Edaphic algae are an important component of salt marsh food-webs: evidence from multiple stable isotope analyses

Michael J. Sullivan, Cynthia A. Moncreiff

Department of Biological Sciences, PO Drawer GY, Mississippi State University, Mississippi State, Mississippi 39762, USA

ABSTRACT: Carbon, sulfur, and nitrogen stable isotope ratios were measured in an irregularly flooded Mississippi, USA, salt marsh to evaluate the relative importance of vascular plants and algae in the food web. Primary producers had distinct $\delta^{13}C$ values: Spartina alterniflora (hereafter Spartina) ($-13‰$), Juncus roemerianus (hereafter Juncus) ($-26‰$), and edaphic algae ($-21‰$). A pure zooplankton sample, which should closely approximate the phytoplankton, had a $\delta^{13}C$ value of $-23‰$. Values for $\delta^{34}S$ ranged from 0 to $+2‰$ for vascular plants, while those for edaphic algae and zooplankton were $+14$ and $+11‰$, respectively; $\delta^{15}N$ for all primary producers ranged from $+5$ to $+6‰$. For 49 of the 56 (88 %) consumers sampled, $\delta^{13}C$ fell within a range of -22 to $-18‰$; this range centered around edaphic algae and zooplankton, but was distinct from the $\delta^{13}C$ of Spartina and Juncus. Values of $\delta^{34}S$ for 48 of the 56 (86 %) estuarine consumers ranged from $+9$ to $+16‰$, which included edaphic algae and zooplankton but was 8 to 15 ‰ more enriched in ^{34}S than Spartina or Juncus. Dual isotope plots showed the fauna to tightly cluster around values for edaphic algae and zooplankton; no consumer even moderately resembled Spartina or Juncus. This strongly suggests that the ultimate food sources for this marsh's invertebrate and fish fauna are the benthic and planktonic algae; direct contributions from vascular plants appear to be minor.

INTRODUCTION

The great productivity of coastal salt marshes supports a diverse array of consumers which exhibit a variety of feeding modes. The major contributors to this productivity are the highly visible vascular plants and microscopic edaphic and planktonic algae. The edaphic algae includes diatoms, green, yellow-green, and blue-green algae inhabiting exposed and subtidal sediments of tidal creeks, as well as those associated with the sediments beneath the extensive and dense canopy of vascular plants on the marsh proper. Despite their small size, the edaphic algae exhibit significant primary production rates (Sullivan & Moncreiff 1988 and references cited therein). The ratio of annual edaphic algal to vascular plant net aerial production (EAP/VPP) typically ranges from 8 to 33 % in coastal salt marshes of the Atlantic and Gulf Coasts of the USA. However, an EAP/VPP value of 61 % was reported by Sullivan & Moncreiff (1988) in the Scirpus olneyi Gray zone of a Mississippi marsh and Zedler (1980) found values of 76 to 140 % in 4 vascular plant zones of a

California marsh. Furthermore, benthic microalgae may greatly augment the primary production of the water column in well-mixed shallow estuaries via their resuspension (Shaffer & Sullivan 1988).

Until relatively recently, it was universally assumed that salt marsh vascular plants supported food webs both within the marsh itself and in adjacent estuarine waters via a detritus-based pathway. However, stable carbon isotope work carried out in Georgia salt marshes (Haines 1976a, 1977) showed that the isotopic signature of detritus in tidal creeks did not match that of the dominant vascular plant Spartina alterniflora Loisel (hereafter Spartina). This unexpected finding began an extensive re-examination of the long-standing dogma that Spartina was a major component in the food web after it died and then decomposed. Subsequent work (Haines 1976b, Haines & Montague 1979, Hackney & Haines 1980, Sherr 1982, Hughes & Sherr 1983, Mariotti et al. 1983, Schwinghamer et al. 1983, Jackson et al. 1986) in salt marshes has amplified and extended this earlier work with different investigators claiming varying importance of Spartina and algal car-

bon as food sources. In all of these studies, a single isotope approach was utilized, and the benthic algae were either ignored, emphasized as a possible food source for selected consumers, or were lumped together with phytoplankton in discussions of the importance of algal carbon.

Peterson and co-workers (1985, 1986) were first to apply multiple stable isotopes (carbon, sulfur, and nitrogen) to the study of salt marsh food-webs. Dual isotope plots led to the conclusion that the fauna of a Massachusetts marsh were more dependent on *Spartina* than on phytoplankton, but they did not consider the benthic algae as a potential food source. Later multiple isotope work in a Georgia salt marsh (Peterson & Howarth 1987) led to the conclusion that assimilation of *Spartina* and algal carbon by consumers was approximately equal, and the possibility that edaphic algae might be important was stressed.

The present study has taken the multiple stable isotope approach to a Gulf Coast, USA, salt marsh. This was done to extend regional comparisons beyond the Atlantic coast of the United States and because of our belief that the importance of benthic algae in salt marsh food webs has been underestimated.

MATERIALS AND METHODS

Study area. All field work was conducted in Graveline Bay Marsh, Mississippi, USA, (30° 21′ 26″ N, 88° 40′ 59″ W), located ca 11 km southeast of the Gulf Coast Research Laboratory at Ocean Springs. The Graveline system is irregularly flooded with a mean tidal range of 0.6 m; inputs of freshwater are minimal. The dominant vascular plants are *Spartina* and *Juncus roemerianus* Scheele (hereafter *Juncus*). The production rates of the edaphic algae on the marsh proper (Sullivan & Moncreiff 1988) and the phytoplankton and subtidal microalgae of tidal creeks (Zimba unpubl. data) have been determined.

Sampling strategy. Floral and faunal samples for stable isotope analysis were collected during March, May, June, July, September, and October of 1987 and January, April, June, August, and September of 1988. The sampling effort was concentrated at three stations. Stn 1 was located near the upper end of Graveline Bayou; Stn 2 was approximately at the mid-point of the bayou; and Stn 3 was immediately upstream of the mouth. The 3 stations were floristically similar in terms of vascular plant composition.

Sample collection. Vascular plants (*Spartina* and *Juncus*) were carefully procured in the field to prevent contamination by the introduction of foreign material to the samples. Only living, aboveground biomass was collected. Benthic algae were collected whenever pos-

sible; samples were removed from the marsh surface with care to exclude as much non-algal material as conditions would permit. All plant samples were frozen if immediate processing was not possible.

