

Extreme food webs: Foraging strategies and diets of scavenging amphipods from the ocean's deepest 5 kilometers

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Abstract

We explore hypotheses that alternate foraging strategies, diet, or nutrient partitioning could help explain the success of scavenging Lysianassoids (Amphipoda) in hadal oligotrophic trenches (depths of 6–11 km) by examining the nutritional strategies of four lysianassoid species (*Eurythenes gryllus*, *Scopelocheirus schellenbergi*, *Hirondellea dubia*, and *Uristes* sp. nov.) collected with baited traps (6.3–10.8 km) from the oligotrophic Tonga and Kermadec Trenches (southwest Pacific Ocean). Diets and foraging strategies were examined by use of (1) the nascent DNA-based analysis of hindgut contents, which provides a 'snapshot' of recently ingested organisms, and (2) natural abundance isotopic signatures, which reflect the source of nutrition and relative trophic position. The scavenging guild exhibits remarkable trophic plasticity, and each amphipod species employs alternate foraging modes, including detritivory or predation, to supplement necrophagy. The nutritional strategies of some species appear to shift with age, depth, and even between trenches. Thus, there is no single ubiquitous hadal food web; rather it is influenced by depth and overlying surface productivity. Isotopic data suggest that coexisting species partition the dietary items, providing evidence of competition among members of the scavenging guild. The extreme foraging flexibility of scavenging amphipods may ultimately contribute to their success in severely food-limited hadal ecosystems.

The hadal zone (6,000–11,000 m) constitutes the deepest 45% of the ocean's vertical range and is located almost exclusively in ocean trenches. This zone is renowned for its extreme hydrostatic pressure, which contributes to high endemism within the hadal zone by (1) excluding those deep-sea taxonomic groups seemingly incapable of adapting to such great pressures (e.g., decapods), and (2) ecologically isolating hadal fauna from the shallower adjacent abyssal waters (Wolff 1960; Belyaev 1989; Vinogradova 1997). Biological sampling at these depths is exceptionally expensive and difficult (Angel 1982; Perrone 2002). Consequently, there is little knowledge of fundamental ecological features such as trophic links and food web structure.

Large nekton carcasses are a sporadic, yet significant source of nutrient input to deep-sea benthic communities (Stockton and DeLaca 1982). Many deep-sea species are

adapted to locate and exploit these nutrient hotspots (Smith and Baldwin 1982; Tamburri and Barry 1999; Smith and Baco 2003), and necrophagy as a nutritional strategy becomes more prevalent with depth (Stockton and DeLaca 1982; Britton and Morton 1994). Crustacean amphipods belonging to the superfamily Lysianassoidea often dominate scavenging assemblages at great depths (Thurston 1990; Gage and Tyler 1996; De Broyer et al. 2004), and lysianassoids appear to be the exclusive taxon assuming the role of scavenger in the deep hadal zone (Hessler et al. 1978; Perrone et al. 2002). Hence, scavenging amphipods are key members in hadal food webs (Kamenskaya 1995).

In 2001, an unprecedented sampling effort of two of the world's deepest trenches yielded tens of thousands of necrophagous amphipods. The collection from the abyssal and hadal depths of the Tonga Trench and hadal depths of the Kermadec Trench (southwest Pacific Ocean) contained the following four lysianassoid species: *Eurythenes gryllus*, *Scopelocheirus schellenbergi*, *Hirondellea dubia*, and *Uristes* sp. nov. (Blankenship et al. 2006). The recovery of four species from the same superfamily with seemingly similar nutritional modes was unexpected. The hadal environment of both the Tonga and Kermadec Trenches is resource-poor. Because of the oligotrophic overlying waters, carrion input into these trenches is undoubtedly low, as evidenced by the extremely low biomass of organisms collected during a 17-h midwater trawl over the Tonga Trench (L. Blankenship, unpubl. data). Detrital flux should also be of extremely low quantity and quality (Sokolova 1994), and this is evidenced by the paucity of hadal meiofaunal diversity and biomass, even of Foraminifera (Belyaev 1989; Tundo 2003). Moreover, the steep slopes of the trench walls restrict the spatial extent of the habitat. Consequently, we expect that competition among scavengers for both food and space is likely to be severe (Iken et al. 2001). Some of

