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Molecular and isotopic tracers used to examine sources of organic matter and its incorporation into the food webs of San Francisco Bay

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Abstract

Multiple indicators (Chl a, C : N ratios, [δ^{13}C]POC, and two classes of lipid biomarker compounds—sterols and phospholipid ester-linked fatty acids) were used to evaluate spatial and temporal variations in the origin of particulate organic matter (POM) in the San Francisco Bay (SFB) estuary. Comparisons were made between the northern and southern subestuaries of SFB, as well as along the salinity gradient of northern SFB. Two sample types were collected—seston, which was used to characterize the bulk POM, and tissues of the suspension-feeding bivalve *Potamocorbula amurensis*—in order to evaluate the assimilable portion of the POM. Samples were collected around biological and physical events (phytoplankton blooms and freshwater inflow) thought to be the primary mechanisms controlling temporal variability in organic matter sources. Seston samples indicate that phytoplankton sources of POM are important throughout the entire SFB system, with additional inputs of organic matter from bacterial and terrestrial vascular plant sources delivered to the northern region. Analysis of biomarker compounds in P. *amurensis* tissues indicates that phytoplankton supply a large fraction of the assimilable carbon to clams throughout SFB, although isotopic analysis of clam tissues suggests that the origin of this reactive carbon varies spatially and that freshwater algae are an important source of reactive organic matter to clams living in northern SFB.

Estuaries are characterized by abundant and diverse sources of organic matter including inputs from a variety of marine and terrestrial origins, each of which vary spatially along the estuarine salinity continuum. In addition to this spatial variability in organic matter sources, the dynamic nature of the estuarine environment results in temporal changes in the delivery of organic matter. Temporal variability exists over time scales as short as minutes to hours (e.g. in response to tidal resuspension or advection). Changes also occur over seasonal and annual time scales as well as over geologic time. These spatial and temporal variations give rise to fluctuations in the abundance and composition of reactive organic matter, including that assimilated by heterotrophs and incorporated into pelagic and benthic food webs.

Estuaries act not only as sites of exchange between the reservoirs of terrestrial and oceanic organic matter but also as active zones where dissolved and particulate materials are produced, transformed, or removed by physical and biological processes. These environments are characterized by high levels of biological production and are also active sites of heterotrophic metabolism (Smith et al. 1991). For example, biologically important compounds containing C, N, 0 , and S are transformed in response to a variety of heterotrophic processes (oxygenic respiration, denitrification, sulfate reduction, methanogenesis). Because these biogeochemical transformations require chemical energy in the form of labile organic matter, a fundamental issue in estuarine science involves understanding the origin, abundance, and reactivity of the organic matter that sustains these characteristic high levels of heterotrophic activity.

Although the pool of organic matter in estuaries is typically large, it consists primarily ofrefractory components or constituents which are unreactive relative to the time required for them to be flushed out of the system (Day

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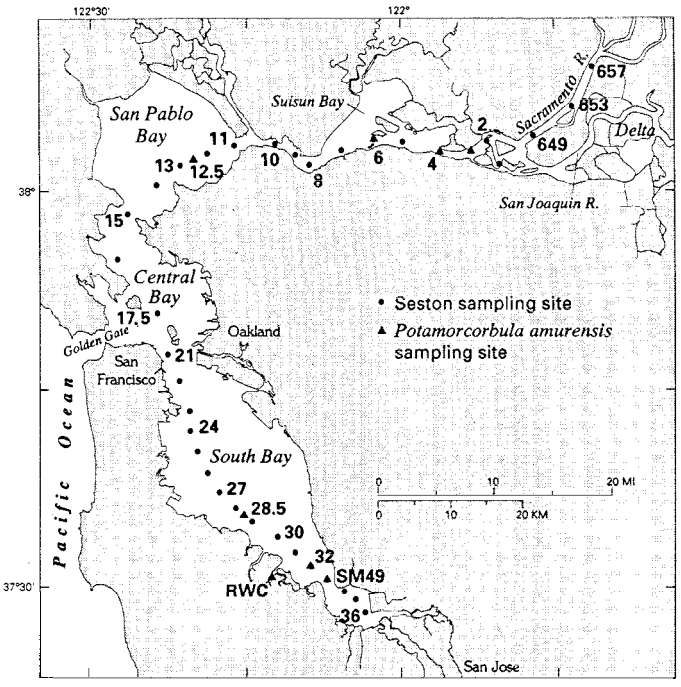

Fig. 1. Map of San Francisco Bay indicating sites where seston and P. *amurensis* samples were collected. The bay is divided into northern and southern regions by the Golden Gate. The northern region (NB) includes the lower Sacramento River (to Sta. 657), Suisun Bay, and San Pablo Bay; South Bay (SB) is the region to the south of the Golden Gate.

et al. 1989). As a result, only a portion of the total organic matter supports heterotrophic activity. Unfortunately, it is difficult to characterize the origin, chemical nature, and abundance of the labile pool of organic molecules which are assimilated and utilized by heterotrophs. Previous estuarine studies have applied various techniques, including the analysis of stable isotopes (Haines 1977; Spiker and Scheme1 1979; Peterson et al. 1985), lignin oxidation products (Reeves and Preston 1989), or lipid biomarker compounds (Mayzaud et al. 1989), to evaluate the relative importance of these different sources of organic matter. A second complicating factor is that estuaries have many potential sources of organic matter. Estuaries can receive inputs of organic matter originating in the surrounding watershed and delivered by river inflow, organic matter produced within the river itself or produced in surrounding wetland habitats and tidal flats, and marine-derived organic matter from the adjacent coastal ocean. Autochthonous primary production by diverse plant communities (phytoplankton, benthic algae, and vascular plants) can also be an important source of organic matter. Industrial and municipal discharge may be important in some estuaries as well. Although each of these sources may contribute substantially to the input of organic matter, the relative importance of these sources may vary spatially and temporally within an individual estuary (Jassby et al. 1993).

Our study used multiple indicators (stable carbon isotopes, lipid biomarker compounds, C: N, and Chl *a)* to evaluate the origin and nature of particulate organic matter (POM) in the San Francisco Bay estuary. Two different types of samples were collected and analyzed: suspended particulate matter (seston), which was used to characterize the pools of POM within the estuary, and tissues of the suspension-feeding bivalve *Potamocorbula amurensis,* which is widely distributed throughout the bay. P. *amurensis* tissues were used to evaluate sources of labile (i.e. assimilable) organic matter used by this consumer organism.

The study site

The San Francisco Bay (SFB) estuary system, the largest coastal embayment on the Pacific coast of the U.S., receives inputs from 40% (153,000 km^2) of California's surface area (Conomos et al. 1985). The bay can be divided into two distinct hydrographic provinces—the northern bay (NB), which is strongly influenced by riverine inputs originating in the area north of the Golden Gate, and the more lagoonlike southern bay (SB) (Fig. 1). In the NB, salinities typically range from <1 psu (practical salinity units) at the San Joaquin and Sacramento River end-members to 30 psu near the Golden Gate. Freshwater inputs to the SB are limited to local runoff, sewage effluent, and the influx of waters derived from the NB during periods of high freshwater inflows that occur during winter or spring storms.

Delivery of organic matter to the bay system varies in response to local habitat, inflow of freshwater, and relative importance of marine vs. terrestrial sources of organic matter. Potential sources of organic matter include primary production by phytoplankton, benthic algae, marsh flora, and seagrasses in addition to sewage and riverine inputs. The relative importance of these sources differs significantly between the northern and southern regions of the bay (Jassby et al. 1993). For example, the largest source of organic C (OC) to the SB is autochthonous (microalgal production), and its input is strongly seasonal. Riverine dissolved organic C (DOC) and particulate organic C (POC) are the dominant allochthonous sources of OC to the NB, although the nature and reactivity of this riverine OC is largely unknown. As a result, the seasonal abundance and nutritional quality of the organic matter available to consumer organisms may vary along the watershed-ocean continuum of the NB as well as between the river-influenced NB and the lagoonlike SB. The abundance and quality of this organic matter may also vary temporally in response to short-term (Wienke and Cloern 1987) and long-term (Jassby et al. 1993) variability in the estuary itself.

Late in 1986, the Asian clam P. *amurensis* was introduced to the NB, presumably from ship ballast waters (Carlton et al. 1990). Since that time, this filter-feeding bivalve has successfully colonized the entire salinity range of the bay (<1–33 psu) and a wide variety of sediment types (Carlton et al. 1990). Abundance of this organism in the bay is extremely high, with localized densities >10,000 m^{-2} at some sites (Carlton et al. 1990). We have capitalized on the success of *P. amurensis* across the full

spectrum of bay habitats by using tissues from this organism to evaluate spatial and seasonal changes in the sources of the particulate organic matter (POM) it assimilates.

Methods

Sample collection—At various times during 1990 and 1991, seston and P. amurensis samples were collected at sites throughout the bay. Collection dates and hydrographic conditions are listed in Table 1, and station locations are shown in Fig. 1. To address spatial variability, we made comparisons between samples collected in the NB and SB subestuaries as well as along the salinity gradient (0–30 psu) in the NB. Temporal variability was addressed by collecting samples around events that we perceived as the primary mechanisms of temporal variability (i.e. phytoplankton blooms and freshwater inflow); we also sampled around seasonal cycles of production by plants and riverine inputs from the surrounding watershed. The sampling included a broad range of seston abundance and composition, particularly with respect to the relative abundance of the organic and mineral fractions. This variability was derived from riverine input of suspended particulate matter (SPM) during flood events, wind-wave resuspension of the bed sediments, and a large phytoplankton bloom in spring 1990.

