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Isotopic and biochemical composition of particulate organic matter in a shallow water estuary (Great Ouse, North Sea, England)

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Abstract

The biogeochemistry of particulate organic matter was studied in the Great Ouse estuary draining to the North Sea embayment known as the Wash from March 1990 to January 1991. Eleven locations were sampled monthly on a 50 km transect across the shallow estuary from the tidal weir to the middle of the Wash. Particulate organic carbon (POC) and total carbohydrate, protein and lipid analyses were combined with the determination of stable carbon isotopes. $\delta^{13}\text{C}$ often increased from -30‰ in the river to -22‰ in the tidal freshwater reach. The mixing zone between fresh and marine tidal waters displayed only a slight increase in $\delta^{13}\text{C}$ to -19‰ . The change in $\delta^{13}\text{C}$ values in the freshwater tidal reach demonstrated that mixing of riverborne and marine suspended POC was not the only process affecting the carbon stable isotope composition. Complementary sources, interfering considerably with the two end-member sources, may be identified as autochthonous primary production and resuspension of sediment that may be transported upstream. The respective importance of these sources is subject to seasonal variation. From March to August, high concentrations in carbohydrate and protein through the whole estuary indicate that despite turbidity significant primary production occurred. The proportional importance of the uncharacterized fraction of POC, which is considered as complex organic matter, was high from September to January and low from March to August. During most of the year, the biochemical compositions of particulate organic matter in the turbidity maximum and the rest of the estuary were similar. This contradicted the principle that owing to the long residence times of particles degradation processes largely dominate the production processes within the turbidity maximum. The occurrence of significant in situ production in such shallow water estuaries may partially compensate for the degradation of suspended particulate organics, resulting in a complex relationship between the biogeochemical cycling and the fate of nutrients.

Introduction

Estuaries are complex mixing zones displaying several interrelated physical, chemical or biological interfaces, where biogeochemical processes significantly affect the fate of riverborne material (Kemp et al., 1982; Saliot et al., 1984; Relexans et al., 1988). Oceanographers are concerned with determining the impact of riverborne pollutants on coastal and offshore marine ecosystems (Gerlach, 1988; Brockman et al., 1988; Jickells

et al., 1991). Eutrophication of riverine and coastal environments is a direct consequence of the increase in nutrient loading and therefore the fate of biologically important elements going through estuaries must be clarified. The first aim of this study was to determine the changes in the biochemical composition of particulate organic matter in an estuary through a seasonal cycle.

Carbon stable isotope analysis has been extensively used in estuaries as a tracer of the major sources of organic carbon (Haines and Montague, 1979; Hughes and Sherr, 1983; Gearing

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et al., 1984; Simenstad and Wissmar, 1985; Saliot et al., 1988; Lucotte, 1989). Stable carbon isotopes have also been used to quantify the mixing processes between continental and marine particulate organic carbon (POC) in estuaries where river and ocean end-members were the only sources (Fontugne and Jouanneau, 1987). However, these mixing models are open to criticism, mainly because of the doubtful accuracy of the $\delta^{13}\text{C}$ estimate for each source, possible changes in the $\delta^{13}\text{C}$ of the transported organic material, and interference from overlooked sources of organics (Fry and Sherr, 1984; Benner et al., 1987). $\delta^{13}\text{C}$ studies in an estuary may not accurately resolve all potential sources but they do provide clues about the respective influence of the main sources, the seasonal variation in export of continental or estuarine organic material and the nature of the sources that combine with the continental and marine end-members. Even though $\delta^{13}\text{C}$ is not an unequivocal tracer of the origin of particulate organic matter in most estuarine and coastal environments it can yield valuable information when combined with other analytical investigations. Most of these multiple approaches deal with simultaneous analyses of several stable isotopes (Peters et al., 1978; Sweeney et al., 1980; Macko, 1983). Combined analysis of stable carbon isotopes and biochemical composition of particulate organic matter (POM) has usually involved chloropigments, POC or particulate organic nitrogen determination (Fontugne and Jouanneau, 1987; Ember et al., 1987; Lucotte, 1989; Harden and Williams, 1989; Matson and Brinson, 1990) while further investigation on the detailed characterization of biochemical compounds is rare (Benner et al., 1987; Saliot et al., 1988). This is a rather surprising gap owing to the conspicuous complementary nature of both approaches in biogeochemical process investigations.

The study presented in this paper was aimed at determining the origin and fate of particulate organic matter in a shallow water estuary, the Great Ouse, on the British coast of the North

Sea. Determination of the major components of organic matter (POC, and total protein, carbohydrate and lipid) were combined with $\delta^{13}\text{C}$ analysis of POC from the river to the marine end-member of the estuary from March 1990 to January 1991. There are evident limitations in measuring the bulk composition of stable carbon isotopes and biochemical constituents that are discussed in this paper. Nevertheless, seasonal coverage of these parameters allows us to assess the influence of variable freshwater input to the estuary on POM composition and flux and to infer the potential significance of the diverse sources of POM. Stable carbon isotopes are especially well adapted to the study of the Great Ouse because of the marked differences between marine and terrestrial organic matter ^{13}C in temperate areas. The present work was part of a general research programme (JONUS) developed to estimate the flux and fate of nutrients through the major estuaries of southeast England and their impact on the North Sea. Particulate organic matter and dissolved inorganic nutrients are obviously related through production and degradation processes and studying POM thus represented an indispensable step in the elucidation of the cycling of nutrients in this estuarine environment.

