged from 1.49 to 1.70, showing the lowest (high terrestrial OM source) and highest (high microbial OM source) values at sites 7 and 10, respectively (Table 1). The maximum emission wavelength (λ_{max}) value of water samples collected from a typical freshwater marsh (site 2, 413.9 nm) was shorter than that of mangrove-influenced sites (sites 6 and 7; 426.3 ± 3.3 nm), and blue shifted (~30 nm) at the FB sites (sites 9–11; 394.5 ± 3.1 nm). F. I. and λ_{max} are often used to estimate the source of DOM such as terrestrial versus marine (microbial) (De Souza Sierra et al. 1994; Battin 1998; Jaffé et al. 2004). In this study, these values were inversely correlated ($r^2 = 0.83$; $p < 0.01$). The

Table 3. Molar composition (mol%) of neutral sugars in UDOM collected from Taylor Slough and Florida Bay.*

Site number	Ara	Rib	Xyl	Rha	Fuc	Man	Gal	Glu	NS† mg C g C^{-1} UDOM
Taylor Slough (TS)									
1	7.0±0.0	0.9±0.1	17.6±0.4	15.7±0.1	11.8±0.2	10.3±0.1	20.9±0.5	16.0±0.2	19.6±0.3
2	6.0±0.1	0.5±0.0	20.6±0.3	13.2±0.1	8.6±0.0	13.4±0.0	18.3±0.0	19.4±0.6	130.6±3.8
3	6.8±0.3	0.4±0.1	20.3±0.7	14.2±0.6	13.1±0.7	10.0±0.0	17.7±0.7	17.5±0.3	123.8±6.9
6	3.7±0.3	0.6±0.2	16.0±0.8	16.8±0.6	4.6±1.2	14.9±0.3	16.8±0.7	26.4±1.9	118.9±25.2
7	6.9±0.0	0.9±0.0	16.5±0.2	15.2±0.3	8.8±0.1	12.1±0.1	16.8±0.1	22.9±0.0	96.1±2.0
Florida Bay (FB)									
9	4.3±0.3	1.0±0.1	18.5±0.3	16.2±1.2	10.1±1.0	12.2±0.5	19.2±2.2	18.5±1.4	111.5±28.5
11	3.9±0.4	2.2±0.1	22.1±0.7	10.9±2.5	11.1±2.8	10.4±0.2	21.2±0.1	18.3±4.4	110.1±36.7
Average (2–7)	6±1	1±0	18±2	15±1	9±3	13±2	17±1	22±4	117±20
Average (9, 11)	4±0	2±1	20±2	14±4	11±2	11±1	20±2	18±4	111±38

* Ara, arabinose; Rib, ribose; Xyl, xylose; Rha, rhamose; Fuc, fucose: Man, mannose; Gal, galactose; Glu, glucose.
† Neural sugar concentration. Site 10 was not analyzed.

Table 4. Yields of lignin phenols from Taylor Slough and Florida Bay UDOM, released by thermochemolysis with TMAH (mg 100 mg C^{-1} UDOM).*

Compound	Group	Site number							
		1	2	3	6	7	9	10	11
Benzaldehyde, 3,4-dimethoxy-	V	0.009	0.019	0.018	0.037	0.022	0.002	0.004	NQ
Benzene, 1,2-dimethoxy-4-(1-propenyl)-	V	0.012	0.012	0.012	0.001	0.003	0.003	NQ	NQ
Ethanone, 1-(3,4-dimethoxyphenyl)-	V	0.017	0.020	0.031	0.032	0.030	0.011	0.012	0.001
Benzoic acid, 3,4-dimethoxy-, ME	V	0.114	0.144	0.181	0.230	0.166	0.050	0.040	0.041
Benzenepropanoic acid, 3,4-dimethoxy-,	V	0.004	NQ	0.003	0.007	0.006	0.001	0.004	NQ
Ethanone, 1-(3,4,5)-trimethoxypheny-	S	0.013	0.008	0.019	0.012	0.010	NQ	NQ	NQ
Benzoic acid, 3,4,5-trimethoxy-, ME	S	0.034	0.055	0.069	0.076	0.049	NQ	NQ	NQ
Benzaldehyde, 3,4,5-trimethoxy-	S	NQ	NQ	NQ	0.003	0.004	NQ	NQ	NQ
Phenol 2-methoxy-	DV	0.015	0.052	0.093	0.072	0.076	0.218	0.260	0.028
Benzene, 1,2-dimethoxy-	DV	0.021	0.024	0.065	0.083	0.057	0.219	0.239	0.166
3,4-Dimethoxytoluene	DV	0.007	NQ	0.008	0.013	0.012	0.038	0.053	NQ
1,2,3-Trimethoxybenzene	DS	0.017	0.047	0.071	0.072	0.054	0.111	0.100	0.153
Benzene, 1,2,3-trimethoxy-5-methyl-	DS	0.024	NQ	0.049	0.070	0.035	0.052	0.057	0.050

* NQ, not quantified compounds below detection limit or with coelution interference; ME, methyl ester.