We collected samples representing three conditions: baseline, bloom (Chl a >10 μg liter^{-1}; SB only), and flood (daily river flow $>1{,}000$ m^3 s^{-1} during the 2-week period before sampling). However, the sampling spanned a period of unusually low river flow, so the "flood" events sampled here (peak daily flows of 1,500 m^3 s^{-1}) were small compared to floods observed during wet years ($>5{,}000$ m^3 s^{-1}). In cases where data are available for multiple cruises covering one of these events, an average value was calculated. Our aim was to sample these events and evaluate their importance in changing the quality of POM in the water column and to determine whether this variability is reflected in the tissues of the consumer organism, P. amurensis.

Water samples were collected at fixed sampling locations in the deep central channel (10–20 m deep). Near-surface water samples (1 m) were collected with a Niskin bottle and screened with 200-pm polyester monofilament (PeCap). Seston was collected from these water samples by vacuum filtration through precombusted glass-fiber filters (Gelman GF/AE) which were then frozen. Sediments were collected with a Van Veen grab sampler, and clams were collected by wet sieving the sediments. Clams were shucked, the gut and hepatopancreas removed by dissection, and the remaining soft tissues stored refrigerated in 4% Formalin. Experiments with radiolabeled formaldehyde indicate that it is removed during sample preparation and therefore does not interfere with the lipid analysis (Federle and White 1982). Formalin preservation has been shown to decrease $\delta^{13}C$ by 2–3% (Mullin et al. 1984). This should be taken into account when making comparisons with other literature values; however, the relative trends should be unaffected because all clams were treated identically. Clam samples consisted of 8–10 animals per site, and attempts were made to collect individuals in the size range of 10–15 mm.

Table 1. Sampling dates and conditions.

	Peak daily low (m^3 s^{-1})*	Samples	Cruise type
17–18 Jan 90	900	seston	baseline
18 Apr 90	250	seston, clams	bloom
30–31 Jul 90	160	seston, clams	baseline?
6–7 Dec 90	290	seston	baseline
11 Mar 91	1,130	seston	flood
11 Apr 91	1,590	seston, clams	flood

* For 2-week period prior to sampling. Delta outflow index from California Dept. Water Resources.
† Small isolated bloom in San Pablo Bay (Sta. 12–13).

Lipid analysis—Clam tissues were initially ground in methanol and then extracted in a 150-ml separatory funnel by a modification of the Bligh and Dyer (1959) method, which involved the addition of phosphate buffer (White et al. 1979). The tissues were extracted for at least 3 h in the single phase. Following the single-phase extraction, chloroform and distilled water (25 ml each) were added to break the phase. Samples were allowed to separate overnight. The total lipid extract was then dried. Filter samples with collected seston were placed in separatory funnels and extracted by the modified Bligh and Dyer method described above.

A portion of this total extract (30%) was dried under N_2, and 3 ml of 5% KOH in methanol: water (80: 20) was added to the dry extract. Samples were incubated in a heating block at 60°C for 2 h. After the samples cooled, 1 ml of water and 2 ml of hexane : chloroform (4: 1) were added. Samples were mixed by vortex for 5 min. The upper phase was removed to a clean test tube and the lower phase re-extracted twice. The combined upper layers were dried. About 30 μl of BSTFA [bis(trimethylsilyl) trifluoroacetamide] were added to each vial, and samples were heated either in a block or at the gas chromatograph (GC) injector for a minimum of 30 min. Just before GC injection, samples were dried and 100 μl of chloroform added. Sterols [as trimethylsilyl (TMS) ethers] were analyzed on a HP 5880 gas chromatograph with a HP-1 column (50 m × 0.20-mm i.d.). Conditions for GC analysis are as follows: 50°C for 1 min, ramp 30°C min^{-1} to 200°C, then 10°C min^{-1} to 280°C, followed by 2°C min^{-1} to 310°C with a final hold of 5 min. Injector and detector temperatures were set at 290°C. Detection was by flame ionization, linear over a range of 10^7 with a minimum detection limit of 1 pg s^{-1}.

The remainder of the total lipid extract was separated into neutral, glyco-, and polar lipids on a silicic acid column by means of a series of mobile phases of increasing polarity (White et al. 1979). Fatty acids associated with the polar lipid fraction (PLFA; phospholipid ester-linked fatty acids) were further analyzed. Phospholipids are membrane lipids, and fatty acids from this class of lipids

were selected for their ability to yield information about both the uptake and the utilization of organic matter. The polar lipid fraction was dried and resuspended in 1 ml of methanol : toluene (1 : 1, vol/vol), and 1 ml of 0.2 N methanolic KOH was added. The mixture was incubated at 37°C for 15 min, pH was neutralized to 6.0 with 1 M acetic acid, and 2-ml volumes of chloroform and methanol were added. The sample suspension was mixed vigorously on a mechanical mixer for 5 min followed by centrifugation. Fatty-acid methyl esters were recovered in the organic phase.

Fatty-acid methyl esters were run on both polar and nonpolar columns with a dual column IBM GC/9630. Conditions for the polar column (Restek Rt_x-225; 60 m × 0.25-mm i.d.) were 100°C initial temperature, 10°C min^{-1} to 150°C, hold 1 min; 2°C min^{-1} to 200°C; hold 24 min. Conditions for the nonpolar column (Restek Rt_x-1; 60 m × 0.25-mm i.d.) followed the same conditions, except the final ramp was at 3°C min^{-1} to 282°C followed by a 5 min hold at the upper temperature.

Peaks were quantified relative to internal standards—methyl nonadecanoate for fatty-acid methyl esters and 5α(H)-cholestane for sterols. Peak verification was obtained by combined gas chromatography-mass spectrometry (GC-MS) with a VG Trio-3 Mass Spectrometer operated in the EI mode. Double-bond positions of the monounsaturated fatty acids were confirmed by capillary GC-MS of their dimethyl disulfide adducts as described by Nichols et al. (1986).

Elemental and isotopic analyses—Aliquots of water samples (200–1,000 ml) were passed through precombusted 47-mm GF-AE filters. The filters plus retained particles were lyophilized in the laboratory and stored frozen until analyzed. Each filter was saturated with 0.5 N HCl to remove inorganic C and then analyzed for OC and N and isotopic abundances (C) with methods described by Rau et al (1990). The ratios $^{13}C:^{12}C$ are reported as the per-mil difference between the sample ratio and the ratio of a standard (PDB):

$$\delta(‰) = [R_{sample}/R_{std} - 1] \times 1{,}000.$$

The analytical precision of these determinations was typically ±0.2% (Rau et al. 1990). Measurements of *P. amurensis* tissues were done on lyophilized individuals or subsamples thereof by means of the preceding methods.

Dissolved inorganic C (DIC) samples were collected into a 400-ml plastic beaker from the lower port of a Niskin bottle. A 60-ml disposable syringe barrel was immersed in the beaker and slowly filled from the bottom of the beaker. Fine-grained carbonate particles were excluded with a 45-pm in-line filter fitted onto the barrel before installation of a 20-gauge needle. This system was then uprighted and bled of air prior to injection through the septum of an evacuated sample bottle. Phosphoric acid (H_3PO_4) was added (0.2 ml) so that CO, (gas) was the predominant inorganic carbon species. Samples were agitated to mix the acid with the sample and CO, evolved into the headspace. Each sample underwent five separate gas expansions on a vacuum line containing a 2-liter expansion volume. These aliquots were combined to make up the individual CO, sample and then purified through the use of liquid nitrogen and dry ice-isopropanol traps. Mass spectrometry was performed on a Finnegan MAT 251 and reported in per mil notation relative to PDB. The error on the analyses is estimated at ±0.2‰.

Chlorophyll and suspended particulate matter—SPM was measured by filtering 200–1,000-ml aliquots of water onto preweighed 47-mm Nuclepore filters (0.45-pm pore size). SPM concentration was determined gravimetrically following air-drying and correcting for the weight of salts retained in the filter matrix. Chl a (a measure of phytoplankton biomass) was determined from 200 to 1,000-ml aliquots of each water sample on 47-mm GF-AE filters and frozen immediately after collection. The filters were ground and extracted in 90% acetone. Absorbances of the acetone extract were measured with a Hewlett-Packard 8452A diode array spectrophotometer and used to calculate Chl a and pheopigment concentrations by the methods of Lorenzen (1967) and Riemann (1978). A conservative estimate of the phytoplankton component of seston carbon was calculated according to the following relationship (modified from Wienke and Cloern 1987):

$$\text{phytoplankton fraction} = \frac{35 \times \text{Chl a}}{\text{POC } (\mu\text{M}) \times 12} \times 100\%$$

where 35 is an estimated lower limit on the ratio of phytoplankton carbon to Chl a. Salinity was calculated from conductivity and temperature measurements made with a Sea-Bird Electronics SBE-9/11 instrument package that includes a 3-electrode conductivity sensor and thermistor.