Samples for zooplankton were collected on most sampling dates, with a concentrated effort on 13 January 1988. Plankton nets with mesh sizes of 335, 153, and 28 μm were towed for a maximum of 10 min. Collections were placed on ice and later frozen until processed.

Consumers (polychaetes, bryozoans, bivalves, gastropods, crustaceans, and a variety of fish species) were collected from as many habitats and trophic levels as possible. Animals were collected by hand or with dredges, seines, trawls, and gill nets as appropriate. Only live, intact organisms were taken to minimize contamination by shell or other foreign material. All samples were placed in clean plastic bags or containers, labelled as to date and location of collection, and placed on ice for transport to the laboratory, where they were frozen for future processing if immediate preparation was not possible.

Sample processing. All biological materials were washed with 10 % HCl to remove carbonate contaminants followed by repeated rinses with tap water to a neutral pH and a final rinse with distilled water. Excess moisture was removed from samples by gentle vacuum filtration on glass-fiber filter paper. Samples were then oven-dried to constant weight at 60 °C. Zooplankton were carefully separated from particulate material using a saturated NaCl solution; plankton floated on the surface with the majority of the detritus sinking to the bottom of the processing container. Using a dissecting microscope, all remaining visible detrital material was manually separated from the zooplankton to obtain as pure a sample as possible. Consumer samples were handled to minimize contamination with foreign material. Whenever possible, only muscle tissue was used to obtain consistent and comparable samples for each species. Final processing was essentially identical for all sample types. Dried samples were powdered using either a Wiley Mill equipped with a #40 delivery tube or ground with a mortar and pestle to as fine a consistency as possible.

The actual measurements of stable isotope ratios for all samples were performed by Coastal Science Laboratories of Austin, Texas. The accuracy of the $\delta^{13}C$, $\delta^{34}S$, and $\delta^{15}N$ analyses was 0.2, 0.5, and 0.2 parts per mil (‰), respectively. A blind control was included with each set of samples to the commercial firm to test the repeatability of the determinations.

Stable isotope terminology and background. Stable isotope ratios are reported with reference to a standard (Fry & Sherr 1984). The difference between the material under consideration and the standard is

expressed in parts per thousand or per mil (‰) according to the following formula:

$$\delta X = [(R_{sample}/R_{standard}) - 1] \times 10^3,$$

where X is ^{13}C, ^{34}S, or ^{15}N, and R is $^{13}C/^{12}C$, $^{34}S/^{32}S$, or $^{15}N/^{14}N$. The standards for C, S, and N are Peedee Belemnite, Canyon Diablo troilite, and atmospheric diatomic nitrogen, respectively.

Biological materials are usually depleted in ^{13}C relative to the PDB standard and hence have negative $\delta^{13}C$ values (Fry & Sherr 1984). Unlike $\delta^{13}C$ values, those of $\delta^{34}S$ may be positive or negative in the tissues of estuarine plants and animals. Estuarine plants will have positive values if they take up their inorganic sulfur primarily as ionic sulfate and negative if their main sulfur source is inorganic sulfide (Fry et al. 1982). In contrast to carbon and sulfur, few $\delta^{15}N$ values exist for salt marsh flora and fauna. All reported values for marsh plants have been positive.

RESULTS

Stable isotope ratios of producers

The stable carbon isotope ratios of the vascular plants and edaphic algae were distinct (Table 1). The C_4 plant *Spartina* had $\delta^{13}C$ values that exhibited very little seasonal variation (−13.9 to −12.8‰) with an average value of −13.2‰. For the C_3 plant *Juncus*, $\delta^{13}C$ ranged from −26.6 to −24.8‰ with an average value of −25.5‰. The composite sample of edaphic algae had a $\delta^{13}C$ value of −20.6‰. Samples of filamentous algae collected in the bayou yielded the following $\delta^{13}C$ values: *Bostrychia radicans* Montagne (−26.2‰), *Vaucheria* sp. (−19.3‰), *Ectocarpus* sp. (−18.7‰), and *Enteromorpha* sp. (−18.4‰). Excepting the very low value for the red alga *B. radicans*, $\delta^{13}C$ ranged from −20.6 to −18.4‰ for the benthic algae of Graveline Bay Marsh. It was not possible to collect a pure phytoplankton sample but a pure zooplankton sample was obtained; its $\delta^{13}C$ value was −23.3‰.

Stable sulfur isotope ratios were more variable than those of carbon (Table 1). The average $\delta^{34}S$ value for

Spartina was +1.4‰ but such values ranged from −8.5 to +13.9‰ over an annual cycle. However, with the exception of 2 high values, $\delta^{34}S$ was always less than +8‰. The range in $\delta^{34}S$ for *Juncus* (−4.8 to +5.4‰) was less than that for *Spartina*, with an average value of +0.4‰ being recorded for the former. The edaphic algae composite had a $\delta^{34}S$ value of +14.3‰ whereas the pure zooplankton sample was more depleted in ^{34}S with a value of +10.7‰. A single collection of the brown alga *Ectocarpus* sp. yielded a value of +13.1‰.

Stable nitrogen isotope values ranged from +3.4 to +6.7‰ and +3.7 to +7.3‰ for *Spartina* and *Juncus*, respectively. Mean $\delta^{15}N$ values for these two vascular plants were +5.2 and +5.3‰, respectively (Table 1). The edaphic algae composite had a $\delta^{15}N$ value of +6.1‰ whereas the pure zooplankton sample was +7.1‰. Samples of filamentous algae collected in the bayou yielded the following values: *Vaucheria* sp. (+4.7‰), *Bostrychia radicans* (+6.5‰), *Enteromorpha* sp. (+9.0‰), and *Ectocarpus* sp. (+10.6‰).