F. I. values of microbially and terrestrially derived fulvic acids are reported to be ~1.9 and ~1.4, respectively (McKnight et al. 2001). Therefore, the higher fluorescence index value at site 2 compared with sites 6 and 7 may indicate a larger contribution of periphyton or microbial-derived DOM in the freshwater marshes (Table 1). On the other hand, the low F. I. and elevated λ_{max} at site 7 suggests a strong influence of mangrove-derived DOM. The F. I. and λ_{max} values of FB sites suggest significantly higher microbial DOM sources compared with the freshwater sites.

The % 285 mean value in the riverine zone (sites 1–7) was 32% ± 3% (Table 1). It increased consistently from site 1 to site 6, and after dropping at site 7, it reached its highest value in FB (55% ± 10%). The fluorescence emission at an excitation of 285 nm is often attributed to the presence of dissolved proteinaceous materials (e.g., Coble et al. 1990; Yamashita and Tanoue 2003). With the exception of site 7, the increase in the % 285 index indicates increased relative amounts of proteinaceous materials and thus may also be an indication of the presence of more labile DOM components. The lower % 285 value at site 1 is ascribed to the canal water input, where DOM has a longer residence time and therefore contains less labile DOM due to bio- and photo-degradation (Lu et al. 2003). The increase of % 285 value along the TS transect may suggest that the relative abundance of proteinaceous materials is increasing along this transect due to autochthonous inputs. This observation is in agreement with previous reports on this system, which suggested significant biogenic inputs of nonhumic materials in the freshwater marshes of the Everglades (Lu et al. 2003).

The blue shift of λ_{max} in FB indicates the dominant DOM source shifted from terrestrial to marine, which is commonly reported in the literature (e.g., De Souza Sierra et al. 1994; Jaffé et al. 2004). Seagrass, the dominant vegetation of FB, is known to produce significant amounts of DOM (Ziegler and Benner 1999). Based on the relatively high DOC concentration and high % 285 values, labile, seagrass-derived DOM is probably an important component in FB waters.

In summary, the DOC concentrations and optical characteristics of the DOM along the TS and FB suggest that the DOM characteristics in the riverine zone are different from inflowing canal waters. DOM characteristics are dominated by terrestrial sources, with strong indication of periphyton inputs in the freshwater marsh portion of the slough. These autochthonous DOM inputs result in a high relative abundance of potentially biolabile proteinaceous compounds (see also Lu et al. 2003). Water samples from FB contain elevated concentrations of DOM, most likely from autochthonous sources, characterized by a significantly higher relative abundance of proteinaceous components compared with TS, indicating a clear DOM source change between TS and FB, as further evidenced by a significant blue shift in the λ_{max} value.

Chemical characterization of UDOM—CPMAS ^{13}C-NMR: UDOM samples fell into three groups based on the similarity of ^{13}C NMR spectra, that is, site 1 (canal inflow point), sites 2, 3, 6, 7 (riverine zone), and sites 9, 10, 11 (FB estuary; Fig. 2). The aliphatic C region (0–45 ppm) for UDOM from TS had a broad signal, suggesting high heterogeneity of alkyl carbons. The absence of a long-chain aliphatic C peak at around 32 ppm suggested minor contributions of organic compounds derived from cutin and suberin (Kögel-Knabner et al. 1992). The broad peak at around 30–55 ppm is ascribed to branched alkyl C and C_α of amino acids in proteins (Kögel-Knabner 1997). The methoxyl peak at 58 ppm and small phenolic C peak at around 140–160 ppm, which are often attributed to lignin-derived phenols, were observed for the TS samples. The latter peak can also be attributed to tannins (Preston 1999). However, because intensities of those peaks were small, those compounds are believed to have suffered diagenetic reworking.