Results

Seston: General parameters—Suspended particulate matter varied spatially during whole-bay transacts (6–53 mg $liter^{-1}$) representative of baseline conditions (January, July, and December 1990), although variations were more prominant during flood events in March and April 1991 (Fig. 2). Within the NB, SPM concentrations were highest (109 mg $liter^{-1}$) during floods. Temporal variability was greatest in the SB with the highest SPM concentrations (308 mg liter-') recorded at station 30 during the April 1991 (flood) cruise when winds were extremely strong and SPM concentrations were elevated by wind-wave resuspension. SPM concentration (<2 mg $liter^{-1}$) was lowest in SB in April 1990 (bloom; Fig. 2), when windspeeds and tidal currents were small. Chl a concentrations were also highest in the SB during the April 1990 bloom, with the maximum (62 μg liter-') measured at station 32 (Fig. 2). Phytoplankton biomass was consistently low in the NB except for a small localized bloom in San Pablo Bay during March 1991, when Chl a concentration reached a maximum of 8 μg liter-'.

POC concentrations ranged from 22 to 500 μM during

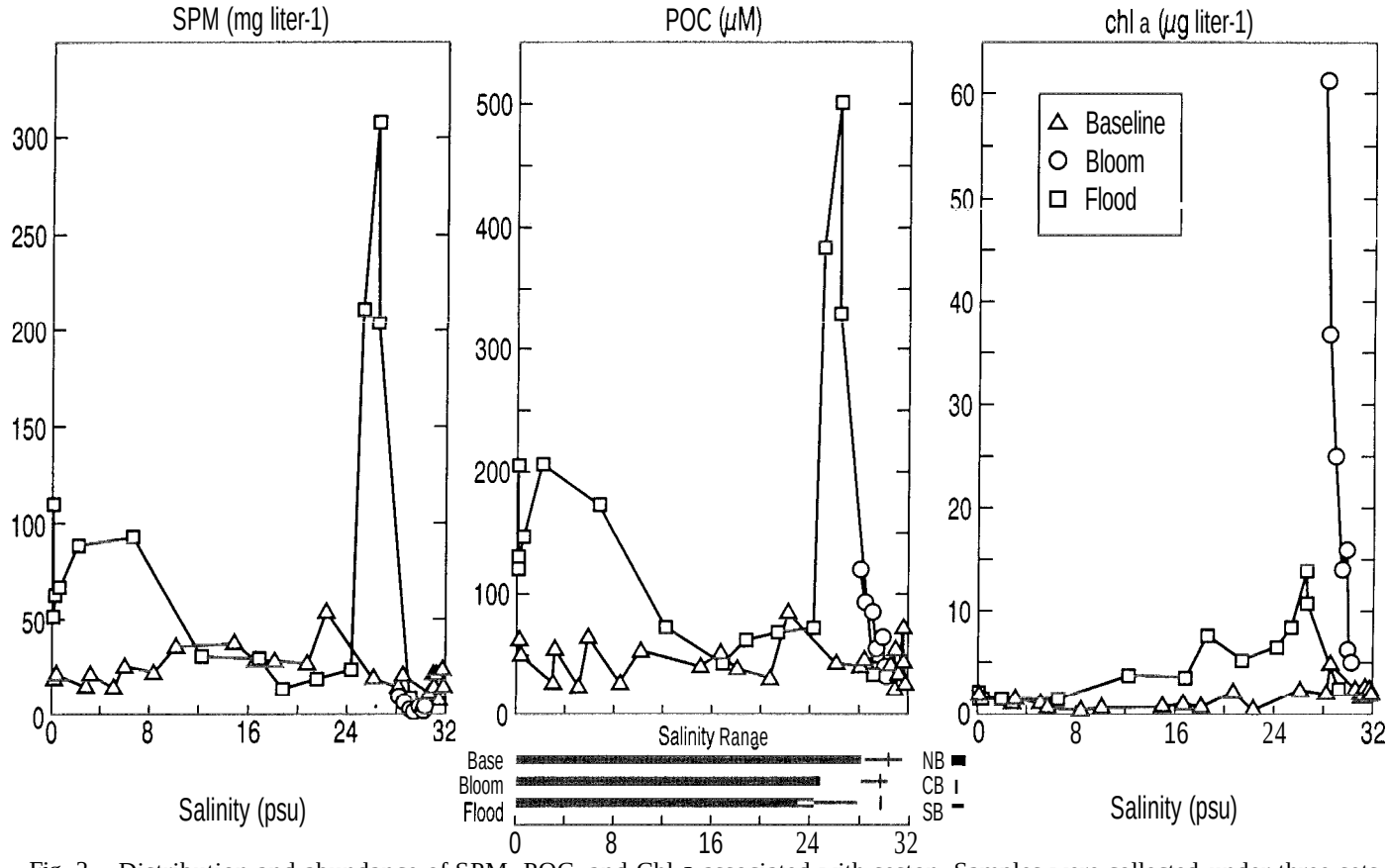

Fig. 2. Distribution and abundance of SPM, POC, and Chl a associated with seston. Samples were collected under three sets of conditions: the spring phytoplankton bloom in the SB (April 1990); winter storms, which include periods of freshwater inflow in the NB and wind-driven resuspension in the SB (March and April 1991); and baseline conditions (January, July, and December 1990). The data have been averaged in cases where there were multiple cruises. Salinity ranges for each sampling condition (baseline, bloom, and flood) are provided below the center panel (CB—the central bay).

the study (Fig. 2). Concentrations were highest in the SB during the April 1991 period of strong resuspension and elevated SPM concentration. Excluding this event, POC concentrations were highest in the NB, particularly during flood conditions. In general, POC tracked SPM except during the 1990 SB bloom, when SPM was extremely low. No significant correlation with salinity was found.

In the SB, C : N ratios of the seston increased from values resembling the Redfield ratio (6.6) during the April 1990 bloom to as high as 14 during periods of low phytoplankton biomass (Fig. 3). There was a strong correlation between salinity and C : N ratios in the NB ($r^2 = 0.73$, $P < 0.001$; Fig. 4), with the highest ratios (−16) at the riverine end and the lowest ratios (−7) at the seaward end. The stable carbon isotopic composition of the seston [δ^{13}]POC was generally in the range of −24 to −27‰ except during the April 1990 phytoplankton bloom in the SB, when values were significantly enriched in ^{13}C ([δ^{13}C]POC = −17 to −19‰, Fig. 3). During baseline conditions, [δ^{13}C]POC ranged from ~ −29% near the riverine end to ~ −23‰ at the seaward end of the NB. Like the C : N ratios, [δ^{13}C]POC was significantly correlated with salinity ($P < 0.001$) in the NB (Fig. 4). The [δ^{13}C]POC data appeared to follow a conservative mixing line (typically bowed for isotope conservation), as was seen in previous work (Spiker and Scheme1 1979). The isotopic [δ^{13}C]POC composition of the seston increased (became more positive) with increasing salinity, as did the [δ^{13}C]DIC (−11.0 to −0.2‰). The slope of the [δ^{13}C]POC vs. salinity relationship, however, differed from that of the [δ^{13}C]DIC vs. salinity relationship (Fig. 4).

The phytoplankton component of seston carbon varied spatially, with values ranging from 2 to 28% during baseline conditions (Fig. **3).** During the 1990 SB spring bloom, the C : Chl *a* relationship did not hold, as values exceeded 100% of the seston carbon at the southernmost sites (Sta. 30 and 32). Assuming 100% of the seston carbon was phytoplankton at these sites, the phytoplankton C : Chl *a* ratios would be 23 and 29 rather than 35, as used above.

Seston: Lipid *biomarker* compounds—The distributions of individual fatty-acid and sterol components (identified in Tables 2 and 3) associated with seston samples are presented in Table 4. In 1990, seston samples were collected during a phytoplankton bloom in the SB (April) and during a nonbloom period in the NB (July). The most abundant fatty acids in seston collected from the SB were 14 : 0, 16 : 0, and 16 : 1ω7c (Table 4). These three constituents made up 61% of the fatty-acid com-