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Acknowledgments

We thank Art Yayanos for leading the expedition to the Tonga-Kermadec Trench, and the RV *Melville* Captain Dave Murline and crew, Roger Chastain, and Erica Goetze for help with sampling and collection. Peter Lonsdale and Christina Massel provided bathymetric charts of the Tonga Trench. Olga Kamenskaya provided information regarding hadal scavenging amphipod diets. We thank Ray Lee for determining the stable isotope values via mass spectrometry and Jen Gonzalez for assisting with the preparation of isotope samples. Ken Halanych directed us to appropriate universal 18S primers for metazoans. Valerie Allain provided unpublished isotope data for tuna collected in the South Pacific. Suggestions from two anonymous reviewers greatly improved this manuscript.

This research was supported by National Science Foundation grants OCE 99-07651 to A. Yayanos, IGERT 0333444 to Nancy Knowlton et al. (Scripps Institution of Oceanography), and IBN0076604 to R. Lee.

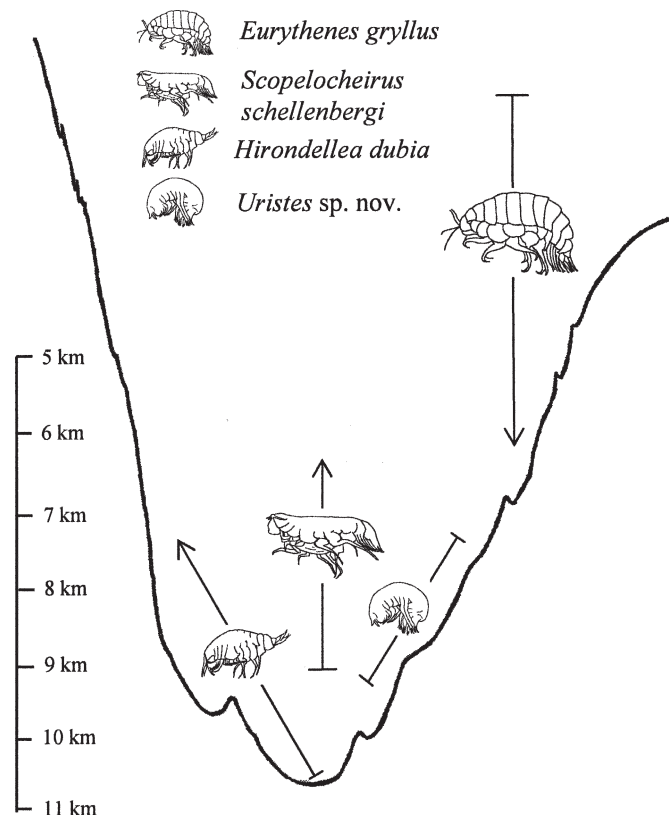


Fig. 1. Vertical habitat partitioning exhibited by the Tonga Trench scavenging amphipod assemblage (adapted from Blankenship et al. 2006). Bars represent the known vertical range of each species; vertical bars signify the species is benthopelagic whereas slanted bars denote a demersal species. Arrows indicate the depth zone containing most or all of the juveniles. The depiction of the trench cross section and amphipods are not drawn to scale.

this competition is probably relieved through habitat partitioning, which is manifested by species inhabiting distinct depth zones (Fig. 1; Blankenship et al. 2006). However, the vertical ranges of species are not mutually exclusive, and up to three lysianassoid species coexist at some hadal depths (Blankenship et al. 2006). Interspecific competition for carrion is probably still intense, and we hypothesized that each lysianassoid species may exercise alternate foraging strategies to further reduce competition.

Current understanding of deep-sea lysianassoid feeding ecology is based on studies detailing their anatomical designs, behavioral displays, and rapid convergence on and sequestering of carrion arriving on the deep-sea floor (Dahl 1979; Thurston 1990; Sainte-Marie 1992). The prevailing view is that deep-sea lysianassoids rely on necrophagy heavily, if not exclusively, for food (De Broyer et al. 2004). Yet, the importance of necrophagy is possibly overstressed, as conclusive diet analyses are rare. Traditional approaches used to study diet are not easily applied to deep-sea lysianassoids. First, the deep sea is notoriously difficult to observe. Second, the majority of deep-sea scavenging amphipods are collected with baited traps (e.g., Shulenberg and Hessler 1974; Christiansen et al. 1990; De Broyer et al. 2004); amphipods with access to bait in traps almost

always satiate themselves with it. Collection methods designed to prohibit deep-sea amphipods from consuming bait do exist, but they often collect amphipods with empty guts or guts containing amorphous material (Smith and Baldwin 1984; Sainte-Marie 1992). Consequently, specimens with native and identifiable gut contents are scarce, preventing dietary analyses based on examination of gut contents (Hessler et al. 1978; Sainte-Marie 1992).