A large, sharp peak at around 72 ppm, which is mainly ascribed to polysaccharides, was observed for all samples except for site 1. When looking at the proportion of each C species of UDOM in the riverine zone (sites 2–7; Table 2), alkyl C (0–45 ppm), *O*-alkyl C (45–110 ppm), aromatic C (110–160 ppm), and carbonyl C (160–210 ppm) were 20% ± 1%, 54% ± 4%, 13% ± 3%, and 13% ± 2%, respec-
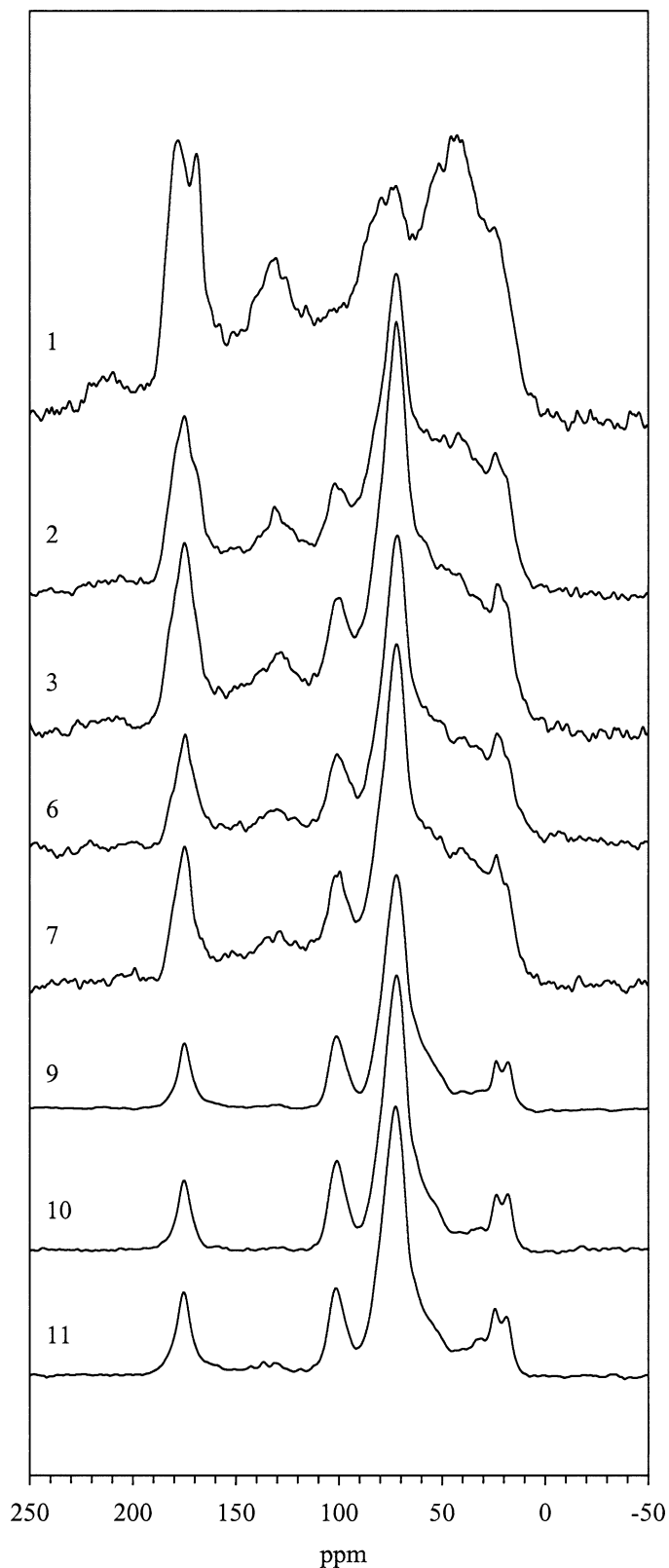
tively. *O*-alkyl C made up the largest proportion of UDOM, and the abundance increased downstream.

The portion of the *O*-alkyl C region between 45 and 60 ppm includes the *N*-alkyl C as well as methoxyl-C (-O-CH$_3$) (Kögel-Knabner 1997). The former derives from C$_\alpha$ of amino acids in proteins, while the latter is mainly derived from lignin. From the C:N ratio of our UDOM samples (Table 1), the N concentration in UDOM was calculated to be 4.4% and 6.1 mol% of C on average for sites 2–7 and 9–11, respectively. Under the assumption that the entire N is in the form of protein, the C$_\alpha$ of amino acids would also account for 4.4 and 6.1 mol% of total C, representing the *N*-alkyl C signal contribution to the total C in the NMR spectra. Therefore, its contribution to the total NMR signal intensity is only minor. The signal between 60 and 110 ppm will be most representative of carbohydrates because all the ^{13}C NMR signals of neutral sugars fall in this region. Table 2 shows the *O*-alkyl C abundance based on the 45–110-ppm range and, in parentheses, the abundance excluding the contribution from the 45–60-ppm signal. In either case, the *O*-alkyl C or carbohydrate abundance in UDOM is very high in the FCE compared with other aquatic environments (e.g., Engelhaupt and Bianchi 2001; Benner 2003; Bianchi et al. 2004). Considering the low relative abundance of *O*-alkyl C in UDOM in an oligotrophic river draining the European Alps (Kaiser et al. 2003), both greater preservation and production of carbohydrates at the Everglades riverine zone are considered to be responsible for the high *O*-alkyl C abundance in the FCE.