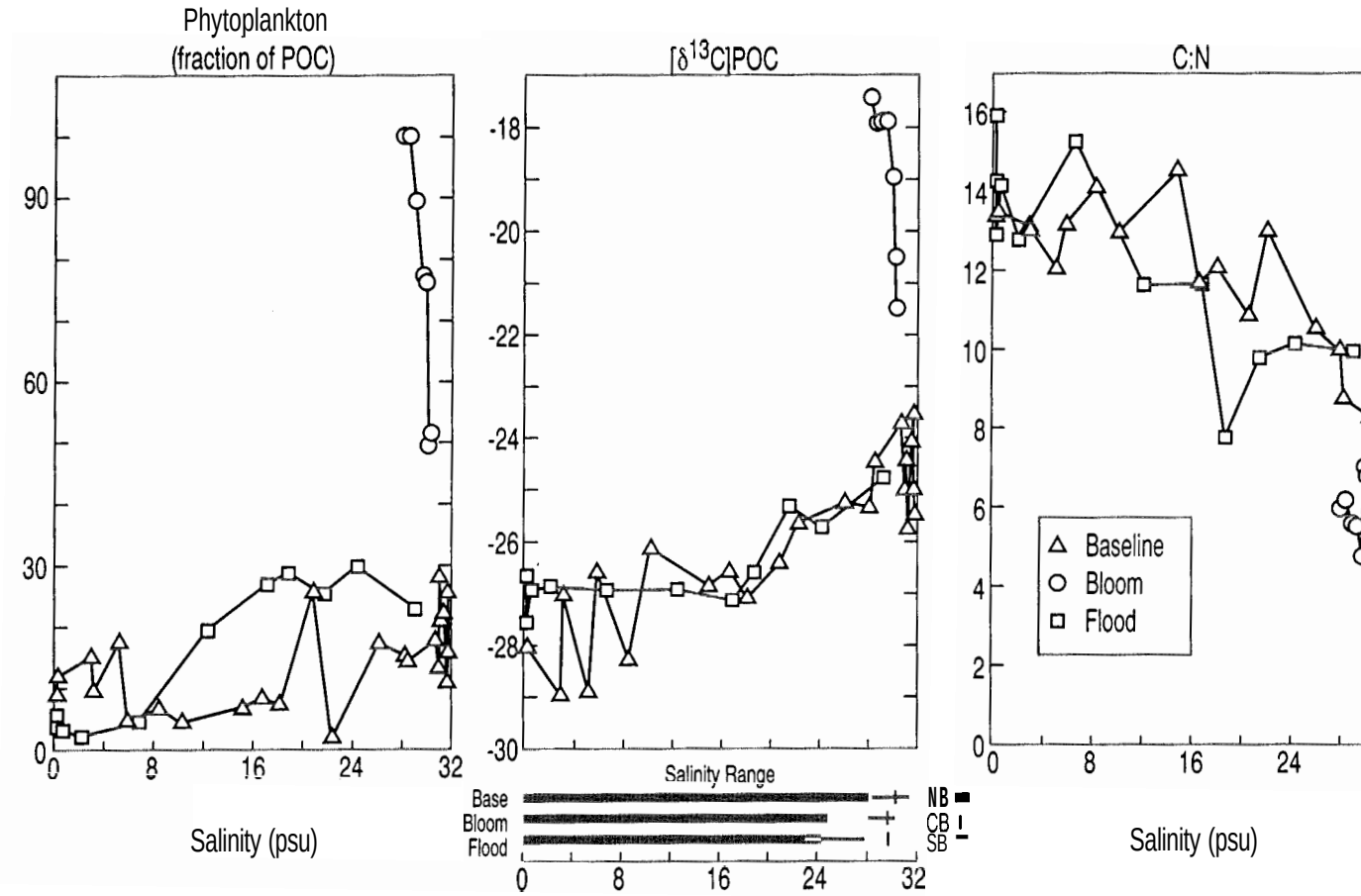

Fig. 3. Three parameters were used to evaluate how the sources of OC associated with the seston varied spatially and tempora The portion of the POC derived from phytoplankton was calculated according to Wienke and Cloem (1987). Stable carbon isoto [δ^{13}C]POC and C : N ratios were also used to evaluate variations in the importance of various sources. Salinity ranges for ea sampling condition are provided below the center panel as in Fig. 2.

Table 2. Sterol and fatty acid abbreviations.

	Sterols
S-1	24-norcholesta-5,22E-dien-3β-ol (24-norcholesterol)
S-2	cholesta-5,22E-dien-3β-ol (trans-22-dehydrocholesterol)
S-3	cholest-5-en-36-01 (cholesterol)
S-4	cholesta-5,24E-dien-3β-ol (desmosterol)
S-5	24-methylcholesta-5,22E-dien-3β-ol (brassicasterol)
S-6	24-methylcholest-5,24(28)-dien-3β-ol (24-methylenecholesterol)
S-7	24-methylcholest-5-en-3P-01 (campesterol)
S-8	24-ethylcholest-5-en-30-01 (sitosterol)
S-9	24-ethylcholesta-5,24(28)-dien-3β-ol (iso-fucosterol)
Σsterols refers to sum of all sterol components	
	Fatty acids
A : BωC	Designation where A is the number of carbon atoms, B is the number of double bonds, and C is the position of the double bond from the aliphatic (w) end of the molecule
PUFA	Polyunsaturated fatty acids (e.g. 18 : 4ω3, 20 : 5ω3, 22 : 6ω3)
PLFA	Phospholipid ester-linked fatty acids
ΣPLFAs refers to the sum of all PLFA components	
EFA	Essential fatty acids (e.g. ω3 and ω6 PUFAs)
iso-	Branched fatty acid with methyl group (CH_3) at ω-1 position
anteiso-	Branched fatty acid with methyl group (CH_3) at ω-2 position

ponents identified in the SB bloom samples. Polyunsaturated fatty acids (PUFAs) were remarkably low in abundance (14%) and contained roughly equal proportions of $C_{,,}$, $C_{,,}$ and $C_{,,}$ fatty acids. The most abundant fatty acid associated with seston collected from the NB was 16 : 0; however, this component represented only 11.7% of the total fatty acids. NB samples included low levels of several C_{20} and C_{22} PUFAs, probably derived from phytoplankton. Longer chained saturated fatty acids (C_{20}, C_{22}, and C_{24}) were more abundant at the freshwater endmember in the NB samples.

During 1991, seston samples were collected along the NB, including San Pablo Bay (SPB) and the seaward central bay (CB) during March, and on a weekly basis from the SB (Sta. SM49) between February and April. The most abundant fatty acids in most of these samples were 16 : 0, 16 : 1ω7c, and 18 : 1ω7c. Two polyunsaturated $C_{,,}$ fatty acids (18 : 4ω3 and 18 : 3ω3) were abundant in SPB, making up 35% of the distribution.

Sterol distributions were generally dominated by cholest-5-en-3P-ol (cholesterol; S-3) except in SPB, where 24-methylcholesta-5,22-dien-3β-ol (brassicasterol; S-5) was the most abundant sterol (Table 4). In the SB, the sterol composition of the seston varied between the bloom and nonbloom years (1990 vs. 1991). During the 1990 spring bloom, sterol distributions were characterized by high levels (21%) of 24-methylcholest-5,24(28)-dien-3β-ol (24-rnethylenecholesterol; S-6) and cholesta-5,22-dien-3β-ol (desmosterol; S-4); in 1991, 24-methylcholesta-5,22-dien-3β-ol (S-5) and 24-methylcholest-5, 24(28)-dien-3β-ol (S-6) were abundant. High concentrations of 24-methylcholesta-5,22-dien-3β-ol (S-5) and 24-ethylcholest-5-en-3P-ol (S-8) were characteristic of the NB and CB regions.

Composition of P. amurensis—In general, *C* : N ratios of the clam tissues (5.3–7.9) were similar to that of other animals. The isotopic composition ([δ^{13}C]POC) of the tissues ranged from −27.8 to −18.1% (Fig. 5). The [δ^{13}C]POC values increased consistently along the freshwater-seawater continuum and were significantly correlated with salinity ($P < 0.001$; Fig. 4); there was little variation among sampling dates. Carbon isotopes were generally heavier in clams collected from the SB and were heaviest during the April 1990 bloom (Fig. 5).

The distributions of fatty acid and sterol components in the tissues of P. *amurensis* are presented in Table 5. The most abundant fatty acid component was 16 : 0, while 20 : 5ω3 and 22 : 6ω3 reached moderate abundances in most collections. Polyunsaturated C_{20} and C_{22} fatty acids were most abundant in clams collected from the SB during the April 1990 bloom. In contrast to the seston samples, high concentrations of polyunsaturated C_{18} fatty acids were not found in clams. During 1990, clams collected during nonbloom periods (NB and SB) had significant (>10%) levels of one or more monounsaturated C_{20} fatty acids (20 : 1w11c and 20 : 1w13c which could not be separated by our methods).

Clam tissues contained a diverse assemblage of sterol components. Throughout the study, cholest-5-en-3β-ol

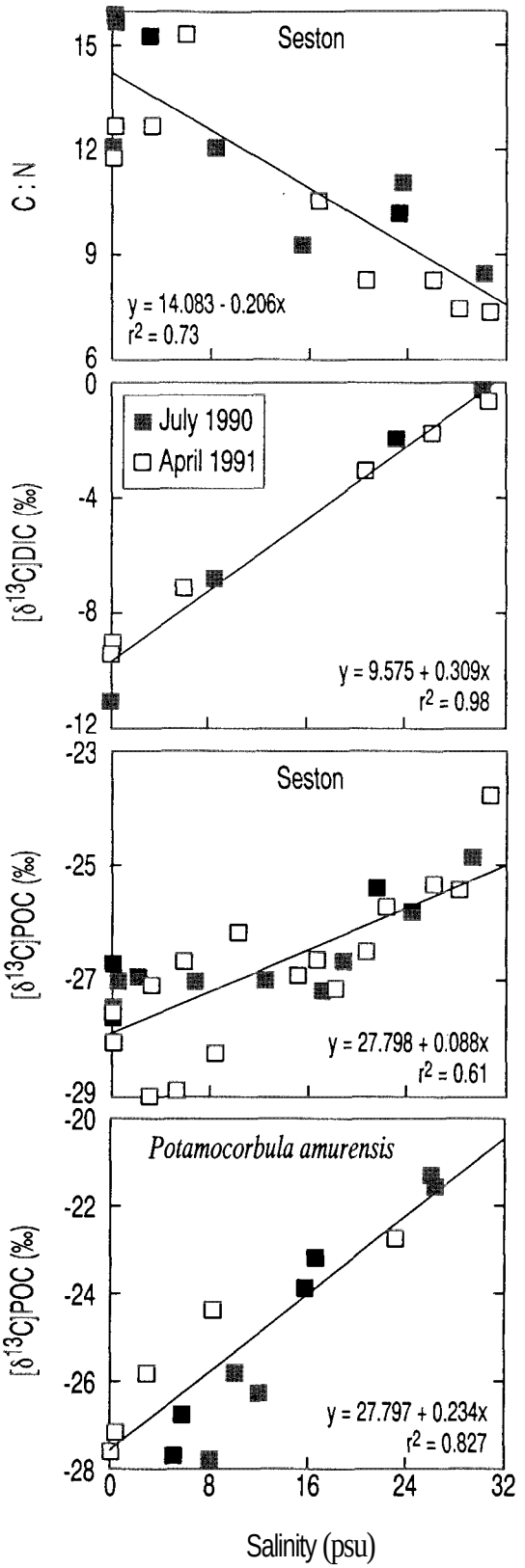

Fig. 4. Significant correlation ($P < 0.05$) with salinity was found for several parameters measured along the salinity gradient in the NB. Data are presented for baseline (July 1990) and flood (April 1991) conditions.

