

# Use of chemosynthetic biomass by large, mobile, benthic predators in the Gulf of Mexico

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**ABSTRACT:** The Gulf of Mexico supports communities of chemoautotrophic and heterotrophic fauna associated with hydrocarbon seeps. The chemoautotrophic invertebrates, mostly dense epifaunal assemblages of mussels and tubeworms, derive their nutrition from symbiotic relationships with sulfide- or methane-oxidizing bacteria. The extent to which benthic predatory fauna feed on the chemoautotrophic assemblages has been an open question. Owing to differences in stable isotope values between chemosynthetic- and photosynthetic-derived biomass, isotopic characterization of predatory fauna would be useful in determining their dependence upon chemoautotrophs for food. Carbon and sulfur stable isotope analyses reveal that fishes with similar feeding strategies (rat tail [*Nezumia* sp.] and eels [*Synaphobranchus* sp., *Ophichthus cruentifer* and *Dysomma rugosa*]) have similar  $\delta^{13}\text{C}$  values ( $-32.7$  and  $-42.5\%$ , respectively), reflecting chemoautotrophic carbon. Large vagrant predators/scavengers such as isopods *Bathynomus giganteus*, hagfish *Eptatretus* sp. and spider crabs *Rochina crassa*, have isotope values closer to oceanic ranges ( $\delta^{13}\text{C}$ :  $-20$  to  $-18\%$ ;  $\delta^{34}\text{S}$ :  $18$  to  $20\%$ ), although some individual *Eptatretus* sp. and *R. crassa* show a chemosynthetic component. Colonist invertebrates, such as the sea star (*Sclerasterias* cf. *tanneri*) and a predatory snail *Buccinum canetae*, have greatly depleted  $\delta^{13}\text{C}$  and  $\delta^{34}\text{S}$  values, indicating an almost 100% reliance on seep production. Nitrogen isotope ratios are depleted ( $-19\%$ ) in some seep areas, and *B. canetae* and *Synaphobranchus* sp. reflect the  $^{15}\text{N}$  depletion ( $-10$  and  $-3\%$  respectively). On a species-specific basis, some mobile benthic predators from the background fauna obtain close to 100% of their nutrition from seep production, indicating that they are residents of the seeps. There is a high degree of movement in and out of the seep habitat by vagrant benthic predators, and although the majority derive most of their nutritional needs from photosynthetic production, the incorporation of chemosynthetic production is substantial.

**KEY WORDS:** Chemosynthetic production · Benthic marine predators · Stable isotopes

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## INTRODUCTION

The hydrocarbon seep communities of the deep Gulf of Mexico are fundamentally similar to those of other deep chemosynthetic systems, supporting dense assemblages of chemoautotrophic-based tube worms and mussels occupied by a more diverse group of heterotrophic species (Kennicutt et al. 1985, Brooks et

al. 1987). These very high standing-stock communities are surrounded by a low standing-stock background community of typical deep-sea species with a phytoplankton detritus-based food web. A fundamental question addressed herein arises from the dramatic contrast in apparent productivity of the 2 communities: to what extent do the background species exploit the chemosynthetic production? Indeed, the paucity of food in the deep environment is thought to be so pronounced and persistent that there is extreme competition for food on both ecological and evolutionary time scales (Jumars et al. 1990, Gage & Tyler 1996). Upon

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initial consideration, a high degree of predation by the background fauna would be expected.

Two possible types of exploitation by background species have already been established from the species composition of hydrothermal vents and cold seeps: (1) feeding by a resident community of consumers, and (2) feeding by vagrant species (Tunnicliffe & Jensen 1987, Tunnicliffe 1992). Resident species would be expected to be part of a relatively closed food web, foraging as resident populations only within a seep or vent community. Vagrant species would bridge the chemosynthetic system and the background, forming no resident populations within seep or vent sites, but foraging across their extensive photosynthetically-based background range with forays into the chemosynthetic systems. Owing to the relatively well-established taxonomic database for the deep Gulf of Mexico, it has been possible to distinguish background species from seep residents. In addition to populations of true seep endemic species, the resident consumer community contains species which have colonized from the background. Such colonists are typically rare in the background, and appear to have elevated population densities within the seep sites (Carney 1994). It was the purpose of this study to establish the degree of seep primary-production utilization by both types of mobile predators and to gain a better understanding of trophic export by the vagrant consumer route.

The results of a prior isotopic survey of deep Gulf of Mexico benthos gave initial evidence that trophic export due to vagrant consumption may be substantial (Kennicutt unpubl. data). That study, initiated prior to the discovery of the seeps, collected fauna with a 9 m semi-balloon otter trawl along 4 transects spanning 320 to 2953 m depth and lying between 27° 35.0' N, 93° 33.1' W; 26° 17.2' N, 93° 19.2' W; 28° 27.2' N, 86° 01.0' W; and 28° 04.4' N, 86° 34.8' W (Pequegnat et al. 1990). The slope megafauna (132 specimens of 42 species) showed a well-defined  $\delta^{13}\text{C}$  distribution centered at  $-17.8 \pm 1.2\%$  consistent with a photosynthetic plankton food source, along with an indication of a second cluster of 22 values centered at a more depleted  $-20.8\%$ . Interpretation of the lower values as being due to some dependence on a chemosynthetic food source was strengthened by the identity of the species collected (seep chemosynthetic production may be  $^{13}\text{C}$ -depleted relative to photosynthetic production, see below). Of the five  $\delta^{13}\text{C}$ -depleted species, 3 are now known to be seep vagrants—the large mobile crab *Chaceon* (Geryon) *quinquedens*, the eel *Synaphobranchus* sp., and the hake *Urophycis cirratus*. Since none of the trawl samples contained seep endemics, it appears that the 17% of the total catch that had indications of seep-derived carbon could be the result of trophic export into the background. The lack of multi-

ple isotope tracers and of information as to seep proximity, however, makes such a conclusion tenuous (for a review of stable isotopic analysis and ecological studies see Lajtha & Michener 1994).

The Gulf of Mexico hydrocarbon seep communities have been observed and sampled for more than 10 yr, resulting in substantial on-site information on species composition, classification of endemics, colonists, and vagrants, and food-web ecology (MacDonald et al. 1989, Carney 1994, Fisher 1996, Kennicutt unpubl. data). The biotic substrate of the communities is formed by various combinations of abundant tube worms and bivalves that harbor chemosynthetic symbiotic bacteria. Two tube worm species, a *Lamellibranchia* species and an *Escarpia*-like species, as well as the comparatively rare mussel *Tamu fisheri* and the peripheral lucinid and vesicomid clams are all symbiotic with sulfur-oxidizing chemoautotrophic bacteria (Brooks et al. 1987, Fisher et al. 1993). The mussel *Bathymodiolus childressi* is the most abundant bivalve at the seeps and it is symbiotic with methanotrophic bacteria (Childress et al. 1986). Closely associated with the tube worm bushes and mussel beds are at least 2 dozen heterotrophic species, dominated by endemic grazing gastropods, smaller decapod crustaceans, and worms of several phyla. Mussel beds usually lack sessile epizoa but, poriferans and hydroids are common on tube worm tubes (Maldonado & Young 1988, Carney 1994). For mussel beds the predominant colonists are both predators—the gastropod *Buccinum canetae* and the sea star *Sclerasterias* cf. *tanneri*. On-site, larger more motile predatory animals classified as vagrants include the hagfish *Eptatretus*, the large spider crab *Rochina crassa*, the eels *Synaphobranchus*, *Ophichthus cruentifer* and *Dysomma rugosa*, the rat-tail fish *Nezumia* sp., and the giant isopod *Bathynomus giganteus*.

The most useful tool for determining the past consumption of any animal sampled on- or off-site is stable isotope analysis. The value of such tracers for determining the nutrient source utilization by and trophic interactions among consumers has been well established in many terrestrial and aquatic systems (for review see Fry & Sherr 1984, Michener & Schell 1994). Consumers reflect the  $\delta^{13}\text{C}$  of their food source (which can differ depending on the process of  $\text{CO}_2$  fixation and  $[\text{CO}_2]$ ) plus approximately 1‰ accounting for the trophic fractionation (Fry & Sherr 1984). Therefore, in systems where isotopically distinct nutrient sources are available the relative importance of sources to consumer species can be determined. Trophic level can be determined among consumer species with  $\delta^{13}\text{C}$  because of the 1‰ enrichment relative to their diet. Nitrogen isotopes can also be used to differentiate nutrient sources and provide an even more definitive

indication of trophic level. Marine and estuarine phytoplankton and their consumers are enriched by several ‰ relative to terrestrial autotrophs. Additionally, there is a 3 to 3.5‰ enrichment in  $\delta^{15}\text{N}$  per trophic level (Michener & Schell 1994 and references therein). Therefore, both carbon and nitrogen isotopes combined can provide nutrient source and trophic information. Sulfur isotopes are also proving useful as nutrient tracers in systems where marine and freshwater nutrients mix (Peterson & Howarth 1987, Hesslein et al. 1991, MacAvoy et al. 1998). Ocean phytoplankton have a fairly uniform  $\delta^{34}\text{S}$  value of about 18.5 to 19‰ (Peterson & Howarth 1987). Terrestrial autotrophs are depleted relative to phytoplankton, and the isotopic composition of  $\text{H}_2\text{S}$  in reducing sediments can reach values of  $-20\%$  or lower (Michener & Schell 1994). Additionally,  $\delta^{34}\text{S}$  is thought to undergo minimal or no fractionation as a function of trophic level, making it an excellent tracer for nutrients since no correction due to trophic fractionation is needed.