When compared with an earlier study where DOM was characterized in the Everglades (Lu et al. 2003), in which powdered DOM samples were obtained by freeze drying after concentration of whole-water samples using a rotary evaporator, UDOM in this study were lower in alkyl C and carboxyl C but enriched in *O*-alkyl C, which suggests that higher percentages of alkyl C in DOM belong to low molecular-weight DOM fraction and are preferentially removed by ultrafiltration. At the FB sites, *O*-alkyl C concentrations were even higher than that for the riverine zone, reaching 78% at site 10. Aromatic C concentrations at the FB sites were very low (2% ± 2%), suggesting an extensive dilution and/or degradation of land-derived UDOM in FB.

Neutral sugar analysis—Neutral sugars are bioreactive and the concentration has been suggested as an indicator of the freshness or amount of labile DOM (e.g., Amon and Benner 2003 and references therein). The neutral sugar concentrations in the UDOM in the FCE were fairly consistent throughout the TS and FB (115 ± 11 mg C g C^{-1} UDOM) except for site 1 (Table 3). The high neutral sugar concentration of UDOM in TS compared with those in other riverine environments (e.g., 36 ± 8 mg C g C^{-1} UDOM for the Amazon River and its major tributaries, Hedges et al. 1994; 17–35 mg C g C^{-1} UDOM for Arctic rivers, Amon and Benner 2003; 13–42 mg C g C^{-1} UDOM for Mississippi river, Benner and Opsahl 2001) suggests UDOM in TS contains relatively large amounts of fresh and/or labile DOM. Table 3 shows the molecular composition and relative abundance (as mol%) of the sugars identified in the UDOM samples. While arabinose and ribose were consistently present at lowest abundance, and rhamnose, fucose, and mannose at

Fig. 2. ^{13}C CPMAS NMR spectra of UDOM from Taylor Slough and Florida Bay. Site numbers are indicated.
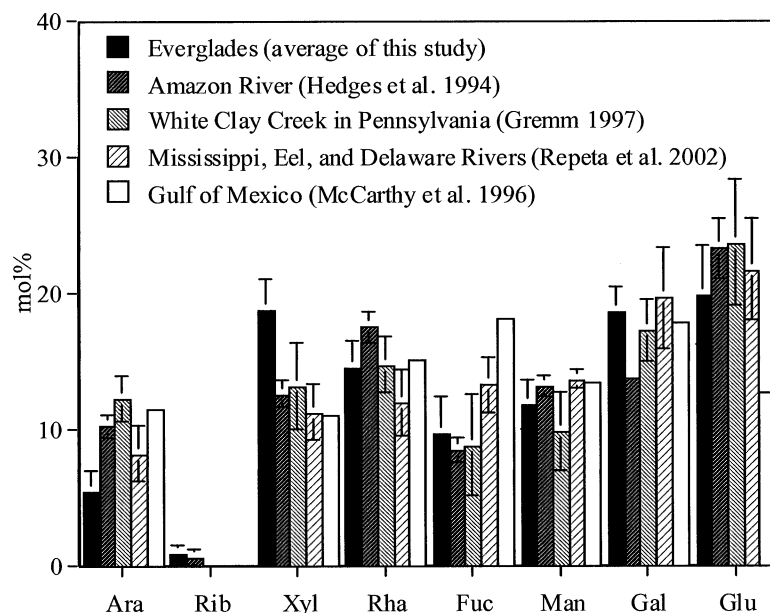
Fig. 3. Comparison of Everglades UDOM neutral sugar composition with that of other riverine systems and the Gulf of Mexico. Abbreviations: Ara = arabinose; Rib = ribose; Xyl = xylose; Rha = rhamnose; Fuc = fucose; Man = mannose; Gal = galactose; Glu = glucose. Sugar composition from other reports was recalculated based on Ara, Rib, Xyl, Rha, Fuc, Man, Gal, and Glu concentrations (mol%) for comparative purposes when needed.

an intermediate level, xylose, galactose, and glucose were the most abundant in all the samples. No major change in the compositions was observed throughout the TS to FB transect, except for site 6, where the sugar composition was lower in fucose and higher in glucose abundance. The low fucose and high glucose concentrations are consistent with neutral sugar compositions often observed for freshly leached UDOM (Ochiai and Hanya 1980; Amon and Benner 2003). The DOM at site 6 in particular is likely to be influenced by inputs from DOM derived from red mangroves as well as aquatic macrophytes and macroalgae, which are all abundant at this very shallow site.