Materials and methods

Site description

The geomorphology and the main characteristics of the Great Ouse estuary (Fig. 1), which is the main freshwater input to the Wash embayment, has been described in previous works (Gould et al., 1986; Fichez et al., 1992). The total catchment of the river system is 8380 km² and the average flow rate is 38.5 m³ s⁻¹ thus making its freshwater flow of the same magnitude as the associated freshwater flows from the Nene, Welland and Witham. Denver Sluice is a tidal weir separating the river from the tidal freshwater reach. Downstream of Denver Sluice a narrow (<70 m wide) and shallow (1–7 m

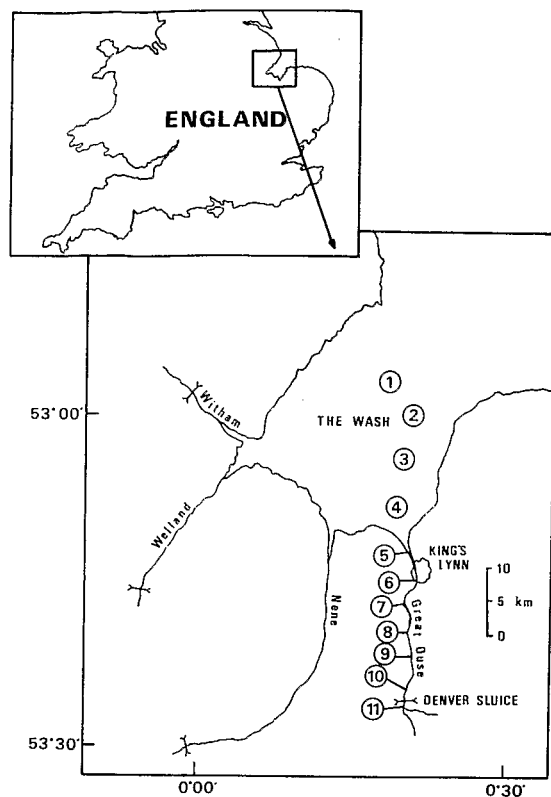


Fig. 1. Location and map of the Great Ouse estuary. Sampling stations are numbered from 11 (river) to 1 (sea) and quoted as TO11 to TO1 in the text (TO standing for Trans-Ouse).

depth at low water) channel stretches for 25 km before opening on the 600 km² Wash embayment. The four rivers (Witham, Welland, Nene, Great Ouse) draining to the Wash may be classified as highly eutrophic as they yield high loads of inorganic nutrients resulting in high productivity. Agricultural activities are mainly responsible for the high level of nitrate and phosphate (400–600 $\mu\text{mol l}^{-1}$ and 40–80 $\mu\text{mol l}^{-1}$, respectively; Fichez et al., 1992) in the river. A monthly averaged freshwater flow of 110 $\text{m}^{-3} \text{s}^{-1}$ was measured at Denver Sluice in February (NRA, unpublished data). It decreased to 38 $\text{m}^{-3} \text{s}^{-1}$ and 19 $\text{m}^{-3} \text{s}^{-1}$ in March and April, respectively, to reach minimum values of 3–5 $\text{m}^{-3} \text{s}^{-1}$ from June to September and increased sharply after October (9 $\text{m}^{-3} \text{s}^{-1}$) to maximum values (110 $\text{m}^{-3} \text{s}^{-1}$) in January.

Sampling and sample preparation

Water was collected at 1 m depth at 11 sampling locations (TO1–TO11) on a 50 km long transect along the estuary from Denver Sluice (TO11) to the Roaring Middle buoy (TO1) in the centre of the Wash. A monthly survey was done during high water at neap tide from March 1990 to February 1991.

Water samples were filtered in the laboratory on precleaned (500°C, 4 h) Whatman GF/C glass fibre filters (Whatman International, Maidstone, UK) under moderate vacuum within a few hours of collection. Loaded filters were dried (50°C, 48 h) and stored in the deep freeze prior to analysis. Salinity was measured at each sampling location.

$\delta^{13}\text{C}$ analysis

Mass spectrometry of carbon stable isotopes is performed on CO_2 gas samples. Two different methods for producing CO_2 from organic carbon are classically described in the literature, both of them displaying advantages and drawbacks. The first method consists of submitting the carbonate fraction to acid (usually HCl) attack and drying the remaining organic material which is then combusted at 900–1000°C in the presence of oxygen provided from diverse sources (copper oxide or direct oxygen gas flow) (Fontugne and Duplessy, 1978; Jeffrey et al., 1983; Macko, 1989). High temperatures ensure that the organic carbon is completely combusted to CO_2 . The disadvantage is that hydrolysis of some organic molecules may occur during acid attack. Moreover, acidified sediments or suspended material often need to be rinsed prior to combustion (Rau et al., 1982; Ember et al., 1987), and such a process may contribute to a significant loss of organic material (Byers et al., 1978). In the second method, the raw sample is directly combusted at a moderate temperature (500–600°C, 0.5–2 h) in the presence of excess oxygen (Sofer, 1980; Hackney and Haines, 1980; Hughes and Sherr, 1983; Fry et al., 1984). Inorganic carbon is not oxi-

dized at temperatures below 500°C (Hirota and Szyper, 1975; Kristensen and Andersen, 1987) and the organic carbon fraction alone is oxidized to CO₂ gas. Unfortunately, such a temperature-based differentiation between inorganic and organic carbon seems to be only theoretical and there is no doubt that a significant loss of inorganic carbon occurs at 550°C (Hirota and Szyper, 1975; Byers et al., 1978) while some refractory organic material remains uncombusted at 500°C (Fröelich, 1980; Kristensen and Andersen, 1987).