There is a special utility for stable isotope analysis in deep-sea chemoautotrophic systems owing to the distinctiveness of the produced biomass. It was the fact that vent tube worms had very distinct  $\delta^{13}\text{C}$  values relative to typical deep-ocean tissue carbon values, that led Rau & Hedges (1979) to postulate that the dominant invertebrates had some non-photosynthetic food source. In hydrocarbon seep communities,  $\delta^{13}\text{C}$  has been the primary isotope value examined. It has been used to help differentiate animals with chemoautotrophic symbionts ( $-20$  to  $-40\%$ ) from those with methanotrophic symbionts ( $\leq -40\%$ ) (Brooks et al. 1987, Kennicutt et al. 1992) and to identify the source methane pool as either thermogenic ( $\delta^{13}\text{C} = -40$  to  $-45\%$ ) or biogenic ( $\delta^{13}\text{C} \leq -45\%$ ) methane (Sassen et al. 1999). Some  $\delta^{15}\text{N}$  and  $\delta^{34}\text{S}$  measurements have also been made on seep chemoautotrophs and heterotrophs (Brooks et al. 1987). Chemoautotrophs tended to have lower  $\delta^{15}\text{N}$  values ( $-5$  to  $-12\%$ ) than heterotrophs (2.8 to 13‰) or marine phytoplankton. The  $\delta^{34}\text{S}$  values of the thiotrophic animals were depleted relative to the methanotrophic bivalves (13‰) and seep heterotrophs (13 to 16‰) (Brooks et al. 1987). Because of the large and significant differences in isotope signature between photosynthetically derived biomass in the ocean and chemosynthetically derived biomass at the seep, examination of the isotopic ratios of predators in and around the seep should reveal their degree of utilization of seep primary production.

## MATERIAL AND METHODS

On- and off-site sampling of fauna was carried out on 1997 and 1998 cruises of the RV 'Edwin Link' and the

'Johnson Sea Link' submersible in the Green Canyon Lease Area region of the Gulf of Mexico. This is a 22 000 km<sup>2</sup> region of seafloor under US economic control lying between the 200 and 2400 m isobaths on the topographically and geochemically complex continental slope off the coast of Louisiana. For the management of oil and gas development the area is subdivided into 4.8 × 4.8 km numbered lease blocks (Fig. 1). The sites sampled for this study were in lease blocks Green Canyon 185: GC 185, 'Bush Hill'; GC 233: 'Brine Pool'; and GC 234. These sites are active hydrocarbon seep areas with chemoautotrophic fauna that have been studied for various purposes for more than a decade.

The GC 185 (Bush Hill) site (27° 46.96' N, 91° 30.46' W) is at a depth of 540 to 580 m. The dominant fauna at the site are tube worms, although mussel beds are found throughout the site as well. Methane and oil are actively seeping from the sediments and carbonate outcrops are scattered over the site (Nix et al. 1995). The GC 234 site (27° 44.7' N, 91° 13.3' W) is at a depth of approximately 540 m. Similar to GC 185, fauna at this site is dominated by tube worms with abundant mussel beds. Methane gas and oil has been observed leaking from sediments (Nix et al. 1995). The GC 233 site (27° 43.4' N, 91° 16.8' W) is at a depth of 640 m. It is dominated by an anoxic brine pool that arises from saline seepage along a fault (Reilly et al. 1996, Sassen et al. 1999). Biogenic methane-utilizing *Bathymodiulus childressi* mussels are the dominant fauna at the site, tube worms are scarce (MacDonald et al. 1990, Smith et al. 2001).

On-site collection of mobile predatory fauna was carried out with small (approx. 3 × 4 × 2 ft: 0.91 × 1.22 × 0.61 m) wire-mesh traps deployed within and around tube worm and mussel beds at GC 234 and GC 233. Trap collections were augmented by direct capture by the submersible via a suction sampler and bushmaster collection nets (Urcuyo et al. 2001). Off-site mobile predators were caught with surface-deployed Z-frame traps, 5 × 6 × 3 ft (1.52 × 1.83 × 0.91 m), set approximately 2 km off the location of the seep communities in areas known from prior surveys to lack active seep communities. Both small and Z-traps were intended to capture the same types of animals. They both were constructed of 1 inch (2.54 cm) square trap mesh, equipped with two 20 cm entry mouths, and baited with menhaden *Brevoortia tyrannus* in a wire bait cage to minimize consumption. Trap soak time (duration of deployment) was determined by cruise logistics, ranging from 1 to 6 d.

Samples of muscle tissue were taken from captured organisms and kept frozen until shipment to the University of Virginia for analysis. Isotopic determinations were made on samples dried at 60°C for 3 d and homogenized. Approximately 5 to 6 mg of tissue was

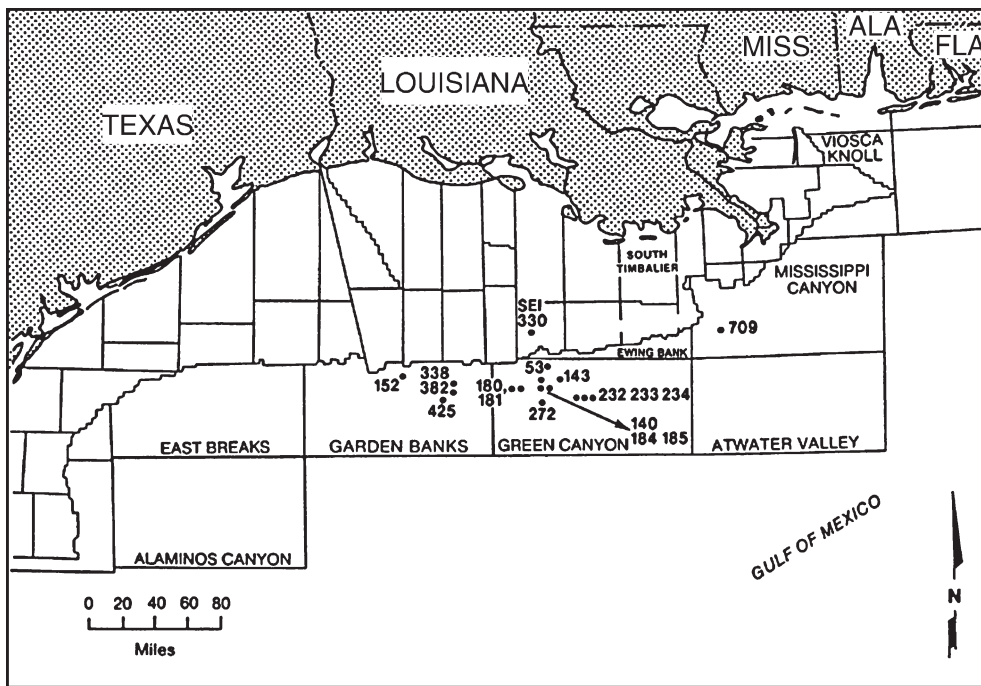


Fig. 1. Locations of the Green Canyon (GC) and Garden Banks lease areas. Chemosynthetic communities and their associated predators at GC 185, GC 233 and GC 234 were the focus of this study (adapted from Roberts & Carney 1997)

used for  $\delta^{34}\text{S}$  measurements and 0.6 to 1.0 mg was used for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  measurements. Computation of the delta value follows the same procedure for all stable isotopic measurements, as follows:

$$\delta^xE = \left[ \frac{(^xE/^yE)_{\text{sample}}}{(^xE/^yE)_{\text{standard}}} - 1 \right] \times 1000 \quad (1)$$

where  $E$  is the element analyzed (C, N or S),  $x$  is the atomic weight of the heavier isotope, and  $y$  is the atomic weight of the lighter isotope ( $x = 13, 15, 34$  and  $y = 12, 14, 32$  for C, N and S respectively). The standard materials to which the samples were compared were PDB (Pee Dee Belemnite) for carbon, air  $\text{N}_2$  for nitrogen and CDT (Canyon Diablo Triolite) for sulfur. A Carlo Erba elemental analyzer coupled to a VG Optima isotope ratio mass spectrometer (Micromass<sup>®</sup>, Manchester, UK) was used for all isotopic measurements (EA/IRMS: Fry et al. 1992, Giesemann et al. 1994). Reproducibility of all measurements was typically 0.3‰ or better.