Neutral sugar compositions of UDOM were reported to be almost identical for a variety of aquatic environments such as rivers, lakes, oceans, and pore water of marine sediments (Repeta et al. 2002). However, other authors have reported small, but significant, differences in the neutral sugar composition between riverine and marine UDOM (Amon and Benner 2003). Neutral sugars are known to undergo quick diagenesis (Ochiai and Hanya 1980; Amon and Benner 2003), and Amon and Benner (2003) concluded that the difference in the composition reflects differences in the diagenetic state rather than the source. The neutral sugar composition of UDOM in the Everglades was almost identical to that of other rivers and the adjacent Gulf of Mexico as reported in the literature (see Fig. 3 for examples), although higher xylose and lower arabinose concentrations were observed for the Everglades. Because plant materials contain relatively large amounts of xylose (Moers et al. 1990) in the form of xylan as an important component of hemicellulose, vegetation, rather than more degraded soil/sediment-derived organic matter, is suggested as a more important source of neutral sugars in the Everglades system. This is in contrast

to other riverine systems (e.g., Hedges et al. 1994; Benner and Opsahl 2001; Repeta et al. 2002), where soils are expected to be the main source of DOM.

Lignin phenol analysis—Lignin is the second most abundant biomolecule after cellulose (Crawford 1981) and it is exclusively of terrestrial/plant origin. Thus, it can be used as a biomarker for terrestrial, higher plant-derived DOM. Lignin is an amorphous, three-dimensional polymer of substituted phenyl-propane units. It is originally insoluble, but after its partial oxidative degradation, it contributes to the DOM pool. From the UDOM collected, 13 kinds of phenolic compounds that have vanillyl and syringyl structures (lignin phenols) were detected and they were categorized into 4 groups (Table 4). The *p*-hydroxy phenols were not included in this study because they are known to be derived from nonlignin compounds (Goñi and Hedges 1995; Opsahl and Benner 1995). Groups V and S represent samples containing vanillyl or syringyl compounds, respectively, which have been reported as lignin-derived phenols (Hatcher et al. 1995; del Rio et al. 1998). Groups DV and DS contain vanillyl and syringyl units, respectively, which have been reported for lignin in a highly degraded state or from nonlignin sources (e.g., 1,2-dimethoxybenzene and 1,2,3-trimethoxybenzene are also derived from condensed tannins; del Rio et al. 1998; Maie unpubl. data).

Benzoic acid, 3,4-dimethoxy-, methyl ester (V) was the most abundant lignin phenol in TS, while 2-methoxy-phenol (DV) and 1,2-dimethoxy-benzene (DV) were dominant in FB (Table 4). The total amount of phenolic compounds belonging to V+S (Fig. 4) increased from site 1 (0.20 mg 100 mg C^{-1} UDOM) to site 6 (0.39 mg 100 mg C^{-1} UDOM), and dropped at sites 9, 10, and 11 (FB) (0.05 mg 100 mg
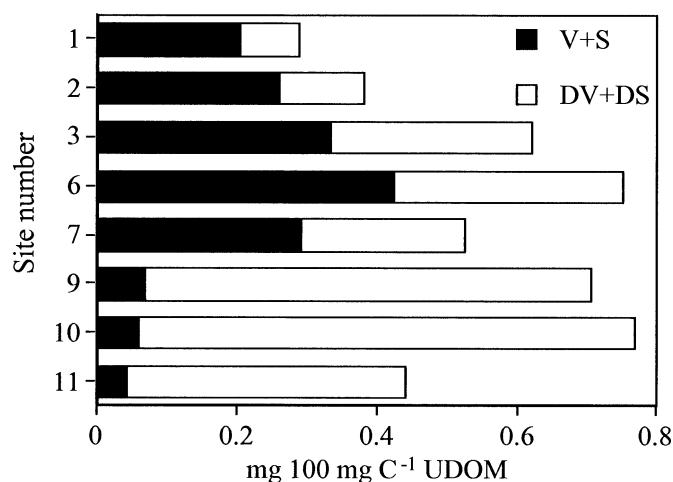
Fig. 4. Yields of lignin phenols in UDOM from Taylor Slough and Florida Bay. V+S and DV+DS refer to the sum of phenolic compounds belonging to groups V and S and groups DV and DS, respectively (see Table 4 for grouping).

Fig. 5. Syringyl-to-vanillyl (S:V) ratio of UDOM from Taylor Slough and Florida Bay. The * indicates not calculated because the yields of S compounds were the below the detection limit.