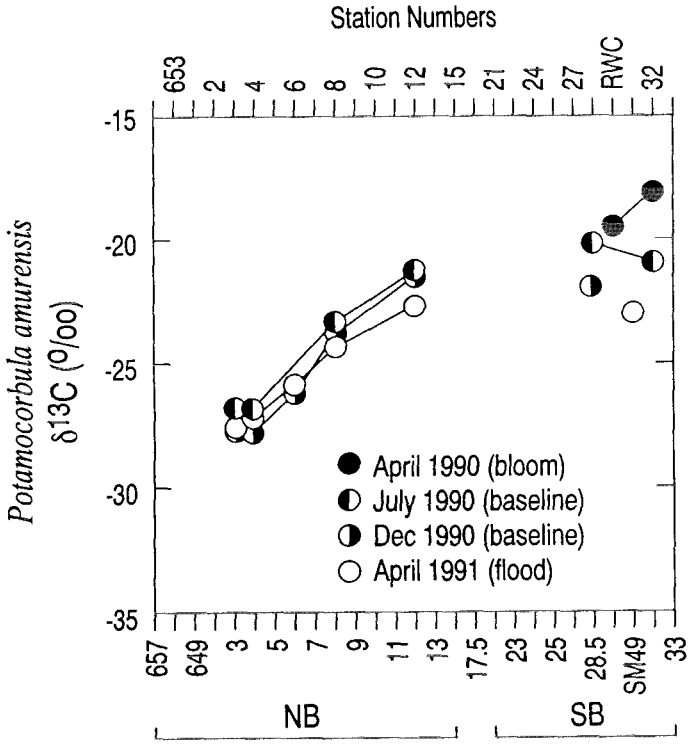

Fig. 5. Isotopic composition of *P. amurensis* collected on four dates. Values of [δ^{13}C]POC in the clam tissues increased consistently from NB to SB. The heaviest values were measured in the SB (Sta. 32) during the spring bloom.

(S-3; cholesterol) was the most abundant sterol except during the bloom in SB, when 24-methylcholest-5,24(28)-dien-3β-ol (S-6) was most abundant (Table 5). Phytoplankton-derived sterols were abundant, with the predominant components including 24-methylcholest-5,22-dien-3β-ol (S-5) and 24-methylcholesta-5, 24(28)-dien-3β-ol (S-6). Sterols generally indicative of terrestrial inputs (e.g. 24-ethylcholest-5-en-3β-ol; S-8) were low in abundance.

Discussion

Variation in sources of organic matter associated with seston—Much of the variation in the seston was associated with two events that influenced the supply of organic matter to the system—the spring phytoplankton bloom, which was strongest in the SB, and the flow of freshwater into the NB during winter-spring months in response to seasonal increases in rainfall and runoff. Lipid biomarker compounds (*see Table 3*) and stable carbon isotopes were used to evaluate how the organic matter supplied by these different processes varied in source and character.

Source-specific fatty-acid components associated with the seston indicated that organic matter derived from phytoplankton was important during the 1990 phytoplankton bloom in the SB and in SPB (Sta. 12 and 13) during the spring 1991 floods. This suggestion is supported by the increased abundance of PUFAs, common to many species of microalgae (Volkman et al. 1989). It is surprising, however, that seston samples collected during the large phytoplankton bloom in the SB (when seston was primarily phytoplankton, Fig. 3) contained relatively low levels of PUFAs (14%) compared to seston samples collected from the much smaller bloom which took place in SPB in spring 1991 (35%, Table 4; Fig. 5). PUFAs also differed compositionally during each of these blooms, with roughly equal proportions of C_{18}, C_{20}, and C_{22} fatty acids in seston collected during the SB bloom, while C_{18} PUFAs were more abundant in the SPB bloom samples. These data suggest that there may be differences in the communities of organisms making up each of these blooms or that there may be an alternative source for the C_{18} PUFAs. Polyunsaturated C_{18} fatty acids are synthesized by unicellular green algae (although generally in association with polyunsaturated C_{16} acids; Volkman et al. 1989 and references therein) but have also been found in dinoflagellates (Nichols et al. 1984) and diatoms of the genus *Thalassiosira* (Volkman and Hallegraeff 1988). Fungi have also been shown to contain C_{18} PUFAs (Shaw and Johns 1985).

Spring blooms in the SB are traditionally dominated by coastal diatoms such as *Thalassiosira rotula, Coscinodiscus* spp., *Skeletonema costatum, Cyclotella* spp., *Rhizosolenia* spp., and *Chaetoceros* spp. (e.g. Cloern et al. 1985). Phytoplankton counts from station 27 during the April 1990 bloom indicated that diatoms (*Coscinodiscus* spp. and *Thalassiosira* spp.) were the dominant organisms. Biomarker compounds associated with the seston samples were consistent with a diatom source (Table 3). These included 14:0, 16:0, 16:1ω7 fatty acids and the sterol, 24-methylcholesta-5,22-dien-3β-ol (Kates and Volcani 1966; Orcutt and Patterson 1975; Volkman et al. 1989). These compounds were abundant in seston samples collected from sites throughout the bay. In the SB, the concentration of 24-methylenecholesterol (S-6) was markedly higher during the 1990 bloom. This sterol

Table 3. Typical source assignments.

Bacterial:	iso- and anteiso-C_{15} and C_{17} saturated and monounsaturated fatty acids; *cis*-vaccenic acid (18 : 1ω7c)
Algal:	Fatty acids generally with chain lengths $<n$-C_{20}; polyunsaturated fatty acids (PUFAs); C_{27} and C_{28} sterols (e.g. diatoms—16 : 1ω7, 16 : 0, 14 : 0, 20 : 5ω3, S-5, S-6; dinoflagellates—16 : 0, 18 : 5ω3, 22 : 6ω3, 4-methyl sterols)
Animal:	16 : 0, 18 : 0, and 18 : 1ω9; cholest-5-en-3β-ol (S-3)
Terrestrial:	Even-numbered saturated fatty acids with chain lengths $>n$-C_{20}; C_{29} and other higher plant sterols (e.g. 24-ethylcholest-5-en-3β-ol; S-8)

Table 4. Lipid components in seston (% ΣPLFAs, Zsterols). Values are means (± standard deviations) calculated for biomarker composition of seston collected from sites from each region of SFB under sampling conditions listed. (Not determined—nd.)

Component	NB Jul 90, baseline	SB Apr 90, bloom	NB*	SPB	CB	SB
				Mar 91, flood		
% ΣPLFAs						
anteiso13.0	2.0(1.3)					
13 : 0	3.4(1.7)					
*iso*14 : 0	0.2(0.3)	1.1(1.1)	1.6(0.9)	0.3(0.03)	0.9	0.3(0.3)
14 : 0	2.1(1.7)	14.4(2.7)	3.1(1.1)	3.6(0.05)	7.5	4.4(2.3)
*iso*15 : 0	0.4(0.8)	0.8(0.7)	2.9(1.3)	0.5(0.2)	1.3	1.2(0.5)
*anteiso*15 : 0	0.3(0.7)		2.8(1.2)	0.6(0.2)	1.2	1.3(0.5)
15 : 0	0.3(0.6)	0.8(0.7)	0.9(0.3)	0.5(0.08)	1.1	1.1(0.4)
16 : 1ω7c	6.1(3.8)	13.0(2.3)	20.6(4.5)	8.0(3.0)	14.0	21.6(4.8)
16 : 0	11.7(7.6)	33.7(3.5)	18.6(2.0)	24.3(7.2)	31.8	27.5(3.2)
br17 : 1	1.5(1.1)					
*iso*17 : 0	2.5(2.7)		0.7(0.2)	0.2(0.1)	0.3	0.4(0.2)
17 : 0	1.7(1.5)	0.4(0.4)	0.8(0.1)	0.5(0.03)	0.8	0.9(0.2)
18 : 4ω3		4.9(4.6)	3.8(5.2)	17.4(6.0)	6.5	5.0(3.3)
18 : 3ω3		2.4(2.4)	3.3(2.9)	13.7(4.4)	3.9	2.2(2.4)
18 : 1ω9c	4.8(1.4)	3.5(1.5)	5.1(0.7)	3.0(0.7)	3.4	3.7(0.7)
18 : 1ω7c	10.5(5.9)	4.8(1.1)	13.6(1.0)	10.3(2.5)	9.7	12.0(2.0)
18 : 0	6.7(1.4)	6.1(3.9)	3.7(0.7)	2.0(0.5)	3.9	3.3(0.6)
20 : 5ω3		3.3(1.3)	2.0(1.8)	3.7(1.8)	2.0	5.2(2.7)
20 : 4ω6	4.9(3.1)					
20 : 4ω3						
20 : 0	6.1(9.2)					
22 : 6ω3		3.0(3.2)				
22 : 5ω6	3.1(3.3)					
22 : 5ω3	4.5(5.3)					
22 : 1ω9c	0.3(0.5)					
22 : 0	3.9(0.6)					
24 : 0	4.1(0.9)					
Σ pg ml^{-1}	161.5(110)	1,404(1,248)	4,443(1,383)	4,987(1,736)	770.6	3,756(1,730)
% Σsterols†						
S-1	nd	9.0(0.9)	3.2(2.1)	10.5(0.3)	4.0	7.4(3.7)
S-2	nd	10.2(1.6)	7.2(1.4)	4.3(4.7)	12.1	5.3(2.3)
S-3	nd	22.5(3.9)	31.7(5.2)	13.7(5.3)	25.8	36.6(19.3)
S-4	nd	10.5(0.7)	1.1(1.3)	2.5(1.1)	3.8	2.5(1.3)
S-5	nd	9.1(1.4)	13.9(9.4)	53.6(6.6)	19.1	13.9(4.0)
S-6	nd	20.9(2.8)	2.6(1.8)	4.6(0.9)	6.5	13.2(12.4)
S-7	nd	6.3(0.4)	9.7(4.0)	2.8(1.3)	5.7	8.7(4.3)
S-8	nd	4.6(1.6)	23.8(9.2)	4.4(1.6)	16.0	8.4(2.5)
S-9	nd	2.2(0.03)	6.6(1.5)	3.6(2.6)	7.0	4.1(2.1)
Σ pg ml^{-1}		4,044(1,976)	1,580(745)	1,269(195)	360.8	2,203(1,231)