Since vagrant species are expected to have a diet combining seep production with phytodetritus organisms, analyses included assessment of food-source mixing. Isotopic mixing equations can be used to quantify food source dependence in systems with isotopically distinct food sources. The general form of the mixing equation used is:

$$\delta^xE_{\text{predator}} - F = (\delta^xE_{\text{seep}} \times f_{\text{seep}}) + (\delta^xE_{\text{ocean}} \times (1 - f_{\text{seep}})) \quad (2)$$

where  $f_{\text{seep}}$  is the fraction of diet from chemosynthetic sources and  $\delta^xE_{\text{seep}}$  and  $\delta^xE_{\text{ocean}}$  are the mean isotopic

signatures of the chemosynthetic material and background ocean respectively. The parameter  $F$  corrects for trophic enrichment and is dependent on the isotope used.

The values used for the mean isotopic signatures of the chemosynthetic prey,  $\delta^xE_{\text{seep}}$ , were calculated site-by-site, averaging values for selected resident fauna exclusive of vagrant species. The phytodetritus-based food sources of the background were considered to be relatively homogenous, and a single set of C, N and S isotope values determined from the literature were used in Eq. (2). The  $\delta^{13}\text{C}_{\text{ocean}}$  value of  $-17\text{‰}$  was used as being typical of values between phytoplankton and background predators (Fry & Sherr 1984, Peterson & Howarth 1987, Michner & Schell 1994, Roelke & Cifuentes 1997). The  $\delta^{15}\text{N}_{\text{ocean}}$  value of  $10\text{‰}$  was used (Fry 1983, Macko et al. 1984). The  $\delta^{34}\text{S}_{\text{ocean}}$  value of  $18.5\text{‰}$  was used (Peterson & Howarth 1987, MacAvoy et al. 1998). When using  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ,  $F$  was given a value of 1 and 3‰ respectively, which is the trophic enrichment typically associated with these isotopes. Sulfur isotopes do not significantly fractionate with trophic transfer (Michener & Schell 1994), therefore  $F = 0$  when using  $\delta^{34}\text{S}$ .

Nonparametric methods were used for all comparisons. Mann-Whitney  $U$ -tests were used for 2 group comparisons and Kruskal-Wallis tests were used for multiple comparisons. The Dunn procedure was used to compare specific differences among groups once Kruskal-Wallis indicated significant differences (Ros-

ner 1990, Lee 1992). Microsoft Excel 5.0 (Microsoft, Inc., Seattle) and Statview SE + Graphics (Abacus Concepts, Inc., Berkeley, CA) were used for individual statistical tests.

## RESULTS

On-site small and off-site Z-traps proved an effective means of collecting 3 vagrant species, the spider crab *Rochina crassa*, the giant isopod *Bathynomus giganteus*, and the hagfish *Eptatretus* sp. *R. crassa* is a true crab reported from 400 to 800 m deep on the upper slope from New England to the Gulf of Mexico (Pequegnat 1970). *Eptatretus* sp., a hagfish, is a cosmopolitan jawless fish found at all depths in the Gulf of Mexico. Although the thin hagfish could easily escape through the trap mesh, they sometimes entered the smaller bait cage and were retained during retrieval. *B. giganteus* is the largest living isopod, reaching a body size of 48 cm in our collections, and is an example of occasional evolutionary gigantism in deep-sea pericardid crustaceans attributed to niche-filling in the comparative absence of large predatory crabs and fishes (Gage & Tyler 1996). The species appears to be restricted to the Gulf of Mexico, Caribbean, and adjacent Atlantic slope, but the genus is cosmopolitan. The capture of other animals by the traps was quite limited, possibly due to voracious feeding in the trap by the giant isopods.

### Site GC 234

Site-specific values for the isotopic signatures of the seep food sources was developed from a selection of 3 symbiont-hosting and 5 resident heterotrophs (Table 1). The values obtained were  $-32.1 \pm 8.3\%$   $\delta^{13}\text{C}$  ( $n = 11$ ),  $6.1 \pm 3.2\%$   $\delta^{15}\text{N}$  ( $n = 11$ ), and  $4.8 \pm 2.8\%$   $\delta^{34}\text{S}$  ( $n = 3$ ). The mean chemosynthetic  $\delta^{13}\text{C}$  value of prey items at GC 234 was statistically indistinguishable from that at GC 185 and virtually identical when tube worms were excluded. For this reason, 1 value for  $\delta^{13}\text{C}_{\text{seep}}$  ( $-32 \pm 6\%$ ) was

Table 1. Stable isotope values for prey fauna from the 3 study sites. The mean and standard deviation of these values were used in isotope mixing equations to determine the exploitation of chemosynthetic production by large, heterotrophic fauna captured both on- and off-site

Species	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{34}\text{S}$
<b>GC 185</b>			
<i>Phascoloma turnerae</i> (sipunculid)	-27.9	6.0	-12.3
<i>Phascoloma turnerae</i> (sipunculid)	-32.5	8.9	2.2
<i>Neretid polychaete</i>	-29.9	5.4	-9.3
<i>Munidopsis</i> Type 1 (squat lobster)	-28.9	7.7	
<i>Munidopsis</i> Type 1 (squat lobster)	-7.5		
<i>Munidopsis</i> Type 1 (squat lobster)	-32.0	6.8	-2.1
<i>Munidopsis</i> Type 1 (squat lobster)	-30.1	6.9	-13.0
<i>Munidopsis</i> Type 1 (squat lobster)	-30.9	6.5	-4.4
Nemertian worm	-23.1	7.5	-6.1
<i>Alvinocaris stactophila</i> (shrimp)	-31.3	6.7	1.5
<i>Escarpia</i> -like species (tube worm)	-21.9	3.7	-33.7
<i>Bathynertia naticoidea</i> (snail)	-29.6	7.0	-3.4
<i>Bathynertia naticoidea</i> (snail)	-29.6	7.0	-3.4
<i>Bathynertia naticoidea</i> (snail)	-30.9	5.8	-7.7
<i>Harmathoe</i> sp. (red head polynoid worm)	-23.7	7.0	-12.6
<i>Eurythoe</i> sp. (hairy polychaete worm)	-40.6	8.1	-12.0
<i>Bathymodiolus childressi</i> (mussel)	-43.0	7.8	-4.1
<i>Bathymodiolus childressi</i> (mussel)	-40.6	8.3	-1.3
<i>Bathymodiolus childressi</i> (mussel)	-41.3	1.5	2.3
Mean $\pm$ SD	$-31.2 \pm 6.0$	$6.9 \pm 1.2$	$-7.3 \pm 8.4$
<b>GC 234</b>			
<i>Phascoloma turnerae</i> (sipunculid)	-30.3	7.8	1.5
<i>Alvinocaris stactophila</i> (shrimp)	-24.1	7.6	
<i>Munidopsis</i> Type 2 (squat lobster)	-27.4	9.6	6.1
<i>Escarpia</i> -like species (tube worm)	-27.5	5.9	
<i>Lamellibranchia</i> sp. (tube worm)	-25.3	4.6	
<i>Alvinocaris stactophila</i> (shrimp)	-28.9	10.1	
Polychaete #2 (worm)	-29.8	10.6	
<i>Bathymodiolus childressi</i> (mussel)	-36.9	5.4	
<i>Bathymodiolus childressi</i> (mussel)	-45.5	2.5	6.7
<i>Bathymodiolus childressi</i> (mussel)	-44.0	1.8	
<i>Bathymodiolus childressi</i> (mussel)	-43.1	3.0	
Mean $\pm$ SD	$-32.1 \pm 8.3$	$6.1 \pm 3.2$	$4.8 \pm 2.8$
<b>GC 233</b>			
<i>Bathynertia naticoidea</i> (snail)	-51.6	-6.5	6.6
<i>Bathynertia naticoidea</i> (snail)	-52.4	-5.6	11.2
<i>Bathymodiolus childressi</i> (mussel)	-69.0	-20.9	11.6
<i>Bathymodiolus childressi</i> (mussel)	-65.0	-17.9	14.6
<i>Bathymodiolus childressi</i> (mussel)	-61.8	-16.8	
Orbinid sp. (worm)	-60.0	-9.3	15.9
<i>Alvinocaris stactophila</i> (shrimp)	-36.5	-4.4	
<i>Cycloporus</i> sp. (flat worm)	-61.3	-16.4	15.9
<i>Cycloporus</i> sp. (flat worm)	-58.9	-14.6	17.2
Mean $\pm$ SD	$-57.3 \pm 9.6$	$-12.5 \pm 6.1$	$13.3 \pm 3.7$

used in mixing equations for both GC 234 and GC 185 (see following subsections).