C^{-1} UDOM). In contrast, the total amount of DV+DS compounds was much higher in FB. The concentration of V+S was positively correlated with % alkyl C that was estimated from ^{13}C NMR ($r^2 = 0.89$; $p < 0.001$), while that of DV+DS was positively and negatively correlated with % O-alkyl C ($r^2 = 0.89$; $p < 0.001$) and % aromatic C ($r^2 = 0.84$; $p < 0.0013$), respectively. DOM rich in alkyl C and aromatic C is known to be produced by oxidative degradation of lignin in peat (Orem and Hatcher 1987). This may explain the observed positive correlation between the concentrations of V+S and alkyl C. The reasons for a positive correlation between the concentrations of DV+DS and O-alkyl C is unclear; these phenols may be mainly produced from nonlignin precursors.

The syringyl-to-vanillyl (S:V) ratio was almost constant throughout TS (0.28–0.36) but significantly lower at the FB sites (0–0.05) (Fig. 5). Photochemical degradation of lignin-derived phenolic compounds decreases the S:V ratio (Opsahl and Benner 1998). The higher sensitivity of DOM to photodegradation in TS relative to FB (unpubl. data) is also consistent with the lower S:V values found in FB. Alternatively, leachate from seagrass may also contribute to the low S:V ratios found in FB. In a leaching experiment by the authors (unpubl. data), the UDOM leached from the sea-grass *Thalassia testudinum* during a 2-week period contained significant amounts of vanillyl phenols while it did not contain detectable amounts of syringyl phenols. Opsahl and Benner (1993) also reported that a seagrass (*Halodule wrightii*) had a high contribution of free and esterified vanillyl phenols that were rapidly leached from plant tissues.

Py-GC/MS characterization—The Py-GC/MS runs showed the presence of more than 100 different compounds, derived from the thermal degradation of the UDOM (see also Lu et al. 2003 and references therein). In this study, Py-GC/MS data were used to assess the similarity of UDOM samples based on the relative abundance of identifiable pyro-products using HCA. Such analysis clearly separated the
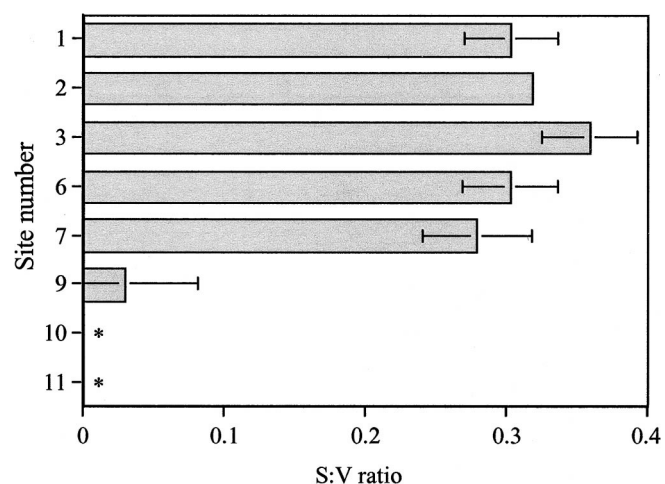
dataset into two broad groups, namely, TS and FB (Fig. 6). The riverine zone is further clustered into subgroups, where the canal inflow point (site 1) is clearly different from the remaining TS sites (sites 2, 3, 6, and 7). Furthermore, mangrove-influenced sites (sites 6 and 7) fell into a small cluster. While the three FB sites formed a separate cluster, sites 9 and 10 correlated more closely than site 11. Waters at this latter site may contain more biodegraded DOM due to higher availability of phosphorous, which is being supplied through water exchange with the Gulf of Mexico.

Molecular characteristics of UDOM at different zones of the ecosystem—Canal overflow point (site 1): Site 1 is anchored at a canal inflow point. Canal water contains higher concentrations of phosphorous than inner marsh sites (Rudnick et al. 1999), which leads to higher microbial activity. In addition, long residence times of canal waters have been suggested to result in higher photochemical degradation (Lu et al. 2003). Molecular characteristics of DOM of site 1 may therefore be influenced by these processes. The ^{13}C NMR
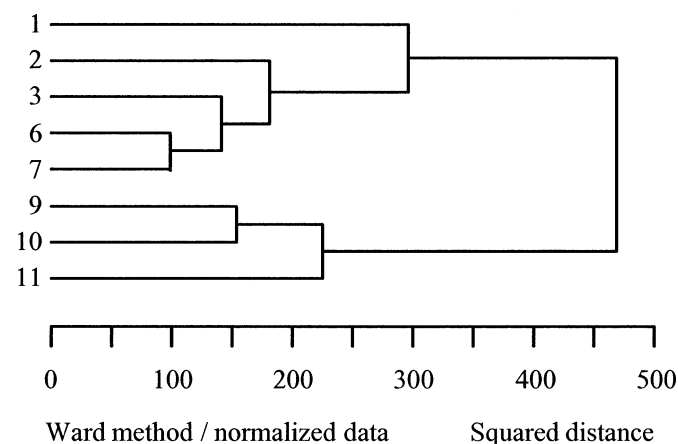