Stable isotope ratios of consumers

Table 2 and Fig. 1 summarize the $\delta^{13}C$ data for consumers sampled in the Graveline Bay Marsh system. Forty-nine of the 56 (88%) $\delta^{13}C$ values listed in Table 2 fell within a range of −22 to −18‰. Of these 49 consumers, 34 fell within an even narrower range of −22 to −20‰. The largemouth bases *Micropterus salmoides*, which was collected from the head of the Graveline system, had the lowest $\delta^{13}C$ value of −25‰, whereas deposit-feeding *Uca* spp. (fiddler crabs) had the highest (−15‰) of any consumer. The pure zooplankton sample possessed one of the lowest $\delta^{13}C$ values (−23‰) of any consumer. Filter-feeding bivalves (*Polymesoda caroliniana*, *Rangia cuneata*, *Ischadium recurvum*, *Geukensia demissa*, and *Crassostrea virginica*) exhibited a very narrow range of −22 to −21‰; this range is slightly more enriched in ^{13}C than zooplankton in the Graveline system.

The range in $\delta^{34}S$ values (+6 to +16‰) for marsh consumers was the greatest of the 3 isotope ratios (Table 2, Fig. 2). Values of $\delta^{34}S$ for 48 of 56 (86%) consumers

Table 1. Summary of $\delta^{13}C$, $\delta^{34}S$, and $\delta^{15}N$ values for primary producers and zooplankton of Graveline Bay Marsh in parts per mil (‰). Each mean is followed by its standard error and the number of samples (N)

Producers	$\delta^{13}C$	$\delta^{34}S$	$\delta^{15}N$
Spartina	−13.2 ± 0.1 (12)	+1.4 ± 2.2 (12)	+5.2 ± 0.5 (6)
Juncus	−25.5 ± 0.2 (12)	+0.4 ± 0.8 (12)	+5.3 ± 0.8 (4)
Edaphic algae	−20.6 (1)	+14.3 (1)	+6.1 (1)
Zooplankton	−23.3 (1)	+10.7 (1)	+7.1 (1)

Table 2. Summary of stable isotope ratios for consumers of Graveline Bay Marsh. Numbers for C, S and N indicate number of samples. Values are in parts per mil (‰); –: no analyses performed

Consumer	$\delta^{13}C$	$\delta^{34}S$	$\delta^{15}N$	C, S	N
Micropterus salmoides (largemouth bass)	−25.1	3.6	6.7	1	1
Micropogon undulatus roe	−23.4	7.4	8.1	1	1
Trinectes maculatus (hogchoker)	−23.3	8.1	–	2	–
Zooplankton, all species	−23.3	10.7	7.1	1	1
Micropogon undulatus (Atlantic croaker)	−22.4	10.9	10.2	13	6
Polymesoda caroliniana (marsh clam)	−22.0	11.3	8.5	1	1
Lagodon rhomboides (pinfish)	−22.0	12.4	10.1	7	4
Rangia cuneata	−21.9	12.0	7.2	1	1
Ischadium recurvum (hooked mussel)	−21.9	12.4	6.6	6	4
Geukensia demissa (ribbed mussel)	−21.8	11.2	7.3	2	2
Littorina irrorata (marsh periwinkle)	−21.6	14.3	–	4	–
Crassostrea virginica (American oyster)	−21.4	13.9	8.1	6	3
Brevoortia patronus (Gulf menhaden)	−21.3	14.5	11.3	7	6
Elops saurus (ladyfish)	−21.3	10.5	–	2	–
Archosargus probatocephalus (sheepshead)	−21.2	12.4	10.0	6	6
Pogonias cromis (black drum)	−21.1	8.2	8.4	1	1
Brevoortia patronus roe	−21.1	14.0	10.0	1	1
Leiostomus xanthurus (spot)	−21.0	9.9	11.2	7	7
Penaeus setiferus (white shrimp)	−21.0	9.9	–	15	–
Neanthes spp. (nereid worms)	−20.9	8.2	8.2	1	1
Anguinella palmata (bryozoan)	−20.8	13.5	8.1	1	1
Stellifer lanceolatus (star drum)	−20.8	11.5	10.2	1	1
Cynoscion nebulosus (speckled trout)	−20.7	12.2	11.8	11	1
Mugil cephalus roe	−20.6	10.8	7.1	1	1
Anchoa mitchilli (bay anchovy)	−20.6	14.5	12.0	3	2
Penaeus aztecus (brown shrimp)	−20.5	10.7	–	10	–
Cynoscion arenarius, larval/juv. (white trout)	−20.4	12.8	11.0	2	2
Bugula sp. (bryozoan)	−20.4	14.0	8.9	1	1
Arius felis (hardhead catfish)	−20.3	12.7	11.5	11	7
Brevoortia patronus, larval	−20.3	10.0	–	3, 2	–
Membras/Menidia, juv.	−20.2	13.6	–	1	–
Lolliguncula brevis (estuarine squid)	−20.2	13.2	11.1	2	2
Menidia beryllina (tidewater silverside)	−20.2	12.3	11.2	10	7
Membras martinica (rough silverside)	−20.2	16.0	11.9	3	1
Larval clupeids	−20.1	15.4	11.5	3	3
Paralichthys lethostigma (southern flounder)	−20.0	10.9	9.7	6	2
Sciaenops ocellatus (redfish)	−20.0	9.8	10.2	9	7
Symphurus plagiusa (blackcheek tonguefish)	−20.0	9.3	–	1	–
Membras martinica, larval/juv.	−20.0	14.4	11.5	1	1
Clibanarius vittatus (striped hermit crab)	−19.6	14.2	9.4	2	1
Callinectes sapidus (blue crab)	−19.6	10.7	8.6	10	5
Anchoa nasuta (longnose anchovy)	−19.5	14.0	12.4	1	1
Bairdiella chrysura (silver perch)	−19.4	13.1	12.2	7	4
Paleomonetes pugio (grass shrimp)	−19.4	6.2	8.6	4, 3	2
Mugil cephalus, juv.	−19.3	10.3	6.4	2	1
Fundulus pulvereus (bayou killifish)	−19.1	7.3	7.8	1	1
Mugil cephalus (striped mullet)	−18.9	10.3	8.0	7	1
Menticirrhus sp. (kingfish)	−18.8	11.5	11.1	1	1
Sciaenops ocellatus, juv.	−18.7	12.9	–	1	–
Fundulus majalis (longnose killifish)	−18.6	7.8	9.6	3	2
Larval spot/croaker	−18.6	14.6	–	1	–
Poecilia latipinna (sailfin molly)	−18.1	10.0	6.2	1	1
Fundulus grandis (Gulf killifish)	−18.1	9.6	–	1	–
Polinices duplicatus (moon snail)	−18.1	10.5	8.4	1	1
Harengula pensacolae (scaled sardine)	−17.8	16.4	12.1	1	1
Uca spp. (fiddler crabs)	−15.0	14.9	7.2	1	1
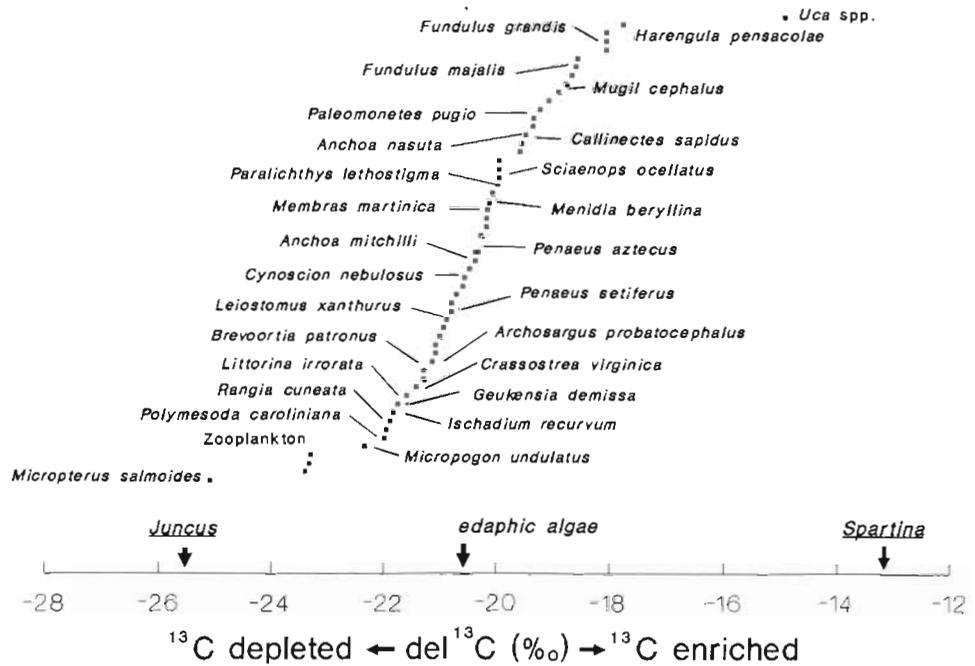
Fig. 1. $\delta^{13}C$ values of consumers in Graveline Bay Marsh