It is certain that neither method is completely satisfactory. However, we found that the second method had a significant advantage as the moderate-temperature technique does not require quartz glassware, thus significantly reducing the expense for routine determination of the ¹³C : ¹²C ratio. Furthermore, the method also significantly decreases the blank level when compared with the high-temperature combustion technique (Chesselet et al., 1981). Precombusted filters are cleaned of organics and are not further affected by the moderate-temperature combustion step thus giving almost undetectable blanks at the spectrometer detection limits. Such advantages persuaded us to select the moderate-temperature combustion technique for the routine preparation of organic CO₂ samples.

Filters or sediment samples were sealed under vacuum in pyrex glass tubes in the presence of an excess of copper oxide. The tubes were progressively heated to 500°C in a muffle furnace, kept at 500°C for 2 h and cooled down progressively. Progressive heating and cooling minimized the occurrence of glass-tube cracking and the subsequent loss of the sample. Purification of the gas sample was achieved on a vacuum cracking line equipped with cold traps for the separation of H₂O and CO₂. Isotope ratios were measured on a V.G. Isogas SIRA 2 (V.G. Instruments, Middlewich, UK) isotope mass spectrometer. The ratios are expressed as δ¹³C in ‰ deviation from the Peedee belemite (PDB) carbonate standard (Craig, 1957). The analytical reproducibility for the δ¹³C composition of internal

graphite standards was within ± 0.1 ‰ and the natural sample variation was in the range of ± 0.5 ‰ for suspended particles and ± 0.4 ‰ for sediment.

Biochemical composition

Total carbohydrate (CH), protein (PR) and lipid (LI) contents of suspended particles were analysed together with δ¹³C determination. Carbohydrates were analysed according to Dubois et al. (1956) and expressed as glucose equivalents. Proteins were analysed according to Lowry et al. (1951) and expressed as bovine sero-albumine equivalents. Lipids were extracted according to Bligh and Dyer (1959), analysed according to Marsh and Weinstein (1966) and expressed as tripalmitic acid equivalents.

Techniques for the analysis of organic compounds have been discussed extensively (Dawson and Liebezeit, 1981; Williams, 1985). The choice for the analytical techniques used in this study arose from previous works on suspended or sediment organic matter (Fichez, 1991a, b) and proved to be suitable for the analysis of estuarine POM (Moal et al., 1985; Poulet et al., 1986). Chemically assessing the bulk amount of organic compounds is subject to several biases, such as the importance of the oxidation of macromolecules owing to the reaction reagents, the accuracy of the chemical identification of molecules or molecular bonds and the choice of the standard. The relative lack of accuracy of each bulk analysis is balanced by the advantage of simultaneously determining the main categories of organic compounds constituting POM. Such simultaneous measurements of the three main classes of compounds along with the bulk organic carbon content yield an estimate of the gross composition of organic matter (Spitzky and Ittekkot, 1991). To assess the composition of POM, carbohydrate, protein and lipid concentrations were converted to carbon using conversion factors of 0.45 gC g⁻¹, 0.50 gC g⁻¹ and 0.70 gC g⁻¹, respectively, according to the composition of the standards used and the natural com-

position of particles (Degens, 1970; Jeffrey, 1970; Saliot et al. 1984; Fukami et al., 1985). It must be emphasized that average conversion factors neglect the diversity in the composition of organic compounds (Fichez, 1991b). However, such a conversion step makes it possible to calculate the residual fraction of organic carbon which is not accounted for by carbohydrate, protein and lipid. This fraction, described as heterogeneous, polyfunctionalized and macromolecular in nature (Gough and Mantoura, 1990), we term complex organic matter (COM) owing to both its biochemical complexity and poorly described molecular composition and origin (Fichez, 1991a,b). Distinct from the uncharacterized COM fraction, the pool of carbohydrate, protein and lipid compounds is referred to as the CPL fraction.

Results

–28.5–28.5 $\delta^{13}\text{C}$

The $\delta^{13}\text{C}$ of suspended particulate organic matter was plotted together with salinity for the transect along the estuary (Fig. 2). The need for such a presentation, instead of usual plots of $\delta^{13}\text{C}$ against salinity, arose from the physical conditions in the upper estuary (tidal freshwater reach) where tidal freshwater extended several kilometres downstream of the sluice during autumn and winter surveys.

There is a general increase of $\delta^{13}\text{C}$ values from –30 ‰ in the river upstream of Denver Sluice to –19 ‰ in the centre of the Wash which could be considered as the effective marine end-member of the estuary. In March the $\delta^{13}\text{C}$ value in the river end-member was –30 ‰, it increased slightly to –28.5 ‰ in the tidal freshwater reach and progressively increased to –20.4 ‰ at TO3 (35 km). The $\delta^{13}\text{C}$ decreased to –22.1 ‰ in the more offshore part of the transect without any associated change in salinity (ca. 33 psu). The April profile showed a similar trend except for the drop in $\delta^{13}\text{C}$ at TO5 (25 km) which is