Several alternative methods have been applied recently to study foraging behavior and diet of marine invertebrates. These approaches include (1) stable isotope ratios and fatty acid signatures as biomarkers of assimilated food (Hobson and Welch 1992; Graeve et al. 2001; Nyssen et al. 2002), and (2) the nascent approach of using universal polymerase chain reaction (PCR) primers to amplify and sequence the DNA from gut contents to identify ingested organisms (Blankenship and Yayanos 2005). Consumed organisms that are digested beyond visually identifiable remains can still leave residual DNA in the digestive tract (Duplessis et al. 2004; Jarman et al. 2004). Recently, it was shown that this dietary DNA can be amplified with highly conserved primers and sequenced (see Blankenship and Yayanos 2005). The resultant DNA sequences are analogous to species barcodes and referencing them against a DNA database such as GenBank should provide insight to their taxonomic origin (Hebert et al. 2003).

$\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ stable isotope ratios (the $^{15}\text{N} : ^{14}\text{N}$ or $^{13}\text{C} : ^{12}\text{C}$ ratio compared to standard reference ratios) are a useful complement to digestive tract analyses, as the isotopic signatures are a reflection of assimilated food (Gearing 1991). Nitrogen isotopes are fractionated during feeding ($\sim 2.3\text{‰}$ per trophic step; McCutchan et al. 2003) and therefore often are used to elucidate relative trophic position (Minagawa and Wada 1984; Hobson and Welch 1992). In contrast, carbon isotopes are more conserved between consumer and prey in the marine environment ($\sim 0.4\text{‰}$ enrichment per trophic level; McCutchan et al. 2003), but primary producers (e.g., phytoplankton, chemosynthetic bacteria, or intertidal plants) can exhibit distinct $\delta^{13}\text{C}$ signatures due to different carbon fixation pathways (Fry and Sherr 1984; Gearing 1991). Thus, $\delta^{13}\text{C}$ values can indicate the nutritional base of a food web (Fry and Sherr 1984) and have been especially valuable in detecting some forms of chemosynthesis-based nutrition (Levin and Michener 2002).

Combining multiple approaches to elucidate food webs should generate a more comprehensive depiction and is more desired than application of a single method (Gearing 1991; Graeve et al. 2001). Here, we use both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ stable isotope signatures and DNA-based analysis of gut contents to assess the feeding ecology of hadal scavenging amphipods and to gain insight into the food web structure in the Tonga and Kermadec Trenches. Specifically, we addressed the following questions. (1) Is nekton carrion the sole source of nutrition for the hadal scavenging guild? If not, what alternative dietary items and foraging modes supplement necrophagy? (2) Do the trophic roles differ among species, bottom depths, amphipod size classes, or between the two trenches? (3) Is there evidence of competition for food among coexisting amphipods?

Table 1. Amphipod species collected from the Tonga and Kermadec Trench hadal zones.

Free vehicle number	Bottom depth (m)	Location	Position of trap deployment	Trapped amphipod species
4	6,252	Tonga Trench	23°49.9'S 174°24.9'W	<i>Eurythenes gryllus</i> , <i>Scopelocheirus schellenbergi</i>
14	6,834	Tonga Trench	17°04.9'S 172°06.1'W	<i>S. schellenbergi</i>
6	8,582	Tonga Trench	24°10.9'S 175°02.5'W	<i>S. schellenbergi</i> , <i>Hirondellea dubia</i> , <i>Uristes</i> sp. nov.
12	8,723	Tonga Trench	17°20.1'S 172°16.3'W	<i>S. schellenbergi</i> , <i>H. dubia</i> , <i>Uristes</i> sp. nov.
9	9,104	Kermadec Trench	32°10.0'S 177°15.9'W	<i>H. dubia</i> , <i>Uristes</i> sp. nov.
10	10,782	Tonga Trench	23°13.7'S 174°43.6'W	<i>H. dubia</i>

Our results, combined with PCR-based gut content studies of *S. schellenbergi* and *E. gryllus* reported in Blankenship and Yayanos (2005), indicate that the scavenging amphipod guild is capable of extensive trophic plasticity; this feature creates a complex and dynamic hadal food web and may also explain their worldwide dominance in oligotrophic trenches.