^{13}C depleted $\leftarrow$ del ^{13}C (‰) $\rightarrow$ ^{13}C enriched
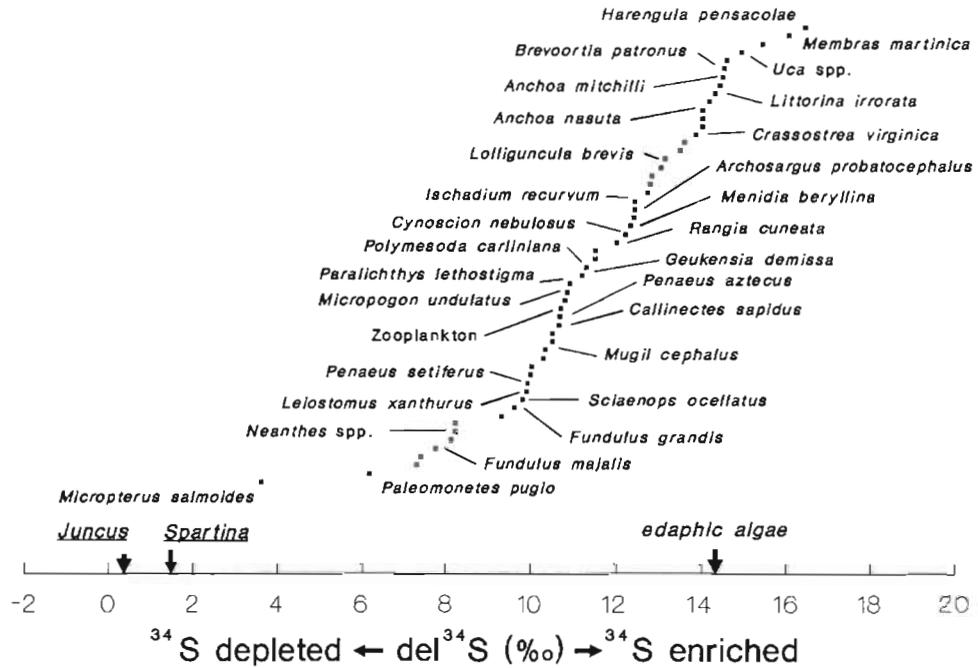
Fig. 2. $\delta^{34}S$ values of consumers in Graveline Bay Marsh

^{34}S depleted $\leftarrow$ del ^{34}S (‰) $\rightarrow$ ^{34}S enriched

ranged from +9 to +16‰. The freshwater bass *Micropterus salmoides* was an outlier with a $\delta^{34}S$ value of 3.6‰.

Table 2 and Fig. 3 summarize the $\delta^{15}N$ data for consumers; such data was obtained for 45 of the 56 consumer categories. $\delta^{15}N$ for the fauna ranged from +6 to +12‰; thus consumers were enriched in ^{15}N relative to edaphic algae, vascular plants, and zooplankton. As a group, invertebrates had lower $\delta^{15}N$ values than did the fish species sampled.