Fig. 6. Cluster analysis of UDOM from Taylor Slough and Florida Bay based on pyrolysis-GC/MS products.

showed depletion of *O*-alkyl C and enrichment of other types of C, such as aromatic C and carbonyl C (Table 2; Fig. 2), which is typical for UDOM that has undergone more diagenetic reworking (Engelhaupt and Bianchi 2001). Furthermore, very low neutral sugar concentrations at this site compared with other sites (Table 3) also support the conclusion that the DOM in this site has undergone extensive diagenesis (Amon and Benner 2003). The Py-GC/MS data show that the nature of UDOM at this site is quite different from UDOM at other riverine sites (Fig. 6), in agreement with previous suggestions by Lu et al. (2003). This shows that DOM at this canal-influenced site is unlike typical DOM in the southern Everglades and suggests that DOM from such inputs is not the predominant source at the riverine sites.

Riverine sites (sites 2, 3, 6, and 7): The higher *O*-alkyl C concentration and lower % alkyl C/% *O*-alkyl C of UDOM in TS (Table 2) compared with values reported for other rivers (Engelhaupt and Bianchi 2001; Benner 2003; Bianchi et al. 2004) seems to be characteristic of the UDOM in the FCE. Because oxidative degradation of DOM is usually accompanied by a decrease of *O*-alkyl C and production of carboxylic C (Orem and Hatcher 1987; Knicker and Lüdemann 1995; Engelhaupt and Bianchi 2001), the consistent increase of % *O*-alkyl C and decrease of % carbonyl C along the TS river section transect (Fig. 2; Table 2) suggest an enrichment of nonhumic substances that are potentially less bio- and photo-degraded. In addition, comparatively low lignin phenol concentrations expressed as V + S (<0.4 mg 100 mg C UDOM; Fig. 4) than in other aquatic environments (0.5–1.5 mg 100 mg C^{-1} UDOM for surface water in Gulf of Mexico, Bianchi et al. 1997; 0.45–1.94 mg 100 mg C^{-1} UDOM for a southern Louisiana tidal stream, Engelhaupt and Bianchi 2001; 0.30–0.90 mg 100 mg C^{-1} for Mississippi river, Bianchi et al. 2004) indicates a relatively minor contribution of lignin-derived organic matter to this UDOM pool. Orem and Hatcher (1987) reported that UDOM in pore water produced under reducing conditions was rich in alkyl C and *O*-alkyl C, while that produced under oxidative conditions contained high concentrations of alkyl C and aromatic C. They attributed the difference to the different degradability of lignin under oxidative and reduced conditions. Although the soil Eh in the ENP is usually indicative of aerobic or slightly reducing conditions (Bachoon and Jones 1992), the aromatic C concentration and aromaticity in UDOM in this area are very low but the *O*-alkyl C concentration is very high (Fig. 2; Table 2). This may be because overall microbial degradation of organic matter, including that of lignin, has been suppressed in this extremely oligotrophic environment. Alternatively, DOM rich in *O*-alkyl C leached from biomass, such as periphyton (Lu et al. 2003), has diluted lignin phenol concentrations. The high neutral sugar concentration (Table 3) corroborates that UDOM samples are rich in labile compounds and have undergone a low degree of diagenetic reworking.

DOM in the pore water of sediments is considered to be an important source of DOM in wetlands because of its high concentration compared with the surface water (Orem and Hatcher 1987). In the ENP, however, the DOC concentration of pore water is not much higher than that of surface water

(Qualls and Richardson 2003). Thus, the contribution to surface waters of pore water-derived DOM seems minor, especially in the marl soil, where organic C concentration is low compared with peat soils. Furthermore, because surface-water DOC concentrations have been positively correlated with plant primary productivity in the Everglades (Davis 1991; Qualls and Richardson 2003), plant exudates and DOM leached from senescent biomass (Engelhaupt and Bianchi 2001) are considered to be very important sources of DOM in the studied ecosystem.