explained below. During most of the following months there is a steep increase in $\delta^{13}\text{C}$ in the freshwater reach, the extent of which is receding from 12 km in May to 2.5 km in August. For example, $\delta^{13}\text{C}$ values increased from –30 ‰ in the river to –22 ‰ throughout the tidal freshwater reach in May; this increase is not related to the mixing of fresh and marine waters. Within the region of the salinity gradient there was a slight increase of $\delta^{13}\text{C}$ up to a value of –19 ‰ which persists from this part of the estuary as far as the middle of the Wash. In summer, $\delta^{13}\text{C}$ values increased in the river to –28.5 ‰ in July and –26 ‰ in August. Profiles changed markedly by the September sampling with a 8 km downstream shift of the salt wedge together with a strong decrease of $\delta^{13}\text{C}$ in the river (–33 ‰). $\delta^{13}\text{C}$ values rose rapidly (–28.4 ‰) at the very beginning of the tidal freshwater reach then progressively increased to –22 ‰ together with the mixing of fresh and marine water. During autumn and winter, river POC $\delta^{13}\text{C}$ increased from –29.5 ‰ in October to –26 ‰ in January. Except for January, $\delta^{13}\text{C}$ values in the tidal freshwater reach were close to –22 ‰, slightly decreasing downstream. The January data displayed a rather different shape from all other samplings, $\delta^{13}\text{C}$ values progressively increasing from –26 ‰ in the river to –22 ‰ at the lower limit of the freshwater reach, decreasing to –24.2 ‰ in the fresh–marine water mixing zone and increasing to –21.8 ‰ in the marine part of the estuary.

An exceptional drop in $\delta^{13}\text{C}$ values was observed in April at TO5 (25 km) which has to be explained. This specific sampling was made at the beginning of the ebbing tide in front of the King's Lynn sewage outfall. At this time of the tide the gates of the sewage plant are opened to discharge the effluent into the estuary where it is then flushed to the sea by the flowing water. The input of anthropic organic material depleted in ^{13}C is clearly identified by the 6 ‰ drop in the $\delta^{13}\text{C}$ value which is localized at TO5 due to the limited dilution of the plume at the beginning of the release process.

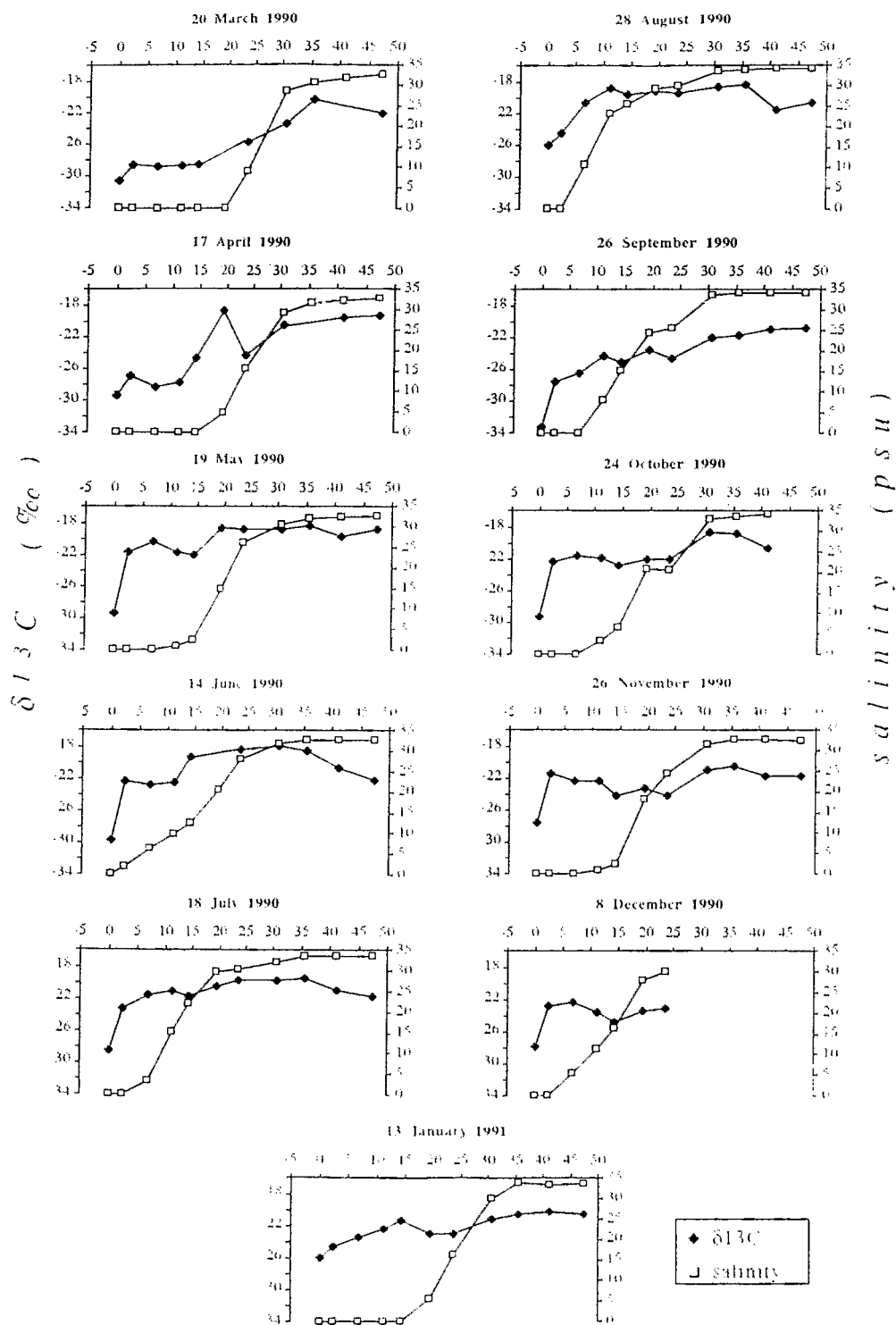


Fig. 2. Axial distribution of $\delta^{13}\text{C}$ and salinity within the Great Ouse estuary. $\delta^{13}\text{C}$ in ‰ and salinity in psu on y-axis, distance in kilometres from the riverine end-member sampling point (TO1) upstream of Denver Sluice on the x-axis.