* Does not include San Pablo Bay.
† Sterol identifications listed in Table 2.

has been shown to be a major component in marine diatoms of the genus Thalassiosira (Volkman and Hallegraeff 1988).

The isotopic composition of the seston is also consistent with a phytoplankton source with significantly heavier values ([δ^{13}C]POC = −17 to −19‰) found in the SB during the April 1990 bloom (Fig. 5). This enrichment in ^{13}C has been seen in other environments during spring diatom blooms (e.g. Georges Bank, USA; Fry and Wainwright 1991), and the phenomenon has been attributed to carbon limitation during high primary production and the accumulation of cellular bicarbonate (Fogel et al. 1992).

Bacterial sources of organic matter were inferred by the presence of iso- and anteiso-branched acids (Perry et al. 1979). These fatty acids are most commonly associated with gram-positive bacteria. Monounsaturated fatty acids (e.g. vaccenic acid, 18 : 1ω7), which are synthesized via the anaerobic pathway, are also commonly associated with a bacterial source (Gillan and Johns 1986). Monounsaturated acids, however, can be biosynthesized by a variety of organisms (see above), and thus they cannot be ascribed solely to a bacterial source as can the iso- and anteiso-branched acids. Although iso- and anteiso-branched acids were not very abundant (2–10%) through-

Table 5. As Table 4, but in Potamocorbula amurensis.

Component	NB Jul 90, baseline	SB Apr 90, bloom	SB Jul 90, baseline	NB Apr 91, flood	SB Apr 91, flood
% ΣPLFAs					
14 : 0	0.7(0.3)	1.3(0.3)	0.9(0.1)	2.6(0.8)	2.2(0.4)
15 : 0	1.0(0.3)	0.6(0.1)	0.9(0.1)	2.0(0.4)	1.3(0.3)
16 : 1ω7c	2.2(0.9)	4.7(1.0)	3.7(1.0)	4.2(0.9)	5.1(0.5)
16 : 0	14.0(1.7)	22.8(2.0)	19.7(1.2)	37.6(6.1)	39.7(3.0)
anteiso17 : 1ω8c	3.0(0.5)	0.3(0.2)	1.7(0.6)	4.0(1.9)	3.3(0.3)
*iso*17 : 0	1.0(0.2)	0.5(0.2)	1.3(0.4)	2.4(0.6)	2.1(0.2)
anteiso17 : 0				1.2(0.4)	0.6(0.1)
17 : 0	1.9(0.1)	1.0(0.2)	2.6(0.2)	3.5(0.4)	3.5(0.3)
18 : 4ω3	1.1(0.5)	0.6(0.3)	1.9(1.0)		
18 : 3ω3	0.3(0.1)	1.2(0.2)	0.5(0.2)		
18 : 1ω9c	3.9(0.5)	2.3(0.2)	4.4(0.6)	10.0(2.7)	4.3(0.4)
18 : 1ω7c	1.6(0.2)	3.1(0.2)	2.7(0.3)	2.5(0.5)	3.0(0.2)
18 : 0	3.6(0.7)	2.7(0.1)	6.8(1.1)	5.0(1.6)	7.1(0.8)
20 : 5ω3	8.5(1.5)	20.8(0.7)	1.0(2.7)	6.3(3.0)	7.2(1.5)
20 : 4ω6	7.5(1.5)	2.2(0.3)	2.9(0.3)	5.0(2.2)	1.9(0.3)
20 : 4ω3	1.6(0.2)	0.9(0.7)	2.5(0.3)	1.1(0.1)	1.5(0.2)
20 : 1ω11c/13c	13.1(2.6)	3.9(1.1)	18.1(1.3)	4.0(1.5)	3.2(1.7)
20 : 1ω9c		1.7(1.2)		2.3(0.8)	1.5(0.8)
20 : 1ω7c	2.6(0.1)	2.3(0.3)	3.6(0.3)	0.9(0.3)	1.2(0.2)
20 : 0					
22 : 6ω3	14.6(2.8)	14.6(1.5)	5.9(0.3)	4.7(2.4)	3.9(0.9)
22 : 5ω6	3.1(0.7)	0.9(0.3)	0.8(0.04)	1.0(0.7)	0.6(0.1)
22 : 5ω3	2.5(0.2)	3.9(0.6)	1.7(0.4)	0.8(0.4)	1.1(0.3)
22 : 4ω6	2.8(0.7)	1.1(0.4)	1.1(0.3)		
22 : 1ω9c				1.0(0.2)	1.0(0.2)
Σ ng mg^{-1}*	3,359(952)	5,786(674)	1,779(773)	1,063(512)	872(255)
Σsterols					
S-1	9.5(2.4)	9.6(0.03)	8.6(1.9)	9.7(2.1)	10.0(1.1)
S-2	10.2(1.6)	6.6(0.4)	7.5(0.2)	12.8(2.2)	7.7(0.3)
S-3	32.3(3.1)	20.3(0.1)	29.8(5.0)	26.9(1.5)	22.4(0.6)
S-4	3.7(1.0)	10.8(0.6)	2.8(1.4)	6.1(1.0)	4.9(0.9)
S-5	18.4(4.6)	8.3(0.6)	14.7(1.7)	19.8(0.9)	17.7(1.3)
S-6	8.1(2.4)	22.4(0.6)	15.1(3.8)	8.7(1.9)	17.8(1.3)
S-7	7.1(1.6)	6.3(0.4)	7.2(1.1)	5.5(0.8)	7.7(0.6)
S-8	4.0(0.9)	4.4(1.8)	5.2(0.5)	4.5(0.8)	4.5(0.4)
S-9	1.9(0.3)	2.2(0.03)	2.3(1.3)	2.3(0.1)	2.2(0.4)
Σ ng mg^{-1}*	3,162(1,169)	1,489(407)	3,139(956)	785(151)	627(216)

* Concentration data are expressed per gram wet tissue. The average water content was 29.2±6.0%.

out the bay, they did vary spatially in the NB, with a marked increase in these components at the river-freshwater end through Suisun Bay in contrast to the SPB or CB regions (Table 4; Bact FAs Fig. 6). Increased levels of bacterial organic matter at the freshwater end-member may be due to the import of soil microbes via river inflow or increased microbial production in this part of the estuary from the metabolism of allochthonous sources of organic matter.

Organic matter from terrestrial higher plants is indicated by long-chained fatty acids ($>n$-C_{24}) and sterols such as 24-methylcholest-5-en-3P-01 (campesterol) and 24-ethylcholest-5-en-3P-ol (sitosterol; reviewed by Volkman 1986). Long-chained fatty acids were not present in seston samples; however, campesterol (S-7) and sitosterol (S-8) represented >30% of the sterol distribution in the NB during 1991 flood sampling. Sitosterol in particular was very abundant from Suisun Bay (Sta. 4) through the low salinity end-member of the NB transect (Sta. 657; Fig. 6). Elevated C : N ratios and light [δ^{13}C]POC values (Figs. 3 and 4) are also consistent with an increased importance of terrestrial organic matter at the freshwater end-member (e.g. Wada et al. 1987; Matson and Brinson 1990). The abundance of sitosterol was high in the CB as well, suggesting an additional marine source for this component, as has been reported elsewhere (Volkman 1986).