Two species of vagrant predators were captured in the small on-site trap at GC 234, *Bathynomus giganteus* (2 specimens), and *Eptatretus* sp. (11 specimens), along with a single specimen of the predatory gastro-

pod *Buccinum canetae* (Fig. 2). Three species of fishes were directly captured (as opposed to trap captured) as part of tube worm aggregation collections at the GC 234 site: the eel, *Synaphobranchus*, the brotulid *Oligopus* sp., and a viperfish *Chauliodis sloani*.

The on-site *Bathynomus giganteus* and *Eptatretus* yielded values reflective of heavy reliance on photosynthetically-derived foods rather than the surrounding chemoautotrophic production, but showed species-specific differences. The 2 specimens of *B. giganteus* were significantly more enriched in  $^{13}\text{C}$  and  $^{15}\text{N}$  ( $p = 0.05$  and  $0.03$  respectively) than the *Eptatretus* and were isotopically similar to 'typical' ocean fauna (Table 2). The relatively large sample size for *Eptatretus* did, however, reveal a pattern for  $\delta^{13}\text{C}$  and  $\delta^{34}\text{S}$  consistent with limited consumption of chemoautotrophic tissue. The ranges of  $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$  and  $\delta^{34}\text{S}$  values for captured *Eptatretus* were  $-18.3$  to  $-16.3$ ‰,  $7.3$  to  $12.2$ ‰ and  $12.1$  to  $18.5$ ‰ respectively with a trend toward depletion in  $^{13}\text{C}$  and  $^{34}\text{S}$  (Fig. 2). Several of the *Eptatretus* had  $\delta^{34}\text{S}$  values that were quite depleted relative to ocean sulfate, and a simple regression of  $\delta^{13}\text{C}$  with  $\delta^{34}\text{S}$  was significant ( $p = 0.02$ ).

Supportive evidence that the *Eptatretus* population consumes some seep production was found in the calculation of the mixing equation based on  $\delta^{34}\text{S}$ . It is estimated that the on-site *Eptatretus* population caught at GC 234 derived, on average, between 7 and 12% of their dietary sulfur from chemosynthetic sources (Table 2). The standard deviations associated with the mixing equations were, however, large and equal to the mean. The median percent of dietary sulfur that originated from chemosynthetic sources was between 3 and 6%, depending on the  $\delta^{34}\text{S}_{\text{seep}}$  value used.

The seep-resident gastropod and fishes showed markedly different trophic dependency from the trapped vagrants. The gastropod *Buccinum canetae*, and the fishes *Synaphobranchus* and *Oligopus* sp. displayed  $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$  and  $\delta^{34}\text{S}$  values consistent with a diet of predominately chemoautotrophic tissue. Solution of the mixing equation for  $\delta^{13}\text{C}$  showed that all 3 species consumed between 50 and 100% seep-produced carbon. The tissue  $\delta^{15}\text{N}$  values of the fishes placed them near the top of the seep-colonist food chain (Table 2). The tissue stable isotope values of the viperfish *Chauliodis sloani* indicated moderate levels of consumption of seep fauna (10 to 24%).

Three species of vagrant predators were caught off-site at Z-traps approximately 2 km from a known seep community at GC 234 ( $27^\circ 45.9' \text{N}$ ,  $91^\circ 11.6' \text{W}$ ). In addition to *Rochina crassa* (3 specimens) and *Bathynomus giganteus* (5 specimens), 1 hake *Urophycis* sp. was captured (Fig. 3). All individuals had isotope values similar to those seen in other pelagic Gulf of

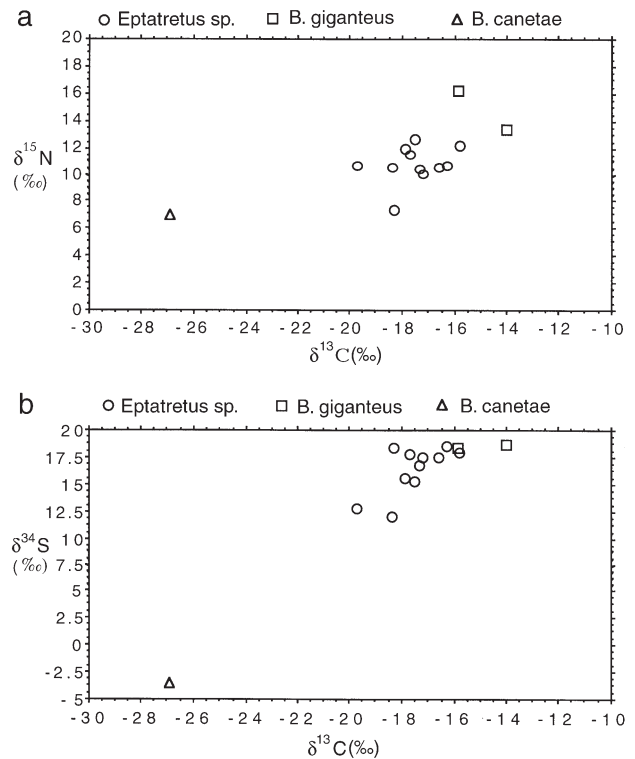


Fig. 2. (a)  $\delta^{13}\text{C}$  vs  $\delta^{15}\text{N}$  and (b)  $\delta^{13}\text{C}$  vs  $\delta^{34}\text{S}$  for animals collected from small traps deployed at GC 234. *Eptatretus* sp. is a hagfish, *Bathynomus giganteus* a giant isopod, and *Buccinum canetae* a predatory snail

Mexico predators. *B. giganteus* on-site were more enriched in  $^{13}\text{C}$  and  $^{15}\text{N}$  than those captured off-site, although not significantly so ( $p = 0.08$ ). Off-site, there was no statistical difference between the  $\delta^{34}\text{S}$  of *R. crassa* and *B. giganteus*. As a group, the predators collected off-site in the Z-trap were significantly more enriched in  $\delta^{34}\text{S}$  than those caught in the small on-site trap ( $p = 0.0002$ ). It is unlikely that these off-site vagrants consumed any appreciable amount of chemosynthetically derived material.

### Site GC 185

A site-specific value for the isotopic signature of the seep food sources was developed from a selection of 2 symbiont-hosting and 8 resident heterotrophs (Table 1). The values obtained were  $-31.2 \pm 6.0$ ‰  $\delta^{13}\text{C}_{\text{seep}}$  ( $n = 18$ ),  $6.9 \pm 1.2$ ‰  $\delta^{15}\text{N}$  ( $n = 18$ ), and  $-7.3 \pm 8.4$ ‰  $\delta^{34}\text{S}$  ( $n = 18$ ). As noted in the previous section, the mean chemosynthetic  $\delta^{13}\text{C}$  value of prey items at GC 185 was statistically indistinguishable from that at GC 234 and therefore an intermediate value of  $\delta^{13}\text{C}_{\text{seep}}$  of  $-32 \pm 6.0$ ‰ was used for both sites.

Table 2. Means  $\pm$  SP and (N) isotope values for animals captured on- and off-site at GC 234, GC 233 and GC 185. % of chemosynthetic material: estimate of biomass carbon or sulfur derived from chemosynthetic production (carbon isotopes were used to calculate this percentage unless noted otherwise)