Biochemical composition

Particulate organic carbon (POC), carbohydrate, protein and lipid concentrations (Table 1) increased from the river to the salt wedge where the turbidity maximum occurred then decreased seaward. Particulate organic carbon concentrations were high in May and maximum during winter. In contrast to POC, concentrations in CPL were high during the first 7 months of the survey and decreased during the last 4 months which covered the end of autumn and winter. In the river end-member concentrations ranged from 96 to 1995 $\mu\text{g l}^{-1}$ for protein, 76 to 1725 $\mu\text{g l}^{-1}$ for carbohydrate and 41 to 1030 $\mu\text{g l}^{-1}$ for lipid. In seawater end-member maximum concentrations were recorded in May for carbohydrate, protein and lipid with especially high values in the range 2000–3600 $\mu\text{g l}^{-1}$ for carbohydrate. Generally proteins were the most important of the three identified organic species and carbohydrates ranked second while lipids, usually far below, ranked third. Maximum concentrations on the estuarine transect were located in the upper or middle estuary always corresponding with the POC maximum and the turbidity maximum. The strong decrease in concentrations at the mouth of the estuary (TO5), owing to dilution of estuarine waters into the Wash, is a signal for the transition from the tidal channelled estuary to the Wash embayment.

An anomaly in this general decreasing trend occurred in April as concentrations of organics dramatically increased at TO5; this is related to the sampling occasion described in the previous section which involved the King's Lynn sewage effluent. This outfall strongly affected the concentration and composition of POM with an especially large increase in lipid concentration. The influence of the sewage was not the subject of this study and such sampling was not repeated during the following months and will not be discussed in detail. However, it must be emphasized that even though the sewage release is a process confined to the beginning of the ebb

tide period, the amount of organic material discharged daily at the mouth of the estuary may be of the same magnitude as the organic material flushed by the estuary during low freshwater-flow periods (summer). Sewage could thus have a major impact on the biogeochemical cycling and the structure of the benthic communities of the surrounding sediments if not on the biogeochemical budget of the estuary.

The biochemical composition of organic matter expressed as a percentage of organic carbon has been plotted against the distance from Denver Sluice (Fig. 3). The uncharacterized fraction of organic carbon which is considered as complex organic matter (COM) represented the largest pool for organic carbon. Nevertheless, from March to July the CPL fraction represented more than 50% of the POC for most of the sampling. From August to January the CPL fraction decreased dramatically progressively reaching values below 10%, COM originating carbon thus accounting for the complementary 90%. An unusual situation was observed in March, lipid accounting for less than 5% of organic carbon in the upper estuary and increasing to almost 50% in the lower estuary and the Wash.

Discussion and conclusions

Carbon isotopic mixing models applied to estuarine studies are mostly based on two sources of suspended particulate organic matter: the riverine and the marine end-members. However, the data from the Great Ouse estuary are not consistent with this simple two end-members mixing model. Given the $\delta^{13}\text{C}$ values of the river and marine end-members of the Great Ouse (-30 and -19 ‰) and allowing for the generally limited isotope fractionation (0 to -2 ‰) owing to degradation processes (Schwinghamer et al., 1983), the strong increase in $\delta^{13}\text{C}$ that occurred in the tidal freshwater reach suggests the presence of at least one more source of POC (Fry and Sherr, 1984).

Table 1

Biochemical composition of particulate organic matter in the Great Ouse estuary following a transect from the river (TO11) to the sea (TO1, 47.6 km downstream TO11) from March 1990 to January 1991. POM, particulate organic matter; POC, particulate organic carbon; CH, carbohydrates; PR, proteins; LI lipids. Concentration in micrograms per litre