FB estuary (sites 9, 10, and 11): Carbohydrates are known to comprise a major fraction of UDOM in the surface waters of ocean basins (e.g., Benner et al. 1992). Even considering that, the abundance of *O*-alkyl C in UDOM from FB was still higher than that of other aquatic environments (Table 2; Benner 2003). Our data suggested that UDOM in FB is a mixture of allochthonous (land-derived) and autochthonous (seagrass/phytoplankton-derived) UDOM. The reduction in the aromatic C concentration in FB (Table 2; Fig. 2) compared with TS indicates a production of large amounts of nonaromatic C, mainly carbohydrates, in FB. Seagrass meadows, which cover 95% of the bottom, have been the dominant vegetation historically in FB (Zieman et al. 1989). However, concurrent with the recent die off of seagrass in FB, phytoplankton, which was not as important in this oligotrophic system, has significantly increased its productivity (Fourqurean and Robblee 1999). Both seagrasses and phytoplankton are known to produce a high amount of DOC (e.g., Ziegler and Benner 1999; Bertilsson and Jones 2003), which contains a high concentration of sugars and phenolic compounds, in the case of the seagrasses (Wilson et al. 1986; Opsahl and Benner 1993). Thus, this biomass-derived DOM is considered to contribute to the UDOM in FB, although it remains unknown which is the dominant source.

The concentration of lignin-derived phenolic compounds (V+S) is about five times smaller in FB than for TS samples (Fig. 4). This might be the result of dilution of UDOM by other organic materials, such as carbohydrates. Because lignin phenols do not solely derive from terrestrial sources, but are also detected in seagrass tissues (e.g., Opsahl and Benner 1993), it is difficult to estimate the contribution of allochthonous UDOM to the overall UDOM pool in FB. Phenolic compounds are very photosensitive and thus are quickly transformed after leaching (Scully et al. 2004). Because the S : V ratio decreased for the FB sites and the concentration of phenolic compounds belonging to DV and DS were quite high in abundance, photo-induced alteration of UDOM (Opsahl and Benner 1998) might be an important biogeochemical process in FB. On the other hand, because sugars do not absorb natural sunlight, they are considered to be less susceptible to photodegradation. Neutral sugars, a major component of the carbohydrate fraction, are known to be very susceptible to microbial degradation (e.g., Amon and Benner 2003). However, microbial activity in FB is low due to the limited availability of phosphorous (Fourqurean et al. 1993; Cotner et al. 2000), particularly in the northeast and central basins (Boyer et al. 1999), and may result in a high neutral sugar concentration (Table 3). High carbohydrate productivity by seagrass and other benthic and planktonic

vegetation in FB may be also important. Based on the discussion above, high autochthonous DOM production, selective photodegradation, and low microbial activity are the main biogeochemical processes controlling the high *O*-alkyl C UDOM concentrations in FB.

In summary, the contribution of soil/sediment-derived UDOM, which is expected to have undergone extensive degradation, seems to be minor in the studied ecosystem. The major portion of UDOM in FB, which is particularly high in *O*-alkyl C, is considered to be autochthonous and derived from seagrass and/or phytoplankton, while the influence of terrestrial-derived UDOM seems minor. While microbial activity is also low in FB, photodegradation of colored DOM (see also Scully et al. 2004) is likely to play an important role in the biogeochemical cycling of UDOM in the region. Thus, local vegetation patterns and geomorphological features typical of such tropical/subtropical, coastal landscapes have an important influence on the nature of DOM, resulting in organic matter dynamics that are clearly different and possibly more complex than the two end-member systems commonly reported in more temperate climates. In addition, the tropical nature and hydrological features of these systems may exert important biogeochemical implications regarding the composition and cycling of DOM. In the FCE and other coastal areas in the tropics and subtropics, the hydrological regime varies tremendously between the dry and wet seasons. Thus, DOM characteristics and the biogeochemical processing of these materials during both wet and dry seasons need to be investigated for these systems.

This study presents, to the best of our knowledge, the first detailed molecular characterization of UDOM preformed in tropical/subtropical environments except for those for the Amazon River basin (e.g., Hedges et al. 1994 and references therein), where most of the DOM has been characterized as of highly degraded, terrestrial origin. In contrast, the data presented here suggest that tropical and subtropical costal wetlands, fringe mangrove forests, and seagrass-dominated estuaries can show enrichments in the carbohydrate fraction of the UDOM, which can be attributed to the high productivity and preservation of such labile UDOM components, particularly in oligotrophic systems. It has been estimated that wetlands occupy 6.8–8.6×10^6 km^2 (Mitsch and Gosselink 2000) and contribute about 10% of all freshwater DOC discharges into the world's oceans and, in fact, swamps and marshes have been recognized as ecosystems with the highest rate of organic carbon loss, on the order of 20 g C m^{-2} yr^{-1} (Schlesinger and Melack 1981). While tropical and subtropical wetlands account for 36–56% of total natural wetlands (Mitsch and Gosselink 2000), studies of DOM dynamics in such environments continue to be very scarce.

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