Dual isotope plots

Dual isotope plots of $\delta^{13}C$ vs $\delta^{34}S$ (Fig. 4) and $\delta^{13}C$ vs $\delta^{15}N$ (Fig. 5) were generated from the pool of stable isotope data. Each plot includes the stable isotope values for the consumer categories listed in Table 2 in addition to those of *Spartina*, *Juncus*, and edaphic algae. Error bars were not plotted since only a single value for the edaphic algae was obtained. A plot of
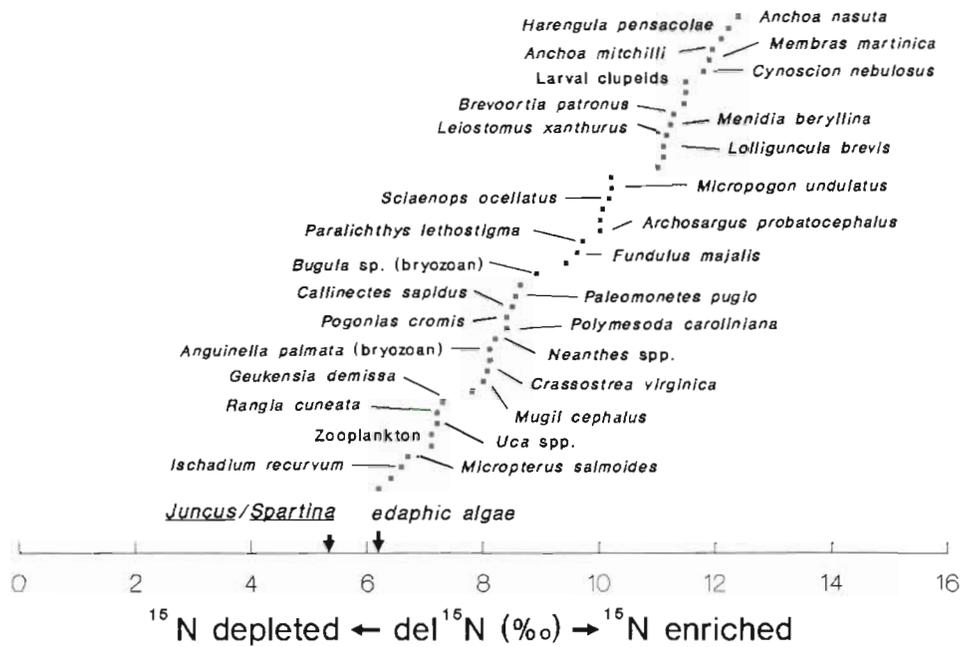
Fig. 3. δ^{15}N values of consumers in Graveline Bay Marsh

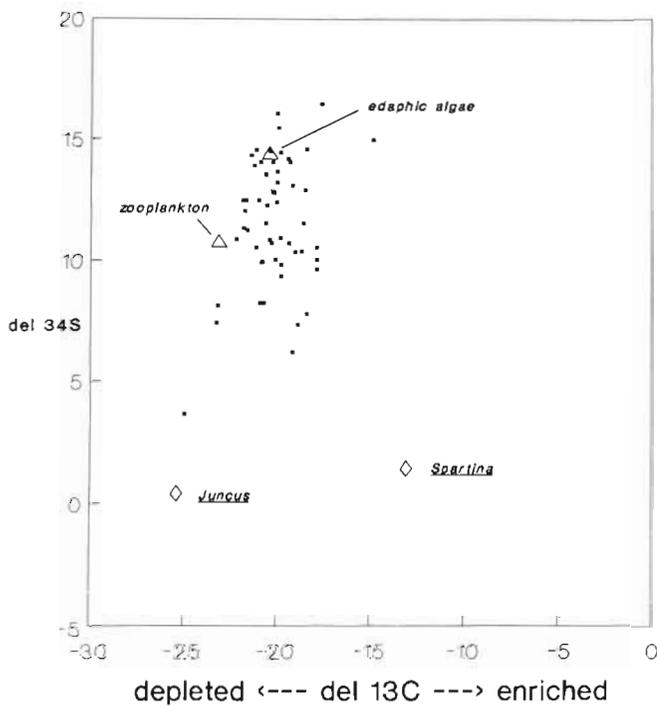^{15}N depleted ← del^{15}N (‰) →^{15}N enriched

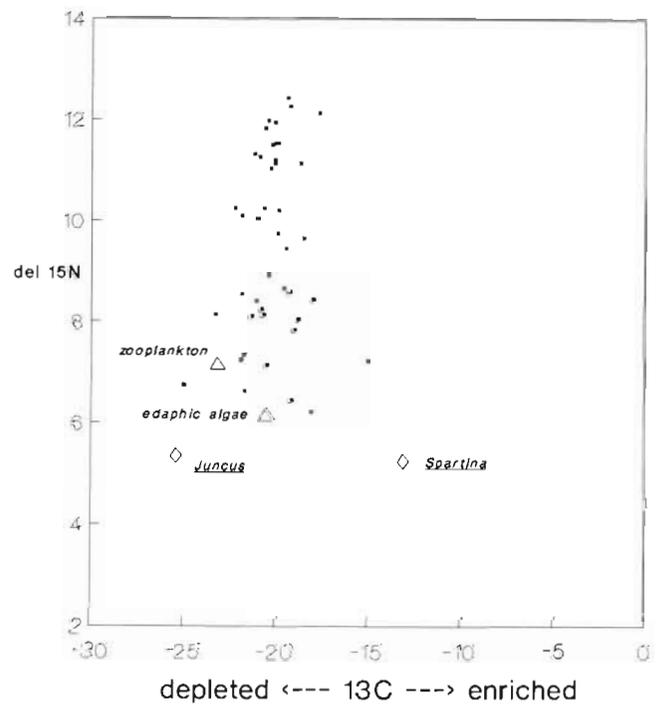Fig. 4. Plot of δ^{13}C vs δ^{34}S values for primary producers and consumers in Graveline Bay Marsh in parts per mil (‰)

Fig. 5. Plot of δ^{13}C vs δ^{15}N values for primary producers and consumers in Graveline Bay Marsh in parts per mil (‰)

δ^{34}S vs δ^{15}N is not shown as the 2 plots listed above were sufficient for interpretation of the data. Our discussion is centered on the plot employing δ^{13}C and δ^{34}S values (Fig. 4), the most reliable indicators for sources of ingested and assimilated organic matter, since isotope shifts of +4 to +5‰ may occur during

assimilation of nitrogen (Peterson & Howarth 1987, Fry 1988).