Species	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{34}\text{S}$	% chemosynthetic material
<b>GC234, on-site</b>				
Vertebrates				
<i>Eptatretus</i> sp.	$-17.5 \pm 1.1$ (11)	$10.8 \pm 1.4$ (11)	$16.2 \pm 2.2$ (12)	7–12
<i>Oligopus</i> sp. <sup>a</sup>	$-27.3 \pm 0.3$ (2)	$9.9 \pm 0.3$ (2)		53–100
<i>Synaphobranchus</i> sp. <sup>a</sup>	–24.9	8.5		42–100
<i>Chauliodis slonani</i> <sup>a</sup>	–18.2	10.7		10–24
Invertebrates				
<i>Bathynomous giganteus</i>	$-14.9 \pm 1.3$ (2)	$14.8 \pm 2.0$ (2)	$18.5 \pm 3.0$ (2)	0
<i>Buccinum canetae</i>	–26.9 (1)	7.0 (1)	–3.5 (1)	50–100
<b>GC234, off-site Z-traps</b>				
Vertebrates				
<i>Urophycis</i> sp.	–17.5 (1)	12.3 (1)	20.0 (1)	0
Invertebrates				
<i>Bathynomous giganteus</i>	$-15.7 \pm 0.3$ (2)	$14.5 \pm 0.2$ (2)	$20.5 \pm 0.7$ (5)	0
<i>Rochina crassa</i>	$-17.9 \pm 0.8$ (3)	$10.5 \pm 0.5$ (3)	$20.4 \pm 0.2$ (2)	0
<b>GC233, on-site</b>				
Vertebrates				
<i>Eptatretus</i> sp. <sup>a</sup>	–28.6	8.2		23–38
<i>Eptatretus</i> sp.	$-19.8 \pm 1.4$ (4)	$10.3 \pm 1.2$ (4)	18.4 (1)	6–9
<i>Synaphobranchus</i> sp.	–42.5 (1)	–3.0 (1)		51–85
<i>Nezumia</i> sp.	–32.7 (1)	2.2 (1)		31–52
Myctophid	$-18.5 \pm 0.3$ (3)	$7.8 \pm 1.5$ (3)		0
Small shark	$-17.8 \pm 0.5$ (2)	$12.7 \pm 0.2$ (2)	$17.8 \pm 0.3$ (2)	0
Invertebrates				
<i>Bathynomous giganteus</i>	$-19.2 \pm 5.8$ (6)	$12.2 \pm 5.9$ (6)	$18.0 \pm 0.9$ (3)	0–45
<i>Rochina crassa</i>	$-23.5 \pm 3.7$ (2)	$7.5 \pm 3.3$ (2)	$13.2 \pm 0.6$ (2)	18–30
<i>Buccinum canetae</i>	$-52.6 \pm 1.7$ (3)	$-10.6 \pm 0.9$ (3)	7.4 (1)	71–100
<b>GC233, off-site Z-traps</b>				
Invertebrates				
<i>Bathynomous giganteus</i>	$-16.4 \pm 1.2$ (11)	$14.1 \pm 1.7$ (11)	$20.1 \pm 1.0$ (9)	0
<i>Rochina crassa</i>	–20.9 (1)	9.8 (1)	12.8 (1)	48 (S)
<b>GC185, on-site</b>				
Vertebrates				
<i>Eptatretus</i> sp.	–20.9 (1)	11.5 (1)	12.2 (1)	23–55
<i>Ophichthus cruentifer</i>	–33.4 (1)	7.1	–13.3 (1)	82–100
<i>Dysomma rugosa</i>	–30.4 (1)	7.4 (1)		68–100
Invertebrates				
<i>Bathynomous giganteus</i>	$-17.2 \pm 1.7$ (2)	$15.2 \pm 0.8$ (2)	$17.7 \pm 0.9$ (2)	5–16 (S)
Unidentified crab	–18.4 (1)	9.3 (1)	19.9 (1)	0
Atelecyliid crab	–24.8 (1)	9.7 (1)		42–97
<i>Buccinum canetae</i>	$-29.1 \pm 9.0$ (2)	$6.2 \pm 1.6$ (2)	$-4.5 \pm 6.3$ (4)	32–100 (C)
<i>Sclerasterias tanneri</i>	$-33.8 \pm 1.6$ (3)	$5.5 \pm 0.1$ (3)	3.9 (1)	45–100 (S)
				80–100
<b>GC185, off-site Z-traps</b>				
Vertebrates				
<i>Eptatretus</i> sp.	$-19.8 \pm 2.1$ (21)	$10.6 \pm 1.5$ (21)	$18.7 \pm 2.7$ (21)	16–40
Invertebrates				
<i>Bathynomous giganteus</i>	$-15.7 \pm 0.1$ (8)	$14.0 \pm 1.1$ (8)	$18.2 \pm 1.6$ (10)	6–11 (S)
<i>Rochina crassa</i>	$-18.6 \pm 1.7$ (12)	$10.4 \pm 1.0$ (13)	$15.6 \pm 1.7$ (9)	8–17
<i>Chaceon</i> sp.	–15.0 (1)	9.3 (1)	19.7 (1)	0

<sup>a</sup>Direct capture as opposed to trap capture

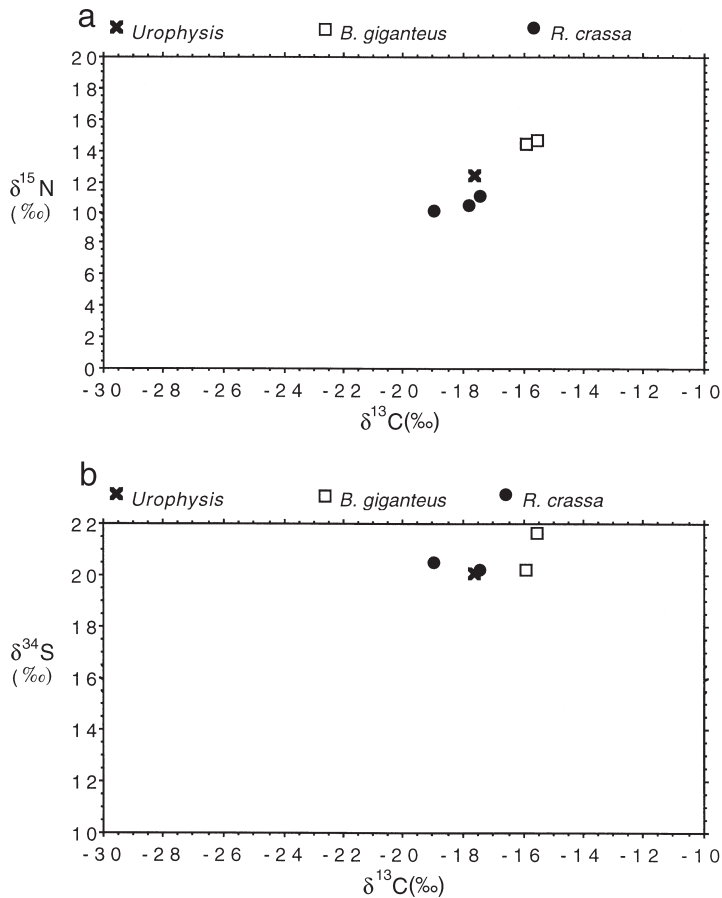


Fig. 3. (a)  $\delta^{13}\text{C}$  vs  $\delta^{15}\text{N}$  and (b)  $\delta^{13}\text{C}$  vs  $\delta^{34}\text{S}$  for predators collected in Z-traps off GC 234. *Urophysis* sp. is a hake, *Bathynomus giganteus* a giant isopod, and *Rochina crassa* a spider crab

On-site traps at GC 185 caught *Bathynomus giganteus* (2 specimens) and *Eptatretus* sp. (1 specimen), while sampling from the submersible produced the predatory sea star *Sclerasterias* cf. *tanneri* (3 specimens), gastropod *Buccinum canetae* (2 specimens), and one specimen each of the eels *Ophichthus cruentifer* and *Dysomma rugosa*, an unidentified crab, and an Atelecyliid crab (Fig. 4).

*Bathynomus giganteus* specimens were only slightly depleted in  $^{13}\text{C}$  and  $^{34}\text{S}$  relative to background fauna, but relative to those caught off-site (Table 2) there was some indication of a small chemosynthetic contribution. Mixing equations for  $\delta^{34}\text{S}$  indicate that between 5 and 16% of biomass sulfur was derived from chemosynthetic sources. The trophic enrichment of *B. giganteus* prevented any meaningful analysis of  $\delta^{13}\text{C}$  in mixing equations. The single *Eptatretus* showed a greater degree of chemosynthetic carbon incorporation (23 to 55%: Table 2).

Among the other on-site predators captured, most had depleted  $\delta^{13}\text{C}$  values, showing clear dependence upon chemosynthetic material. Solution of the mixing equation for  $\delta^{13}\text{C}$  showed the eels, gastropod, sea stars, and atelecyliid crab to be up to 100% reliant on seep-community carbon. In the case of *Buccinum canetae*,  $\delta^{34}\text{S}$  values (ranging from  $-9.6$  to  $3.8$ ‰,  $n = 4$ ) produced a more focused view of consumption. Based on a mixing equation for  $\delta^{34}\text{S}$ , a minimum of 80% of the *B. canetae* biomass sulfur was obtained from chemosynthetic production. The most  $^{34}\text{S}$ -enriched of the *B. canetae*, however, was also the most  $^{13}\text{C}$ -depleted, indicating methanotrophic contributions over thiotrophic sources.