Date	Component	Location										
		TO11	TO10	TO9	TO8	TO7	TO6	TO5	TO4	TO3	TO2	TO1
		Distance (km)										
		0.0	2.4	6.7	11.2	14.2	19.5	23.7	30.6	35.6	41.0	47.5
20/03/90	POC	2407.3	3742.5	4015.1	4566.1	6440.5	5846.3	1696.1	498.5	460.9	529.4	329.8
	CH	618.9	1210.9	1641.7	2417.0	2723.6	1833.2	502.7	134.3	125.8	130.5	67.7
	PR	1020.8	2009.7	2449.2	3630.0	4051.1	3179.2	823.8	278.1	288.8	271.8	162.8
	LI	120.8	230.4	232.3	354.2	448.1	346.6	91.9	146.4	183.1	231.1	223.3
17/04/90	POC	1928.6	3774.1	4495.5	3112.1	1738.2	1807.1	9424.6	587.1	326.0	478.7	288.4
	CH	709.7	1216.1	1065.3	906.9	489.4	627.7	1779.0	214.6	198.5	212.9	112.6
	PR	1041.1	2105.2	2241.5	1559.5	923.0	1124.8	3739.6	480.5	262.1	439.4	231.7
	LI	523.4	821.2	892.1	645.7	267.7	381.3	5268.5	127.1	66.59	107.2	55.9
19/05/90	POC	3066.6	6729.2	7109.2	4051.8	2420.5	3234.6	5485.5	2549.3	2421.6	2423.3	1984.0
	CH	1300.0	2694.0	2026.0	1407.0	1072.8	958.5	2980.0	2002.9	2590.0	2475.7	2125.7
	PR	950.0	3533.3	3204.2	1962.5	1360.0	1879.2	3845.8	1410.7	1363.1	985.1	934.5
	LI	689.1	1181.4	1052.5	690.2	421.0	363.8	1004.9	381.6	391.6	371.6	375.6
14/06/90	POC	1440.6	8156.5	4016.0	3137.6	4629.5	3121.8	2852.0	730.2	701.4	525.1	735.5
	CH	467.6	3750.0	1647.7	1120.0	1370.0	951.4	776.7	186.9	232.8	156.4	254.4
	PR	855.4	3916.7	1933.7	1241.7	1733.3	1221.4	1184.3	449.6	356.7	294.7	254.3
	LI	290.4	1211.0	756.0	677.7	640.7	392.0	365.9	118.1	113.8	123.9	172.5
18/07/90	POC	3550.2	20337.0	7373.5	6407.7	5105.2	4204.5	2806.6	331.8	1055.7	229.9	229.5
	CH	1725.0	5366.7	2876.7	1625.0	3014.0	1361.7	830.8	143.5	213.4	118.2	108.9
	PR	1078.0	5527.8	4191.7	3502.1	2588.3	1720.8	1173.6	232.8	468.9	180.9	144.1
	LI	754.3	1967.4	1105.1	919.9	916.3	622.6	470.1	86.6	162.9	63.6	43.7
28/08/90	POC	6763.4	5387.0	3878.0	2463.8	1523.9	1117.9	1342.8	638.9	368.2	190.9	142.3
	CH	1405.0	1020.0	614.7	370.8	208.6	171.3	148.6	198.6	105.7	77.8	32.2
	PR	1995.2	1504.7	1590.5	700.0	420.3	405.7	317.1	143.4	134.3	34.3	29.9
	LI	1030.0	522.3	534.6	317.7	172.0	102.5	97.3	85.5	55.3	45.1	22.0
26/09/90	POC	2065.5	3787.3	6766.0	6689.0	2695.4	1631.0	1469.1	608.9	273.8	226.6	215.1
	CH	751.5	950.0	1429.4	1127.5	500.0	224.4	295.5	147.3	75.5	67.4	82.0
	PR	385.7	1050.8	966.9	1342.9	229.2	172.0	266.1	85.3	70.3	51.5	50.4
	LI	251.2	404.7	431.0	453.3	141.7	95.5	128.6	45.6	28.1	35.2	33.3
24/10/90	POC	684.5	4239.0	8594.0	12078.0	6844.0	3017.8	5445.0	781.5	789.7	480.9	–
	CH	130.4	634.0	1370.8	1990.0	1026.3	610.0	790.0	149.0	138.8	71.1	–
	PR	251.4	731.0	1318.7	4257.2	1263.3	499.1	2193.7	169.3	164.15	129.5	–
	LI	95.5	210.3	387.1	978.2	243.8	189.4	214.5	105.0	87.1	63.7	–
26/11/90	POC	1081.8	12231.5	18081.5	12494.0	9547.0	3826.8	2401.3	1178.6	1055.2	1056.6	678.5
	CH	351.8	1455.0	1482.1	2271.4	1070.0	730.0	338.8	94.0	140.8	75.2	98.4
	PR	179.6	1782.3	2278.9	963.3	1464.0	850.0	406.3	99.6	129.2	74.6	66.5
	LI	88.1	348.1	330.0	299.0	210.5	168.2	104.2	80.5	130.0	55.1	66.2