Data points in the δ^{13}C vs δ^{34}S plot (Fig. 4) cluster tightly around the edaphic algae and zooplankton. None of the consumers even moderately resemble the δ^{13}C and δ^{34}S values for either *Spartina* or *Juncus*. The

single data point near *Juncus* is that for the freshwater largemouth bass *Micropterus salmoides*, a non-resident species. Only 3 consumers (*Paleomonetes pugio*, *Fundulus pulvereus*, and *F. majalis*) appeared to exhibit any displacement toward *Spartina* (Fig. 4). These species are characteristically found in shallower areas well within the marsh where bacterial degradation of *Spartina* may affect the stable isotope ratios of available pools of organic matter. Neglecting these consumers, the distribution of $\delta^{13}C$ and $\delta^{34}S$ values indicated that consumers in the Graveline system for the most part closely match the $\delta^{13}C$ and $\delta^{34}S$ values of the benthic algae and zooplankton. *Uca* spp. (fiddler crabs) were nearly 6‰ more enriched in $\delta^{13}C$ than the edaphic algae; however, their $\delta^{34}S$ value of +15‰ almost exactly matched that of edaphic algae and was more than 13‰ higher than the mean $\delta^{34}S$ value of *Spartina*. The algal-grazing marsh periwinkle *Littorina irrorata* differed from the edaphic algae only in its $\delta^{13}C$ value, which was only 1‰ less.

The $\delta^{13}C$ vs $\delta^{15}N$ plot (Fig. 5) is less amenable to interpretation because the $\delta^{15}N$ values of the primary producers differ by less than 1‰ (Table 1). It does, however, show the tight clustering of $\delta^{13}C$ values around the edaphic algae and zooplankton and hence corroborates the results shown in Fig. 4.

DISCUSSION

Stable isotope ratios of producers

The $\delta^{13}C$ values of *Spartina* and *Juncus* fell within previously reported ranges. However, edaphic algae in Graveline Bay Marsh were more depleted in ^{13}C than previously reported with a $\delta^{13}C$ value of −21‰. Haines (1976a) measured values of −18 to −16‰ for mudflat algae in a Georgia salt marsh. Peterson & Howarth (1987) reported a value of −17‰ for creekbank algae in a Georgia marsh, whereas Rodelli et al. (1984) found ranges of −20 to −18‰ for 4 benthic diatom species and −22 to −18‰ for filamentous green algae attached to the roots of Malaysian mangroves. Craft et al. (1988) measured $\delta^{13}C$ values of −19 to −14‰ for benthic and floating algae in a North Carolina salt marsh.

As previously mentioned, a pure zooplankton sample was obtained which had a $\delta^{13}C$ value of −23‰. Gearing et al. (1984) found from extensive sampling in Narragansett Bay that the zooplankton were enriched in ^{13}C relative to phytoplankton by 0.6‰. If this is also the case in the Mississippi marsh then the phytoplankton would be −24‰. The filter-feeding bivalves (*Polymesoda caroliniana*, *Rangia cuneata*, *Ischadium recurvum*, *Geukensia demissa* and *Crassostrea virginica*) exhibited a very narrow range of −22 to −21‰; this

range is slightly more enriched in ^{13}C than the zooplankton in the Graveline system and approximately the same as that of phytoplankton in most estuarine systems (Gearing et al. 1984).

The average $\delta^{34}S$ value for *Spartina* (+1.4‰) in Graveline Bay Marsh fell amongst mean values measured by Peterson and co-workers (1985, 1986, 1987), which ranged from −4 to +1‰ in Massachusetts and Georgia salt marshes. A single value of +6‰ for *Juncus* is all that existed before the present study (Peterson & Howarth 1987). *Juncus* in Graveline Bay Marsh had an average $\delta^{34}S$ value of +0.4‰. Edaphic algae with their value of +14‰ compared favorably to $\delta^{34}S$ values of +13 and +18‰ measured by Peterson et al. (1986) for filamentous blue-green algal mats in a Massachusetts salt marsh. These algae must therefore obtain most of their sulfur as seawater sulfate which has a $\delta^{34}S$ value of +20‰ rather than as depleted sulfides which typically range from −30 to −10‰. Zooplankton were +11‰ which is much lower than the values of +19 and +20‰ measured by Peterson et al. (1985, 1986). All detritus was hand-picked from the zooplankton sample so the question naturally arises as to whether or not phytoplankton in the Graveline system are more depleted in ^{34}S than their Atlantic counterparts. A pure phytoplankton sample was not collected in the present study but the filter-feeding bivalves in the marsh were only slightly enriched in ^{34}S relative to the zooplankton (range = +11 to +12.5‰ for 4 species and +14 for the oyster, see Table 2). This suggests that the $\delta^{34}S$ value for the zooplankton may be very close to that actually characterizing the phytoplankton in the Graveline system, despite its low $\delta^{34}S$ value.

Stable nitrogen isotope values for *Spartina* and *Juncus* were within previously reported ranges. The $\delta^{15}N$ value (+6‰) for edaphic algae was higher than values of +4‰ (Mariotti et al. 1983, Peterson & Howarth 1987) and +1‰ (Craft et al. 1988) reported in other salt marshes. The zooplankton, with their $\delta^{15}N$ value of +7‰, were more depleted than the value of 9‰ employed by Peterson and co-workers (1985, 1986, 1987) for phytoplankton in their stable isotope work. Presumably the phytoplankton in the Graveline system should have a $\delta^{15}N$ value equal to or less than that of the edaphic algae.

Stable isotope ratios of consumers

Consumers collected in Graveline Bay Marsh were more depleted in ^{13}C than their counterparts in a Massachusetts (Great Sippewissett) and Georgia (Sapelo Island) salt marsh. Forty-nine of the 56 (88 %) consumers listed in Table 2 (see also Fig. 1) had $\delta^{13}C$ values between −22 and −18‰. Of these 49 consumers, 34 fell within a narrower range of −22 to −20‰. At

Great Sippewissett, $\delta^{13}C$ values for the marsh fauna fell between −17 and −10 ‰ (Peterson et al. 1986), while in Sapelo Island the range for consumers was −18 to −15 ‰ (Peterson & Howarth 1987). The $\delta^{13}C$ values of the Mississippi consumers therefore do not even overlap those of their counterparts in the 2 Atlantic salt marshes. With few exceptions, all consumers were more enriched in ^{13}C than zooplankton and their $\delta^{13}C$ values centered around that for the edaphic algae (Fig. 1). As previously mentioned, $\delta^{13}C$ for all benthic algae in the Graveline system ranged from −21 to −18 ‰, which would include the majority of values for consumers.