The off-site trapping produced numerous specimens of *Eptatretus* (21 specimens), *Rochina crassa* (12 specimens), and *Bathynomus giganteus* (8 specimens) (Fig. 5). The relatively large sample sizes for the 3 species allowed a more thorough analysis of the data. For *Eptatretus* the mean off-site GC 185  $\delta^{13}\text{C}$  was equal to that on-site at GC 233 (see following subsection), consistent with some incorporation of chemosynthetic organic matter (Table 2). In most cases, the  $\delta^{34}\text{S}$  values of off-site *Eptatretus* were much too enriched for thiotrophic material to be an important part of its diet (Table 2). There were 3 exceptions, where the hagfish had  $\delta^{13}\text{C}$  values of  $\leq 20$ ‰ and depleted  $\delta^{34}\text{S}$  values (11.4, 12.4 and 15.4‰) relative to ocean sulfate. From

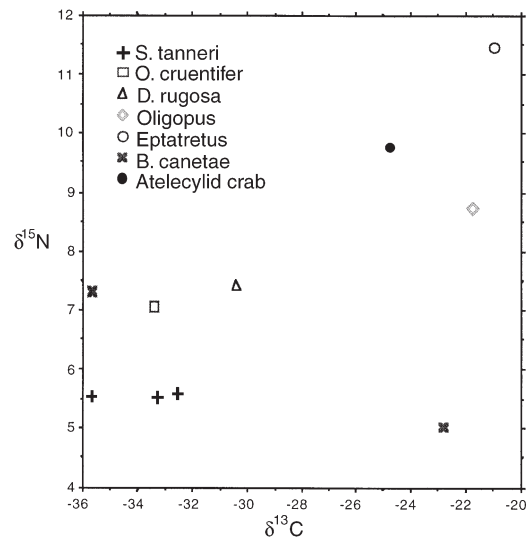


Fig. 4.  $\delta^{13}\text{C}$  vs  $\delta^{15}\text{N}$  for predators collected on-site at GC 185. Note that, although the  $^{13}\text{C}$ -enriched *Buccinum canetae* was close to values expected for photosynthetic production, its  $\delta^{34}\text{S}$  value was very depleted ( $-9.4$  ppt) relative to ocean sulfate (19 to 20‰). *Sclerasterias tanneri* is a sea star, *Ophichthus cruentifer* and *Dysomma rugosa* are eels, *Oligopus* sp. a brotulid fish, *Eptatretus* sp. a hagfish and *Buccinum canetae* sp. a predatory snail



the mixing equation based on  $\delta^{34}\text{S}$  it was estimated that these 3 hagfish derived between 21 to 41, 18 to 35, and 9 to 18% of their biomass sulfur from tube worms or thiotrophically-derived material similar to tube worms. As mentioned above, the remaining *Eptatretus* were enriched in  $^{34}\text{S}$ , generally near the 20‰ value of ocean sulfate. Their  $\delta^{13}\text{C}$  values were slightly depleted relative to typical ocean predators, suggesting limited incorporation of chemosynthetic material that was more probably of methanotrophic than thiotrophic origin.

*Rochina crassa* captured off-site were also depleted in  $^{13}\text{C}$  and  $^{34}\text{S}$  relative to background ocean isotopic values. Their  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values overlapped the range observed for the *Eptatretus* (Fig. 5), but their  $\delta^{34}\text{S}$  was significantly depleted relative to *Eptatretus* ( $p = 0.002$ ). The relatively low  $\delta^{34}\text{S}$  combined with  $\delta^{13}\text{C}$  values, seldom below  $-22\text{‰}$ , suggests that tube worm material or organisms that feed on tube worms contribute to the diet of the *R. crassa*. The fact that the *R. crassa* are approximately 8‰ higher in  $\delta^{15}\text{N}$  than tube worms, strongly suggests that they did not directly graze upon tube worms but consumed tube worm predators. The mixing equation based on  $\delta^{34}\text{S}$  estimated that the mean percentage of *R. crassa* biomass sulfur derived from thiotrophic sources was 8 to 17% (Table 2).

*Bathynomus giganteus* captured off-site were among the most  $^{13}\text{C}$ -enriched of any animal captured in this study (mean:  $16.0 \pm 1.1\text{‰}$ ), however their  $\delta^{34}\text{S}$  values were slightly depleted (17.9‰) relative to values expected for Gulf of Mexico predators. Using the same end-member  $\delta^{34}\text{S}$  values as with the *Rochina crassa* above, it is estimated that between 6 and 11% of the *B. giganteus* biomass sulfur was derived from chemosynthetic production.

#### GC 233

Site-specific values for the isotopic signature of the seep food sources was developed from a selection of 1 symbiont-hosting and 3 resident heterotrophic species (Table 1). The values obtained were:  $57.3 \pm 9.6\text{‰}$   $\delta^{13}\text{C}_{\text{seep}}$  ( $n = 9$ ),  $-12.5 \pm 6.1\text{‰}$   $\delta^{15}\text{N}$  ( $n = 9$ ), and  $13.3 \pm 3.7\text{‰}$   $\delta^{34}\text{S}$  ( $n = 7$ ). These were dramatically different from the values shared between GC 185 and GC 233 for 2 reasons. First, the dominant symbiont-containing population is a monoculture of the methanotrophic mussel *Bathymodiolus childressi*. Second, there are very distinct site-specific isotopic signatures for the methane substrate, which is more biogenic (Brooks et al. 1988, Kennicutt et al. 1992, Sassen et al. 1999), with a depleted  $\delta^{13}\text{C}$  signature ( $\delta^{13}\text{C} \leq -60\text{‰}$ ). The negative and distinctive  $\delta^{15}\text{N}$

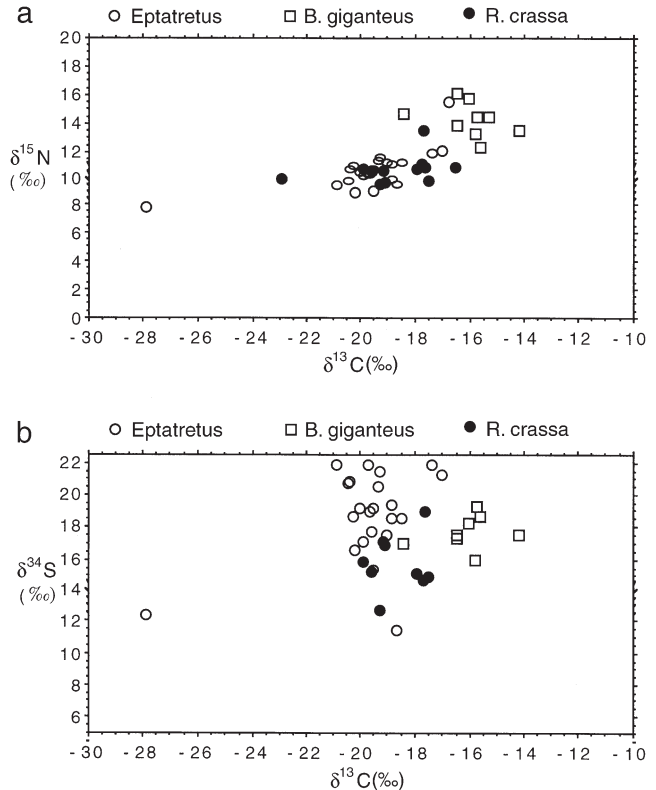


Fig. 5. (a)  $\delta^{13}\text{C}$  vs  $\delta^{15}\text{N}$  and (b)  $\delta^{13}\text{C}$  vs  $\delta^{34}\text{S}$  for predators caught in Z-traps off GC 185. *Eptatretus* sp. is a hagfish, *Bathynomus giganteus* a giant isopod, and *Rochina crassa* a spider crab

values (Table 2) are probably due to high concentrations of ammonium. Because the greatly depleted nitrogen signal is clearly different from that associated with Gulf of Mexico photosynthesis products (Fry 1983, Macko et al. 1984),  $\delta^{15}\text{N}$  was used in the solution of the mixing equation.

On-site trapping at GC 233 caught the three most common vagrants, *Bathynomus giganteus* (6 specimens), *Eptatretus* sp. (4 specimens), and *Rochina crassa* (3 specimens), along with the rattail fish *Nezumia* sp. (1 specimen) and gastropod, *Buccinum canetae* (3 specimens). Direct collection by the submersible produced the eel *Synaphobranchus* sp. and a small demersal shark, a pelagic myctophid fish, and an additional specimen of *Eptatretus* (Fig. 6).