Table 1 Continued

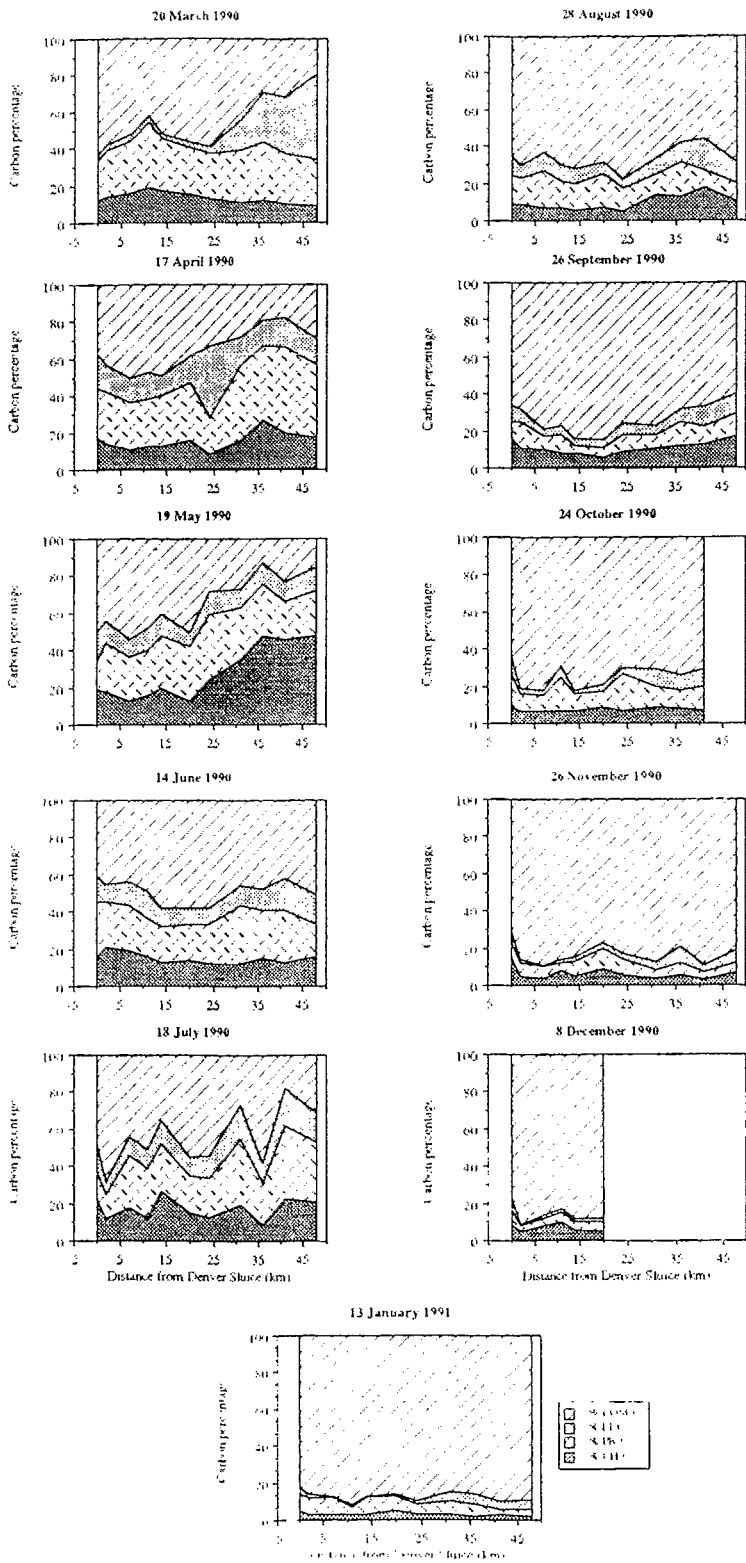
Date	Component	Location										
		TO11	TO10	TO9	TO8	TO7	TO6	TO5	TO4	TO3	TO2	TO1
		Distance (km)										
		0.0	2.4	6.7	11.2	14.2	19.5	23.7	30.6	35.6	41.0	47.5
08/12/90	POC	628.4	21277.5	14848.5	5738.1	2645.2	–	–	–	–	–	–
	CH	110.2	1242.9	1416.3	1518.8	561.0	293.6	–	–	–	–	–
	PR	96.24	2327.4	3006.0	2994.0	671.5	248.5	–	–	–	–	–
	LI	59.4	412.0	390.4	322.0	150.2	61.3	–	–	–	–	–
13/01/91	POC	654.4	4937.0	8078.3	16312.0	9938.5	4196.7	2677.5	770.4	1477.6	1248.8	1578.1
	CH	76.3	373.5	474.7	1068.8	733.8	482.5	181.3	52.5	65.8	81.5	65.6
	PR	114.4	914.3	1406.3	1165.9	1863.1	711.9	283.8	108.4	172.1	71.3	113.7
	LI	41.3	125.6	112.7	233.3	173.2	87.7	61.0	53.2	122.0	78.2	109.1

A first possible source could be the detritic non-living sediment organic material as it may account for a large fraction of the suspended POM in the upper reaches of tidal shallow water estuaries, mainly because of resuspension processes and the long residence times of particles (Eisma and Cadée, 1991). The occurrence of a $\delta^{13}\text{C}$ anomaly in the tidal freshwater reach corresponded to the progressive formation of a suspended particulate matter (SPM) maximum (from $100 \mu\text{g l}^{-1}$ in May to $400 \mu\text{g l}^{-1}$ in December) while its decrease (below $50 \mu\text{g l}^{-1}$ in March) corresponded to the partial flushing of this SPM maximum to the sea by increasing river flow (Fichez et al., 1992). $\delta^{13}\text{C}$ values for sediment POC in the upper estuary of the Great Ouse (salinity 0–10 psu) were in the range -21.7 to -20.3‰ (R. Fichez, unpublished results, 1991), thus demonstrating the benthic boundary layer to be a significant potential source of POM enriched in ^{13}C . Values of $\delta^{13}\text{C}$ higher for the sediment than for SPM indicate that marine sedimentary organic material may be transported upstream. Such a transport, related to the tidal currents at the benthic boundary layer, has already been suggested through stable isotope study (Mook and Tan, 1991).

Additionally, despite a high level of turbidity, evidence for significant primary production in

the upper reaches of the Great Ouse estuary from March to July has been presented in a previous work (Fichez et al., 1992). Autotrophic production, even in the most turbid part of the estuary, is made possible for a large period of the year (March–July) by the turbulent motion of particles in such shallow waters as those from the Wash estuaries. Despite the long residence times of particles primary production is sustained in the tidal freshwater reach and for each transect few spatial changes resulted in the biochemical composition of POM. Adaptation of phytoplankton cells to low light intensities is known to decrease enzymatic isotope fractionation subsequently increasing the $\delta^{13}\text{C}$ value of the algae (Mortain-Bertrand et al., 1988; Descolas-Gros and Fontugne, 1990a, b). Autochthonous primary production thus could be a source of POM enriched in ^{13}C in the freshwater reaches of shallow turbid estuaries.

The influence of both autotrophic production and sediment resuspension combined with two end-members mixing yields a complex pattern of the spatial and temporal distribution of $\delta^{13}\text{C}$. Even if the analysis of $\delta^{13}\text{C}$ is of limited value in the study of systems with more than two organic sources (Fry and Sherr, 1984) it is of considerable interest in giving evidence of the multiplicity of these sources as well as in pro-



ducing some clues to the identification of potential organic sources. From the results obtained in this study we are able to show that POM composition does not depend on a simple water mixing process alone but also on other environmental conditions (light penetration, particle residence time, resuspension).