A word of caution must be expressed when making comparisons with the stable carbon isotope work of Peterson and co-workers (1985, 1986, 1987) as *Spartina* was the dominant vascular plant with *Juncus* a minor component of the flora in their studies. In Graveline Bay Marsh *Juncus* is predominant but *Spartina* nevertheless still contributes significantly to total marsh production. Another important point is that many of the animals collected in the present study were subtidal species. It is thus worthwhile to compare our results with those of Hughes & Sherr (1983), who examined $\delta^{13}C$ values of subtidal consumers from tidal creeks draining either *Spartina*- or *Juncus*-dominated marshes in Georgia. Seven invertebrates and 11 fish were sufficiently similar, i.e. same species or genus or ecological niche, for comparisons to be made. The average $\delta^{13}C$ value of subtidal consumers of this study was −20.5, while values of −20.3 and −18.1 ‰ were found by Hughes & Sherr for subtidal consumers of *Juncus* and *Spartina* tidal creeks, respectively. Thus, the specific identity of the dominant vascular plant may influence the $\delta^{13}C$ of resident consumers. It should be mentioned, however, that the *Juncus*-dominated creeks studied by Hughes & Sherr were more influenced by freshwater flow than was the *Spartina* creek, and this may have been a factor in explaining the observed differences.

The consumers in Graveline Bay Marsh were slightly more enriched in ^{34}S than their counterparts from a Georgia salt marsh. Forty-eight of the Mississippi consumers (86 % of total) fell within a range of +9 to +16 ‰ (Table 2, Fig. 2). Most of the marsh fauna at Sapelo Island had $\delta^{34}S$ values between +6 and +14 ‰ (Peterson & Howarth 1987). Among the consumers with the highest $\delta^{34}S$ values were *Uca* spp., *Littorina irrorata*, and *Crassostrea virginica*; this was also true in our study. Peterson & Howarth considered it unlikely that phytoplankton were the main source of sulfur for *Uca* spp. and *L. irrorata* but that edaphic and epiphytic algae, which also obtain most of their sulfur from seawater sulfate, were their major ultimate source of organic matter This is also probably true of the majority of consumers in the Graveline system, as 86 % of all consumers were 8 to 15 ‰ more enriched in ^{34}S than *Spartina*. Finally, Peter-

son et al. (1986) found that all fauna in the Great Sippewissett salt marsh had $\delta^{34}S$ values between 0 and 10 ‰. Such values are intermediate between those for *Spartina* and for benthic and planktonic algae.

The only other study in which comprehensive $\delta^{15}N$ measurements were made is that of Peterson & Howarth (1987) in the salt marshes of Sapelo Island. They observed that the $\delta^{15}N$ values for consumers (+7 to +11 ‰) centered around the phytoplankton (+9 ‰). In Graveline Bay Marsh, virtually all consumers (+6 to +12 ‰) were enriched in ^{15}N compared to the vascular plants (+5 ‰) and edaphic algae (+6 ‰), and all but 3 consumers were enriched relative to the zooplankton (+7 ‰). Peterson & Howarth (1987) found that fiddler crabs (*Uca* spp.) and small mullet (*Mugil cephalus*) generally had the lowest $\delta^{15}N$ values which was also true in our study.

Stable nitrogen isotope ratios are good indicators of trophic level whereas those of carbon and sulfur are not (Fry 1988). Peterson & Howarth (1987) found that consumers fractionate nitrogen by +1 to +5 ‰ per trophic transfer; fractionation of carbon or sulfur was slight and could be in either the positive or negative direction (see their Table 3). Peterson & Howarth divided the marsh fauna of Sapelo Island into 2 groups: one with $\delta^{15}N$ values less than 9 ‰ which included deposit and suspension feeders, and a second with $\delta^{15}N$ values greater than 9 ‰ which included predators and omnivores. Consumers belonging to the deposit/suspension feeding group were *Uca* spp., small *Mugil cephalus* (small mullet), *Geukensia demissa*, *Littorina irrorata*, and *Crassostrea virginica*. Consumers belonging to the predator/omnivore group were *Paleomonetes pugio*, *Pennaeus setiferus*, *Bairdiella chrysura*, *Callinectes sapidus*, *Fundulus heteroclitus*, and large *Mugil cephalus*. Such an arbitrary division would seem also to characterize the fauna of Graveline Bay Marsh quite well (see Table 2). The 5 filter-feeding bivalves belong to the first group as do *Uca* spp., *Neanthes* spp., and the bryozoans *Anguinella palmata* and *Bugula* sp. *C. sapidus* and *P. pugio* had $\delta^{15}N$ values very close to 9 ‰; however, both species are highly omnivorous and thus utilize a variety of food sources. All fish species except *Pogonias cromis*, *Poecilia latipinna*, and *Fundulus pulvereus* fall into the second predator/ omnivore group based on their $\delta^{15}N$ values which is generally consistent with their biology. Thus, $\delta^{15}N$ measurements appear to be an excellent indicator of a consumer's relative trophic level in the Mississippi marsh.

Dual isotope plots

In sharp contrast to previous studies (Peterson et al. 1986, Peterson & Howarth 1987), plots employing $\delta^{13}C$ and $\delta^{34}S$ values or $\delta^{13}C$ and $\delta^{15}N$ values of primary

producers and consumers did not result in a broad band of values lying between phytoplankton and *Spartina*. Instead, this dual isotope procedure for Graveline Bay Marsh produced a tight clustering of data points around the edaphic algae and zooplankton (Fig. 4 and 5). Consumers varied from moderately to greatly depleted in ^{13}C and enriched in ^{34}S relative to *Spartina*. The marsh periwinkle *Littorina irrorata*, which is known to graze on edaphic algae, was essentially identical to the latter in both its δ^{13}C and δ^{34}S values. The one outlier in this plot, the freshwater bass *Micropterus salmoides*, was only collected once at the periphery of the marsh system; it serves primarily to illustrate the discrete separation of the Graveline system from any bordering freshwater systems. A reasonable conclusion from the stable isotope data is that the major food sources for the fauna of Graveline Bay Marsh are the edaphic and planktonic algae; the contribution of vascular plants appears to be minor. This does not mean of course that all consumers are actually eating algae, but that the algae are the major basis of the food web in Graveline Bay Marsh. Kitting et al. (1984) used stable carbon isotope analysis to show that epiphytic algal carbon, rather than that of seagrasses, formed the primary basis of the food web in Texas seagrass beds; very little of the seagrass leaf biomass was ingested by any invertebrate species.