The on-site *Eptatretus* and *Bathynomus giganteus* values indicated a general reliance on photosynthetic food, but gave indication of a minor trend in depleted  $\delta^{13}\text{C}$  values. Among the *B. giganteus*, there was a 15‰ range in  $\delta^{13}\text{C}$  ( $-30.6$  to  $-15.4\text{‰}$ , Fig. 6a). Of the 6 specimens captured, 2 had slightly depleted  $\delta^{13}\text{C}$  values and 1 had incorporated substantial chemosynthetic

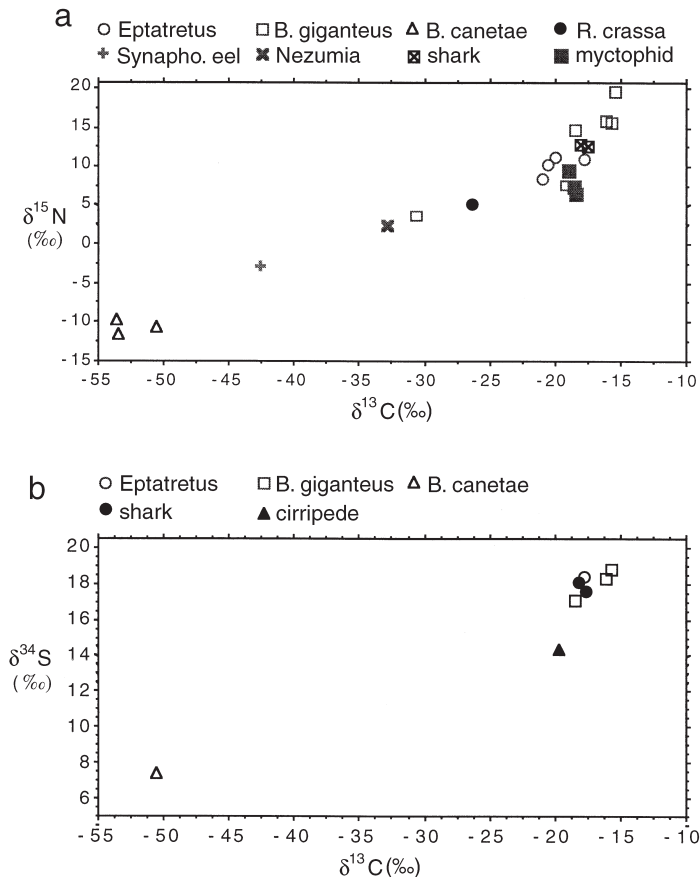


Fig. 6. (a)  $\delta^{13}\text{C}$  vs  $\delta^{15}\text{N}$  and (b)  $\delta^{13}\text{C}$  vs  $\delta^{34}\text{S}$  for predators collected from small traps deployed at GC 233. *Eptatretus* sp. is a hagfish, *Bathynomus giganteus* a giant isopod, *Rochina crassa* a spider crab, *Buccinum canetae* a predatory snail, *Synaphobranchus* sp. an eel, *Nezumia* sp a rattail fish, and *Cirripedia* a barnacle taken from the carapace of *R. crassa*

carbon. The most depleted *B. giganteus* may have incorporated between 27 and 45% of its carbon from methanotrophic sources, the other 2 moderately depleted *B. giganteus* incorporated only 3 to 7% (Table 2). The mean  $\delta^{13}\text{C}$  value of *Eptatretus* from GC 233 was only about 1‰ depleted relative to ocean phytoplankton, but significantly depleted relative to the values of those captured at GC 234 ( $p = 0.02$ ), indicating that the former probably had incorporated a small amount of chemosynthetic carbon. Based on the mixing equation for  $\delta^{13}\text{C}$ , between 6 and 9% of the *Eptatretus* biomass carbon was derived from methanotrophically fixed carbon. The  $\delta^{15}\text{N}$  of the on-site *Eptatretus* and *B. giganteus* were extremely enriched relative to the 'typical' prey  $\delta^{15}\text{N}$  from GC 233 (Tables 1 & 2), however, on-site *B. giganteus* were depleted in  $^{15}\text{N}$  relative to those captured off-site (see below), indicating small chemosynthetic nitrogen components in their diet. The 3 *B. giganteus* analyzed for  $\delta^{34}\text{S}$  were

the individuals with  $\delta^{13}\text{C}$  values similar to open ocean predators. Their enriched  $\delta^{34}\text{S}$  values also indicate their reliance on photosynthetically derived material, although they were not as enriched as *B. giganteus* captured off-site at GC 233 (see below, Figs 6b & 7b).

Of the other species captured at GC 233, *Buccinum canetae*, *Synaphobranchus*, *Nezumia*, and 1 of 3 *Rochina crassa* showed greater dependence on chemoautotrophic production. Solving the mixing equation (Eq. 2) for  $\delta^{13}\text{C}$  produced results of: *R. crassa*, 13 to 23%; *Nezumia* sp., 27 to 47%; *B. canetae*, 69 to 100%; *Synaphobranchus* sp., 47 to 83% (Table 2). Solving the mixing equation for  $\delta^{15}\text{N}$  revealed similar ranges: *R. crassa*, 17 to 30%; *Nezumia* sp., 26 to 45%; *B. canetae*, 72 to 100%; *Synaphobranchus* sp., 46 to 79%.

The off-site trap, deployed approximately 2 km from the GC 233 (27°43.86'N, 91°16.48'W), caught 11 *Bathynomus giganteus* and 1 *Rochina crassa* (Fig. 7). The *R. crassa* was distinct, being depleted in  $^{13}\text{C}$  relative to typical ocean predators and the same species off-site at GC 234 (Table 2), although not as depleted as the *R. crassa* on-site at GC 233. These values, as well as depleted  $\delta^{34}\text{S}$ , were indicative of a consumer utilizing chemosynthetically fixed sulfur. Based on the  $\delta^{34}\text{S}$  values, the *R. crassa* had derived approximately 48% of its sulfur from methanotrophic material (Table 2). There was no statistical difference in  $\delta^{13}\text{C}$  or  $\delta^{15}\text{N}$  between the *B. giganteus* caught on- and off-site at GC 233, however, those caught off-site were significantly more enriched in  $^{34}\text{S}$  ( $p = 0.03$ ) relative to those on-site, indicating less reliance on seep food sources.

## DISCUSSION AND CONCLUSIONS

Two main conclusions can be drawn from the results reported. First, on a species-specific basis some mobile benthic predators from the background fauna do obtain close to 100% of their nutrition from seep production, indicating they are residents of the seeps. Vagrant species utilized less than colonists on an individual-by-individual basis. Second, the extent of utilization found in on-site versus off-site trapped vagrant species becomes indistinct as sample size increases, indicating a high degree of movement in and out of the seep habitat for those species.

The mixing equations can be used to estimate that, at these locations, the sea star *Sclerasterias* cf. *tanneri*, the gastropod *Buccinum canetae*, the atelecyliid crab, and various fishes (*Ophichthus cruentifer*, *Dysomma rugosa*, and *Oligopus* sp.) generally obtain from 50 to 100% of their nutrition from seep production. Given the wide isotopic range of seep food sources, these species are probably completely dependent upon the

seep community for nutrition. The 3 invertebrates have been previously classed as colonists on the basis of high in-seep populations and comparative rarity in the wider background distribution range (Carney 1994). The in-seep versus background abundance of the fishes has yet to be determined, but they seem comparatively rare in the deep fish fauna of the background and may prove to fit within the colonists classification. As such, all these predators represent seep utilization by the background fauna in the sense of becoming part of the seep-site food web during at least part of their life cycle.

The commonly trapped vagrant species are direct exporters of seep production into the background. There was a species-ranking in the degree of utilization consistent with our knowledge of these organisms, *Eptatretus* sp. consuming the most, followed by *Rochina crassa*, and *Bathynomus giganteus* consuming the least. All 3 are opportunists but vary in the versatility of their feeding mechanisms such that their ability to exploit seep production may differ. *Eptatretus* spp. have not been well studied in the Gulf of Mexico, but the relevant ecology of this hagfish genus has been summarized by Martini (1998). Hagfish of that and other genera are opportunistic predators and scavengers capable of consuming large and small prey, and are widely distributed throughout all except the polar ocean regions, with a known depth range from inshore to 5000 m. As burrowing organisms, their abundance usually goes unnoticed, but they have proven to be one of the most abundant species of demersal fishes at upper slope depths. *Eptatretus* spp. can be expected to have a wide range of seep prey available to it. *R. crassa* is less well studied, but is a true crab feeding with elongated chelipeds that carry soft tissues to the mouth for ingestion (Williams 1984). Its relatively small chelae are probably not specialized for shell-crushing, limiting seep prey to small thin-shelled juvenile mussels, gastropods and crustaceans or soft-bodied animals. On occasion, specimens were captured which had been feeding upon hydroids, indicating that they can be omnivorous browsers on the limited seep epizoa. *B. giganteus* appears to have the least adaptable feeding methods and least access to seep prey. Typical of isopods, it lacks specialized food-gathering appendages other than mouthparts. It depends upon small food-handing maxillipeds and powerful, plier-like jaws. The jaws are faced with numerous short spines, making them ideally suited for tearing flesh, but are ill-suited for manipulating and crushing shells. In effect, it may have little access to seep biomass and may also be at a competitive disadvantage with the other 2 vagrant species.