Lipid biosynthesis in bivalves—Although specific information regarding the biosynthesis of lipids by P. *amu-*

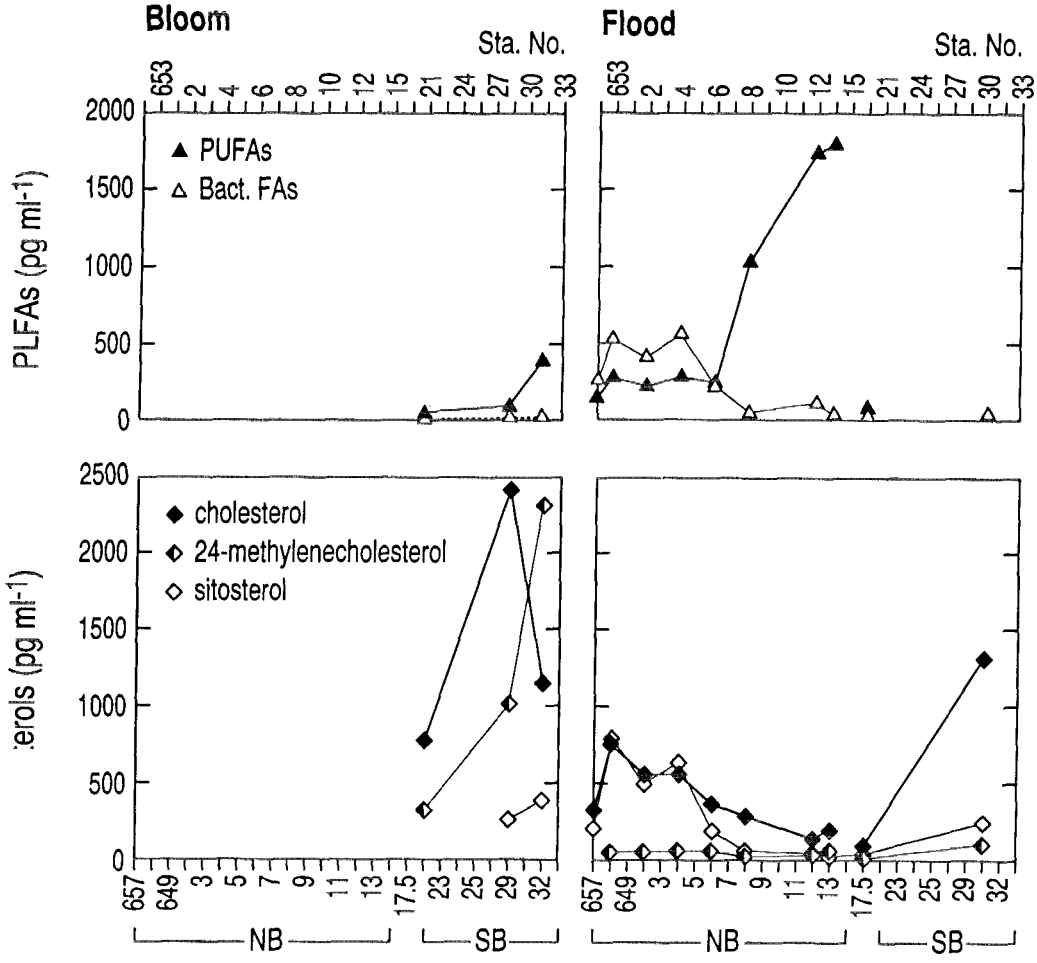

Fig. 6. Source-specific lipid compounds (PUFAs and bacterial fatty acids; sterols) associated with seston compared for two events thought to influence the origin of organic matter: the spring bloom and winter floods. PUFAs indicative of phytoplankton are compared with fatty acids specific to gram-positive bacteria (iso- and anteiso-C_{15}, C_{15}, and C_{17}). PUFAs increased during the spring bloom in the SB as well as during a small bloom in SPB (Sta. 12, 13) that coincided with our "flood" cruise. Bacterial fatty acids (possibly associated with soils) were enriched at the freshwater end-member (Sta. 657-4) during periods of flow. The abundance of three sterols—cholesterol (ubiquitous), 24-methylenecholesterol (phytoplankton), and sitosterol (vascular plants, occasionally phytoplankton)—are compared in the lower panels.

rensis is unavailable, the lipid content of marine bivalves is generally thought to be determined by diet (DeMoreno et al. 1980). As a result, the lipid composition of these organisms can be used to make inferences regarding diet where specific molecules known to be synthesized exclusively by other organisms (algae, higher plants, bacteria) can be detected. We must use caution in making such interpretations, however. The presence of specific lipids in bivalve tissues can indicate that the organism uses certain types of organic matter, but the absence of other source-specific molecules does not necessarily indicate that these are unimportant. Molecules may be transformed during digestion and thus lose the structural features which would allow them to be traced to a specific source.

PUFAs are considered essential fatty acids for animals, and they are generally derived either from the animal's diet directly or following conversion of dietary components. Although the end product of fatty-acid metabolism in bivalves is limited to 14 : 0 (Voogt 1983), bivalves can also achieve a full complement of fatty acids by using chain elongation and desaturation. Although monounsaturation, generally using **A9** desaturase, is most efficient with stearic acid (18 : 0), work by DeMoreno et al. (1977) has shown de novo biosynthesis of 16 : 1 (ω7), 18 : 1 (ω9), and 20 : 1 (ω11) fatty acids for the yellow clam *Mesodesma mactroides*. Animal fatty-acid synthetases cannot, however, insert a double bond into a fatty acid closer to the methyl end of the carbon chain than the ω7 position. Because double bonds can be introduced only at positions between the carboxyl group and the nearest double bond, polyunsaturated fatty acids synthesized by animals are unlikely to have double bonds at "ω" <7 (Conway and Capuzzo 1991). As a result, the ω3 and ω6 polyunsatu-

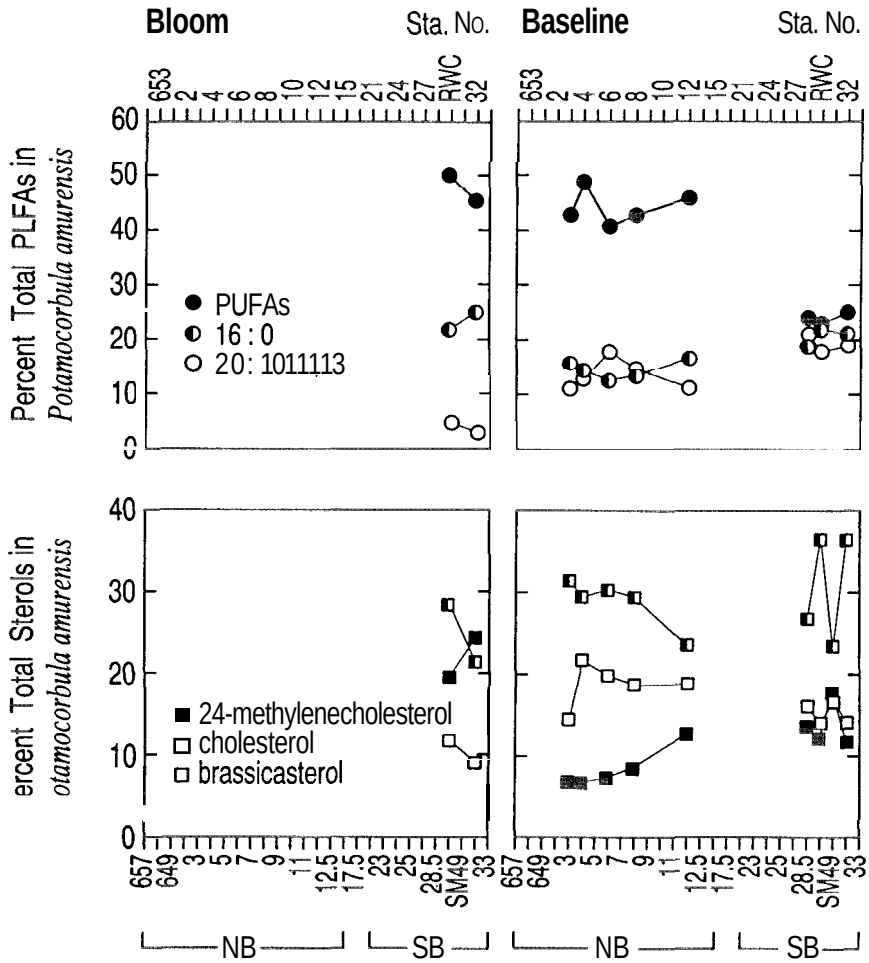

Fig. 7. The lipid composition of P. amurensis showed spatial and temporal changes in the relative abundance of fatty-acid and sterol components collected during bloom and baseline conditions. The greatest differences were measured in clams collected in the SB (Sta. **RWC-32)** during bloom vs. baseline periods. The composition of clams collected along a salinity gradient in the NB was relatively homogeneous and similar to clams collected from the SB during bloom conditions.

rated fatty acids found in P. amurensis tissues were most likely obtained from algae (probably diatoms) in the diet. This is consistent with results obtained for M. *mactroides* (DeMoreno et al. 1976) and the mollusc *Chlamys* te-*huelcha* (Pollero et al. 1979).

Lack of agreement in experiments carried on by other investigators has not resolved whether bivalves are capable of sterol biosynthesis, and this topic remains controversial. Although bivalves are thought to be incapable of sterol synthesis, limited data indicate the incorporation of radiolabeled acetate into the sterols of Mytilus *californianus*, Saxidomus giganteus (Fagerland and Idler 1960), and Crassostrea virginica (Teshima and Patterson 1981). It seems, however, that most bivalves are capable of only limited de novo synthesis (Voogt 1983). Sterol distributions in P. amurensis from SFB are fairly diverse, containing up to 13 components. It is possible that some of these sterols [e.g. cholesterol (S-3)] are synthesized by the clams, but most other components are probably derived from the diet of the organism either directly or after some modification. Sterols such as brassicasterol (S-5), 24-methylenecholesterol (S-6), and trans-22-dehydrocholesterol (S-2) are probably derived from an algal source. Components thought to be derived from higher plants (e.g. 24-ethylcholest-5-en-3β-ol and 24-methylcholest-5-en-3/3-01) were also present in P. amurensis tissues, albeit at low levels.