There is an alternative interpretation of the data. Observed δ^{13}C values for consumers in Graveline Bay Marsh could be due to ingestion of *Juncus* and other vascular plant detritus. It is possible that heterotrophic microorganisms degrading mineral-poor, i.e. high C:N:S ratio, plant detritus could take up their nitrogen and sulfur from the water column. Their δ^{15}N and δ^{34}S values would then resemble those of the algae while their δ^{13}C value would be similar to that of the vascular plant (*Juncus* = $-26‰$). Recent work in salt marsh tidal creeks has suggested that a major carbon pathway may exist from heterotrophic bacteria to protozoa (Sherr et al. 1989). Such protozoans (mainly ciliates and colorless flagellates) are readily grazed by microcrustaceans and fish larvae (Sherr & Sherr 1988). The high δ^{34}S values of the subtidal fauna of Graveline Bay Marsh argue for an algal-based food web, but in the absence of data on the abundance of *Juncus* detritus and stable isotope composition of its bacterial decomposers, the possibility discussed above cannot be discounted.

It is not possible to evaluate the relative importance of edaphic algae and phytoplankton since their δ^{13}C values are so close and the δ^{34}S value of the latter in Graveline Bay Marsh is not precisely known. Furthermore, the stable isotope values for edaphic algae are based on a single composite sample. One therefore, cannot be sure how representative these values are,

both temporally and spatially. What is evident, however, is that this represents the first study to show that vascular plant carbon may be of little direct trophic importance in at least some marsh systems, although the alternative hypothesis discussed above cannot be discounted. Hopefully, the data presented here will encourage other investigators to consider the benthic algae as a potentially important source of organic matter in future studies of trophic relationships in salt marshes and other estuarine systems. This becomes even more important when one considers that a significant proportion of water column productivity, i.e. 'planktonic' productivity, in shallow estuarine systems may sometimes be due to displaced benthic diatoms and other microalgae (Riaux-Gobin 1987, Shaffer & Sullivan 1988).

Regional comparisons

The only other studies employing multiple stable isotopes in salt marshes are those of Peterson et al. (1986) for Great Sippewissett Marsh in Massachusetts and Peterson & Howarth (1987) for Sapelo Island marshes in Georgia. The present study makes possible a comparison of a Gulf Coast marsh with 2 widely separated Atlantic marshes. It is profitable to begin with a comparison of the stable isotope values of consumers listed in both Table 2 of the present study and Table 4 of Peterson & Howarth (1987). All but one (*Uca* sp.) of the 11 species common to both studies were more depleted in ^{13}C in the Mississippi marsh. The Graveline Bay Marsh fauna was on average 2.7‰ more depleted than Sapelo Island fauna (δ^{13}C means = -19.8 and $-17.1‰$, respectively). The opposite was true for δ^{34}S; all but one (*Paleomonetes pugio*) Mississippi consumer were more enriched in ^{34}S than the same species in Georgia. The average enrichment was 2.1‰ (δ^{34}S means = $+11.6$ and $+9.5‰$, respectively). As previously mentioned, these differences in δ^{13}C and δ^{34}S may be due to the dominance of *Spartina* in Georgia and *Juncus* in Mississippi, or to freshwater influences. The average δ^{15}N value for these consumers was 8.9 and 8.4‰ in Mississippi and Georgia, respectively. The difference is only 0.5‰, which probably does not represent a significant enrichment and agrees with previous findings (Fry 1988) that δ^{15}N is most useful as an indicator of trophic level rather than food source.

As one moves from Massachusetts to Georgia to Mississippi, the following trends confront an observer (see also Table 5 of Peterson & Howarth 1987): (1) δ^{13}C values for *Spartina* stay constant at $-13‰$ whereas those for edaphic algae (and perhaps phytoplankton) become increasingly more negative; (2) the

marsh fauna becomes increasingly depleted in ^{13}C and increasingly enriched in ^{34}S; and (3) although δ^{15}N values for consumers increase slightly the differences are insignificant compared to those for carbon and sulfur. Peterson & Howarth (1987) interpreted these trends as indicating that consumers in the Sapelo Island marshes were more dependent on phytoplankton and perhaps benthic algae than the fauna at Great Sippewissett. They stated that the contributions of *Spartina* and algae to the food web at Sapelo Island were 'roughly equal' whereas their previous isotope work (Peterson et al. 1986) at Sippewissett led to the conclusion that *Spartina* was the major food source but phytoplankton were also important. The benthic algae were not considered in their earlier study. In the Mississippi marsh we move more or less 180 degrees from the conclusions of the Sippewissett study in that planktonic and benthic algae are major food sources and the vascular plants presumably play a minor role.

Why the relative importance of algae and vascular plants should change so dramatically as one moves south along the Atlantic Coast and then west to the Gulf Coast is an important question, since it relates to possible differences in the functioning of salt marshes from different geographical regions. Peterson & Howarth (1987) hypothesized that algae would be expected to be more important in Georgia than in Massachusetts if their productivity and availability relative to *Spartina* were higher in the more southerly marsh. The productivity rates of edaphic algae on the marsh proper (Sullivan & Moncreiff 1988) and in the subtidal sediments of Graveline Bayou and lower-order tidal creeks (Zimba unpubl. data) are significant but are less than those measured by Pomeroy (1959) in Sapelo Island marshes. These algae are known to be the preferred food source for a host of estuarine consumers at lower trophic levels including harpacticoid copepods, nematodes, ostracods, mollusks, juvenile shrimp, and certain fish species (Gleason and Zimmerman 1984, Montagna 1984, Gleason 1986, Plante-Cuny & Plante 1986, Decho 1988). It should be pointed out that in addition to the relative dominance of *Spartina* vs *Juncus,* there are differences in basic hydrology between the 3 systems discussed above. Such differences include tidal range, regularity of tidal flooding, and possible freshwater and upland terrestrial inputs. Factors of this type could also be key components leading to the observed differences in stable isotope ratios. In addition, climatic factors affecting rates of biological activity and seasonal changes within each system may contribute to the observed regional shifts in isotopic ratios. However, the bottom line is that benthic algae should not be ignored in estuarine food web studies.

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