Between colonist and vagrant lies a smaller third group of consumers represented in this study only by 1 *Synaphobranchus* sp. and the rattail fish *Nezumia* sp. Both are abundant in the background as with the trapped vagrants, but have seep utilization proportions similar to the background colonists. Both have been directly observed preying on seep crustaceans. In the case of *Nezumia* there is no obvious indication of a large in-seep population. *Synaphobranchus* however, burrows, making population assessment difficult. The possibility exists that these species may have restricted foraging areas and are effectively long-term seep residents.

There was an interesting relationship between sample size and the degree of seep utilization that reinforces the role of residency duration. In off-site samples, increased sample size resulted in increased indication of seep feeding. The smallest off-site sample ( $n = 9$  at GC 234) indicated little incorporation of seep food. Roughly the same sample size at GC 233 ( $n = 10$ ) produced a single specimen of *Rochina crassa* with a marked seep sulfur signal (Fig. 7). The largest sample

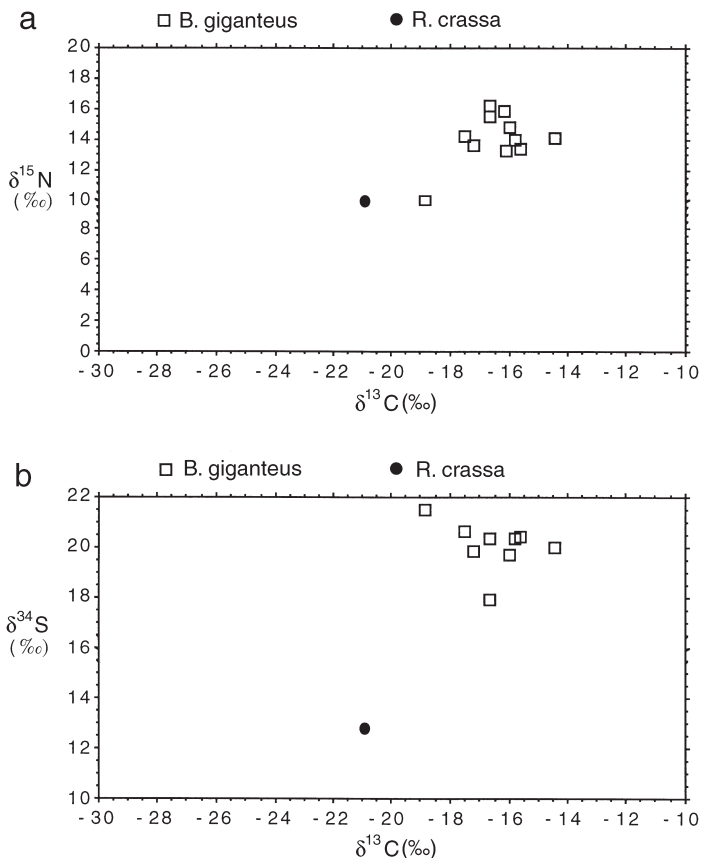


Fig. 7. (a)  $\delta^{13}\text{C}$  vs  $\delta^{15}\text{N}$  and (b)  $\delta^{13}\text{C}$  vs  $\delta^{34}\text{S}$  for predators collected in Z-traps off GC 233. *Bathynomus giganteus* is a giant isopod and *Rochina crassa* a spider crab

at GC 185 ( $n = 43$ ) yielded wide ranges of seep utilization by populations of *R. crassa* and *Eptatretus*. A trend opposite to that observed off-site is apparent on-site, especially for *Eptatretus* with increasing sample size, seep utilization became less apparent. Both trends are to be expected if there is considerable movement of individuals between the local areas being sampled by traps. The larger the sample size, the more often a newly arrived animal carrying the trophic label of its previous foraging region will be trapped. At least on the scales employed in this study, vagrants do export seep production.

The continental slope fauna of the Northern Gulf of Mexico have been sufficiently well studied to support the conclusion that background species that have taken up residency, colonists, are rare in the background and constitute a small fraction of the local predatory and scavenging fauna. Similarly, while the known vagrants are relatively common in the background, they comprise a small percent of the total species richness. The overall impression is that a limited number of background species exploit the seep as either long-term or short-term residents. There are 2 main possibilities for this absence of a broader species base of exploitation: (1) a limited availability of seep prey due to biotic defense and predator exclusion from a noxious environment, or (2) an adequate supply of food in the background.

With respect to prey defense, as a result of the sulfide-binding properties of their blood, the tissue of the tube worms normally contains sufficiently high concentrations of  $H_2S$  to likely render them inedible to most animals (Arp & Childress 1983), while the shells of the mollusks afford some degree of protection. Tunnicliffe (1992), proposed that noxious exclusion at hydrothermal systems, hydrogen sulfide and brines may produce similar effects at some hydrocarbon seep sites. The fact that so few background species utilize the seep habitat may simply reflect physiological tolerance. Unlike most hydrothermal sites, a diverse suite of background species are often present at seep sites (C. R. Fisher pers. obs.), suggesting that noxious exclusion may not be as extreme as at active vents. Both possibilities can be explored through field experimentation.

Our original expectation that many deep-sea background species would prey upon seep organisms was based upon the generally-held view that food is especially scarce in the deep sea. This may be true in benthic systems at more than 3000 m under oligotrophic waters, but its application to the upper continental slope must be questioned. The hydrocarbon seeps in this study are in fairly shallow water (500 to 650 m). Both the empirical Betzer-Suess relationship (Betzer et al. 1983) for particle flux and the Rowe (1983) relation-

ship for benthic biomass indicate that the influx of photo synthetic detritus at these depths is more comparable to that of the continental shelf rather than the abyssal. Reinforcing the possibility of relatively high background food levels is the fact that the upper slope of the Gulf of Mexico supports high populations of large predatory fishes and crabs. This is a common pattern on all upper slopes that is unrelated to the presence or absence to chemosynthetic communities (Gage & Tyler 1996 and references therein). Ultimately, the role of background food limitation in driving exploitation of seep and vent systems will require comparative studies of trophic patterns among endemic, colonist, and vagrant species in regions of contrasting labile detritus influx. The lower the food resources of the background, the greater the expected level of exploitation should be.

While the results of this study presently lack the standing stock and transfer rates needed to fully quantify trophic export from seeps, it is informative to consider the possibilities. The initial isotope survey of background fauna by Kennicutt & Brooks (unpubl.) indicated that up to 17% of the overall benthic fauna in the seep region might have consumed minor amounts of food from seeps. The inability of a single isotope to separate seep carbon from isotopically similar sedimentary organic carbon probably makes this estimate too high, since depleted  $\delta^{13}C$  values were only attributed to seep production. If, however, abundant forms such as the vagrants trapped in the present study have only 1 to 2% seep carbon in their overall population, then there must be a large export which must exert considerable influence upon the prey populations of the seep community. As to the populations of abundant background consumers, seep food may be so greatly exceeded by phytodetritus-based food at this depth that the overall impact of seep exploitation is minimal. Quite the contrary would be the case for the rarer background species which have colonized the seeps. The well-fed sub-populations resident within the seeps may make a higher reproductive contribution to the overall population than the sparse background sub-population. In such cases, overall regional persistence of the species may ultimately be dependent upon seep utilization.

Most large mobile predators captured on-site from the 3 chemosynthetic communities showed significant incorporation of chemosynthetic material. This was particularly apparent among predatory snails (*Buccinum canetae*) and eel-like forms (including *Oligopus* sp., *Synaphobranchus*, *Ophichthus cruentifer*, *Eptatretus*, *Nezumia* and *Dysomma rugosa*). *Bathynomus giganteus* captured within the seep communities generally showed the least chemosynthetic incorporation, although some individuals incorporated a significant

amount. Off-site *Rochina crassa* and *Eptatretus* (from traps deployed in the vicinity of GC 185 and GC 233) displayed a significant chemosynthetic isotope signal. It is estimated that they may derive 20% or more of their biomass carbon and sulfur from chemosynthetic sources. *B. giganteus* captured off-site generally showed little evidence of chemosynthetic usage. These results indicate that chemosynthetic production is exported to Gulf of Mexico predators. Although most mobile benthic predators not physically associated with the chemosynthetic communities derive most of their nutritional needs from photosynthetic production, their incorporation of chemosynthetic production is nevertheless substantial.

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