The rise in $\delta^{13}\text{C}$ to typical marine values at the mouth of the estuary, 30 km downstream of Denver Sluice, demonstrates that, even though dilution has a major impact on estuarine POC, little direct export of continental POC occurs for most of the year, as concluded in several studies on estuaries (Salomon and Mook, 1981; Tan and Strain, 1983; Gearing et al., 1984; Lucotte, 1989). Nevertheless, the progressive increase in $\delta^{13}\text{C}$ values from the river to the sea in March, September or January could indicate that some outwelling fluxes of POM do occur but it must be emphasized that stormy events may also be responsible for a significant shift in SPM $\delta^{13}\text{C}$ at sea.

We cannot give an unambiguous explanation for the few ‰ drop in $\delta^{13}\text{C}$ leading to the values of around -22 ‰ occasionally measured in the middle of the wash. An input of continental organic material through the plume of the large Humber estuary to the Wash is very unlikely, particularly as this trend was clearly observed at the end of spring and during summer when river flows are low. This anomaly was first observed in March and corresponded to high concentrations of lipid, which are known to be very depleted in ^{13}C . However, from June to the end of the year lipids were low but COM increased markedly. We thus suggest that both lipid and COM may be partially responsible for the shift in $\delta^{13}\text{C}$ values in the middle of the Wash. Additionally, the influence from the tidal flats located at the mouth of the estuary (between 25 and 35 km downstream from the sluice) may

contribute to a moderate increase in $\delta^{13}\text{C}$ at stations TO4 and TO3 when compared with TO2 and TO1 which are located in the middle of the Wash. To test this hypothesis, sediments from the Wash embayment and the tidal flats will be analysed for their stable isotope compositions.

The sharp decrease in the $\delta^{13}\text{C}$ ratio of the terrestrial organic material from -26 ‰ in August to -33 ‰ in September may be related to the increase in the river flow with monthly averaged values of $3\text{ m}^3\text{ s}^{-1}$ in August and $5\text{ m}^3\text{ s}^{-1}$ in September (National Rivers Authority (NRA), personal communication). After a long period of relative dryness and proliferation of planktonic and benthic algae in the ponds and agricultural drains surrounding the river system, strong rainfall washed this organic material to the river and through the estuary to the sea. Evidence for the strong depletion of ^{13}C in the terrestrial organic material flushed to the estuary arises from the very negative and marked seasonal fluctuations in $\delta^{13}\text{C}$ values measured in standing fresh water (Rau, 1978; Fry and Sherr, 1984; Mook and Tan, 1991). The downstream extent of the several ‰ drop in $\delta^{13}\text{C}$ in September as compared with August indicates that the export of continental organic material reached 35 km downstream of Denver Sluice. However, the two sampling stations located in the middle of the Wash (TO1, TO2) were not affected by this outwelling of continental organic material.

The significant importance of COM in riverine and estuarine organics, is already well known (Laane, 1983; Berger et al., 1984; Saliot et al., 1984; Ittekkot, 1988). This has been confirmed in this study as carbon associated with COM commonly accounted for more than 40% of bulk POC. Complex organic matter is generally considered to be highly refractory to decomposition and to display a conservative behaviour in estuaries (Courtot et al., 1985; Ittekkot, 1988; Spitzky and Ittekkot, 1991). We did not find any strong increase in the proportional importance of COM in the turbidity maximum as shown in

Fig. 3. As for Fig. 2 for the proportional particulate organic carbon content (%) of the diverse biochemical compounds: carbohydrates (CH), proteins (PR), lipids (LI) and the remaining fraction of POC which is assumed to be complex organic matter (COM).

the Loire estuary (Saliot et al., 1984). From March to July the regular decrease in the importance of COM as a carbon source from the river to the middle of the Wash agreed with other studies demonstrating the uncharacterized fraction of organic matter to be higher in river than at sea (Ittekkot and Zhang, 1989; Spitzzy and Ittekkot, 1991). However, from August to January no significant differences were observed from the river to the middle of the Wash, thus demonstrating the increasing importance of the humic fraction in marine POC.

Carbohydrates peaked during the phytoplankton bloom periods, especially during May in the marine part of the estuary, corresponding to a classical feature in phytoplankton growth (Anonymous, 1985; Mayzaud et al., 1989). Proteins, or more restrictively amino acids, have been reported to be of insignificant importance during winter periods but to represent up to 69% of organic matter in phytoplankton blooming (Laane, 1983). Nevertheless, our results showed protein carbon to represent a significant fraction of POC during most of the survey as well as being the main characterized organic compound ahead of carbohydrate and lipid. A similar high content of proteins or amino acids in POM have been described for riverine (Wafar et al., 1989) and estuarine systems (Saliot et al., 1984; Poulet et al., 1986; Relexans et al., 1988) and it is possible to question whether or not the excess of N nutrients is of importance in the context of protein production.

Turbidity maxima are generally accepted as locations for heterotrophic decomposition of POM produced upstream, as the residence time of particles is high and light penetration low (Kranck, 1979). However, the behaviour of POM in the Great Ouse estuary may be regarded as a possible example of how other shallow tidal estuaries function. Furthermore, it may be inferred from this study that in shallow water and turbid estuaries the organic and inorganic reservoirs are more tightly and intricately related than has been assumed.

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