P. amurensis as an indicator of assimilable *carbon*—There was a substantial change in the biochemical composition of P. amurensis tissues collected from the SB during April (bloom) vs. July (baseline) samplings (Table 5, Fig. 7). During the phytoplankton bloom, PUFAs (predominantly C_{20} and C_{22}) were the most abundant components, comprising 46% of the fatty acid distribution (Table 5). In contrast, the composition of clams collected from the SB during baseline conditions (July 1990) had significantly lower levels of the PUFAs (18%) and exhibited a relative enrichment (from 5 to 18%) in the abundance of one or more C_{20} monounsaturated fatty acids (coeluting 20 : Iw11c and 13c). Monounsaturated C_{20} fatty acids have been found in other bivalves (e.g. M. *mactroides*, DeMoreno et al. 1976) although not at the levels

reported here. Palmitic acid (16:0) made up about 20% of the fatty acid distribution in clams collected during bloom, flood, and baseline conditions (Fig. 7, Table 5).

Clams collected from the NB during baseline conditions (July 1990) also had high percentages of the C_{20} and C_{22} fatty acids (42±4%, Table 5; Fig. 7). These levels are comparable to those seen in clams from the SB during the phytoplankton bloom rather than during the nonbloom collection. There was no evidence for significant changes in the PUFA or, for that matter, PLFA composition of P. amurensis along the salinity gradient of the NB (Sta. 3–12.5, Fig. 7). Seston collected from the NB did not contain significantly greater percentages of these components ($\Sigma C_{20} + C_{22}$ PUFAs = 12.5±7.0%) than did the seston collected during the SB phytoplankton bloom (5.1±3.8%), indicating that high concentrations of the PUFAs in NB clams cannot be due to differences in the composition of the seston. It is unclear whether the NB clams have a storage mechanism for maintaining high levels of these labile components during periods of low phytoplankton abundance or whether they selectively consume and assimilate the available phytoplankton.

Although the C_{20} and C_{22} polyunsaturated fatty acids are probably derived from a phytoplankton source, they are not major components in phytoplankton, generally making up <20% of the total fatty acids (Volkman et al. 1989). For example, seston collected from the SB during the spring bloom (primarily phytoplankton, Fig. 3) exhibited relatively low levels of these components (6%; Table 4). The high relative abundance of these labile fatty acids in the clam tissues is therefore striking and suggests that these organisms can exert a dramatic effect on the qualify of particulate matter in the bay through their selective removal (i.e. digestion) of phytoplankton from the total seston pool (e.g. Cloern 1982; Alpine and Cloem 1992). Our findings indicate the opportunistic nature of the Asian clam, P. amurensis and suggest that these organisms are capable of rapid assimilation and incorporation of algal-derived organic matter into their tissues during periods of high phytoplankton abundance. Alternatively, because these compounds are essential to most bivalves, the clams may have developed a mechanism for sequestering these components for use during times of low phytoplankton abundance (e.g. NB clams). The NB, however, is not characterized by the large spring bloom events of the SB, suggesting that these clams must efficiently and selectively filter whatever phytoplankton or algal-derived detritus is available from the water column. If this is the case, then clams resident in the NB may use a different strategy for obtaining these essential components of their diet than do clams in the SB. Seasonal changes in the relative abundance of fatty acids in clams collected from SB may be due to changes in food supply as well as growth and reproduction. For example, P. arnurensis collected from the SB generally undergoes a period of rapid growth following the spring bloom, while clams in the NB are generally smaller (ash-free dry wt) and exhibit less growth during spring and early summer months (J. Thompson unpubl. data). Growth following the SB bloom may explain differences in the composition of clams collected during April vs. July in the SB. Clams collected from the SB are reproductive throughout the year, indicating that this process is not responsible for these temporal variations in fatty-acid composition (Parchaso 1993).

In addition to differences in their fatty-acid composition, P. amurensis tissues exhibited spatial and temporal variations in several sterol components, e.g. cholesterol, 24-methylenecholesterol, and brassicasterol (Table 5; Fig. 7). During nonbloom conditions (NB and SB), cholesterol was the most abundant sterol, followed by brassicasterol and 24-methylenecholesterol, which are both produced by diatoms. However during the 1990 phytoplankton bloom, 24-methylenecholesterol was the most abundant sterol in clam tissues collected at Sta. 32 (Fig. 7, Table 5).

The isotopic composition [δ^{13}C]POC of the clam tissues varied spatially, with lighter values (i.e. an enrichment in C_{12}) at the freshwater end (Figs. 4 and 5). Because animal [δ^{13}C]POC is largely determined by the [δ^{13}C]POC of the diet (e.g. DeNiro and Epstein 1978), the significant spatial (and to a lesser extent seasonal) [δ^{13}C]POC variation observed in P. *amurensis* (range = −27.8 to −17.7‰) requires an equivalent isotopic variation in its food source. This variation must arise because the isotopic composition of the dominant food source (e.g. algae) changes, the dietary contributions of different, isotopically contrasting food sources changes (e.g. marine vs. terrestrial sources), or some combination of the two.

Algal isotopic variations have been linked to changes in CO_2 supply relative to photosynthetic demand (e.g. Degens et al. 1968; Deuser et al. 1968; Fogel et al. 1992). Such an effect is consistent with our observations of a significantly elevated seston and clam [δ^{13}C]POC during an algal bloom and the implied low CO_2 supply-demand within the SB (Figs. 3 and 5). Low phytoplankton abundance in the NB indicates that carbon limitation is an unlikely explanation for the variations observed in this region of the bay. The biomarker data suggest that algae are the dominant food source for P. umurensis. If this is true, then a 10‰ range in algal [δ^{13}C]POC is indicated by the isotopic variations exhibited by the clam. The variation in [δ^{13}C]DIC along the NB salinity gradient, seen both in this study (Fig. 4) and in earlier work by Spiker (1980), provides an explanation for these variations in algal [δ^{13}C]POC. Assuming that phytoplankton fractionation of inorganic C is constant along the salinity gradient in the NB, the spatial variation in algal [δ^{13}C]POC is consistent with that seen for the isotopic composition of DIC in the NB (−10%). This would also account for the significant upstream decrease in clam [δ^{13}]POC observed at the freshwater end-member (Fig. 4). Alternatively, the upstream decrease in clam [δ^{13}C]POC could reflect a greater nutritional reliance on ^{13}C-depleted terrestrial OC. This, however, is not supported by the clam biochemical data because lipid compounds, which would be derived from terrestrial plants (e.g. 24-ethylcholest-5-en-3P-01, long-chained n-fatty acids), are absent or not abundant in P. amurensis tissues (Table 5).

Lastly, it is important to note that the clam [δ^{13}C]POC

gradient in the NB is significantly steeper than that of the seston, with isotopic deviations between clams and seston greatest in the downstream stations (Fig. 4). These data indicate selective, nonrandom particle feeding or assimilation by the clam, with greater preference for food enriched in ^{13}C (relative to the bulk seston) at the downstream NB stations. Alternatively, an intrusion of oceanic phytoplankton could supply ^{13}C-enriched particulate matter to the downstream stations. Similarity between the slope of the [$\delta^{13}C$]POC vs. salinity relationship for P. *amurensis* and that for the [$\delta^{13}C$]DIC vs. salinity relationship, however, suggests that the clams selectively assimilate the local phytoplankton component of the seston.

Conclusions

Molecular and isotopic tracers indicate that the sources, and hence the reactivity of POM, vary spatially and temporally in San Francisco Bay. These variations seem to be associated with seasonal and annual differences in the intensity of the spring phytoplankton bloom and inputs of allochthonous POM during runoff events. Spatial variation in the quality of the organic matter associated with seston reflected these events in the estuary. Lipid biomarker compounds indicate that particulate matter is predominantly derived from phytoplankton, with low contributions from bacterial and higher plant-derived sources of organic matter. The relative importance of these organic matter types appears to vary spatially, with compounds derived from bacterial and higher plant sources elevated at the low salinity region of the NB, particularly during flood conditions.

Tissues of P. *amurensis* collected from throughout the bay indicate that this filter feeder utilizes organic matter principally derived from phytoplankton. In general, the clam tissues reflect their dietary requirements and contain significant levels of essential fatty acids (C_{20} and C_{22} polyunsaturated fatty acids, for example) that the clams are unlikely capable of synthesizing de novo. These components originate from an algal component of the clams' diet. Concentrations of components indicative of phytoplankton ($C_{\text{„}}$ and C_{22} PUFAs, 24-methylenecholesterol) were higher during bloom than nonbloom periods in the SB. Temporal changes in the lipid composition of clams living in the SB may be due to growth, which generally occurs in late spring and may utilize compounds stored during the spring bloom. PUFAs were abundant in clams from the NB during nonbloom conditions, suggesting that the clams are able to assimilate and store these components or that clams living in the NB are highly efficient at selectively ingesting and assimilating algae under conditions of low phytoplankton biomass and high total seston loads.

Values of [$\delta^{13}C$]POC in the clam tissues increased consistently from NB to SB. Our data indicate that while algae supply the bulk of the essential compounds for these clams, the isotopic signature of the algae in this estuary varies significantly. This variation results from C limitation during periods of high productivity in the SB and from isotopically varying DIC sources along the NB salinity gradient. Our data illustrate the value of using multiple tracers (isotopes and lipid biomarker compounds, as done here) and suggest that consumer organisms with widespread distributions might be exploited as biological indicators of the spatial and temporal fluctuations in labile POM within ecosystems that have multiple inputs of organic matter.

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