Materials and methods

Collection sites and methods—The Tonga and Kermadec Trenches are located in the southwest Pacific Ocean (parallel to the 175°W longitude line) and are the second and fourth deepest trenches worldwide. The adjacent trenches are separated by a sill (the Tonga Platform) with a shallow peak approximately 5.5 km in depth (Wright et al. 2000). The Tonga Trench axis extends from approximately 15°S to 25°S with a maximum depth of 10,882 m, and the Kermadec Trench axis continues from 25°S to 35°S with a maximum depth of 10,047 m (Belyaev 1989). The Tonga Trench lies under very oligotrophic waters where primary productivity is estimated to be < 100 mg C m⁻² d⁻¹ (Fig. 2.1 in Herring 2002). The surface waters overlying the Kermadec Trench are slightly more productive caused, in part, by an annual spring bloom (average primary productivity 100–150 mg C m⁻² d⁻¹; Fig. 2.1 in Herring 2002).

Sampling in the Tonga and Kermadec Trenches at hadal depths took place on leg nine of the Cook Cruise aboard the RV *Melville* (October 2001). The amphipods were collected from six benthic, free-vehicle (F/V) baited traps (one or two 30-liter Niskin bottles mounted to an aluminum frame, set 1 m above the bottom and open for 10–17 h) deployed to bottom depths ranging from 6,252 m to 10,787 m (Blankenship et al. 2006; Table 1). Bigeye tuna carcasses (*Thunnus obesus*) wrapped in nitex mesh and secured inside the Niskin bottles served as bait. Traps were recovered within 1 h of surfacing, and collected amphipods were immediately transferred into iced buckets until preservation. Whole amphipods and *T. obesus* (bait) muscle tissue were preserved by one of the following three methods: cold 95% ethanol for use in the molecular diet analysis, or frozen at –20°C for the duration of the cruise with subsequent storage at –70°C, or freeze-dried for stable isotope analysis.

Sediments were collected from two multicorer deployments in the Tonga Trench (23°50.0'S, 174°25.0'W, bottom depth 6,205 m; 23°20.0'S, 174°48.8'W, bottom depth 10,158 m) and one in the Kermadec Trench (32°00.0'S, 177°20.5'W, bottom depth 9,941 m). The top 1.25-cm layer of sediment from one core per deployment was frozen at –20°C onboard the ship, and later frozen at –70°C in the laboratory for stable isotope analysis.

Sex and size assessment—Seventy-five *H. dubia*, 41 *S. schellenbergi*, 19 *Uristes* sp. nov., and 5 *E. gryllus* individuals were selected for δ¹³C and δ¹⁵N stable isotope analysis. Identification of sex and estimation of size (using length of coxa four as a proxy for total size) of individual *E. gryllus*, *S. schellenbergi*, and *H. dubia* were performed as described in Blankenship et al. (2006). Neither sex nor size was determined for *Uristes* sp. nov.; their brittle exoskeleton precluded the specimen manipulation needed to estimate these parameters. Most *Uristes* sp. nov. specimens exhibited the same degree of dorsal curvature. Thus, individuals were separated into three size classes without straightening their dorsal form: juveniles (3–4 mm in length), medium-sized adults (5–8 mm), and large adults (9–10 mm).

Stable isotope analyses—Amphipod guts were flushed with milli-Q water to remove bait tissue (Gearing 1991) and then washed with milli-Q water again and oven-dried overnight. Either whole specimens or chitin and muscle tissue from the head or legs (Gearing 1991) were analyzed for δ¹³C and δ¹⁵N stable isotope ratios by continuous-flow isotope ratio mass spectrometry after acidification with PtCl₂. δ¹³C and δ¹⁵N were also determined for trench sediments from each multicorer deployment. Stable isotope ratios are expressed as (δX) ‰ = [R_{sample}/R_{standard} – 1] × 1,000, where X is either ¹³C or ¹⁵N, and R is either (¹³C:¹²C) or (¹⁵N:¹⁴N). The reference standards for carbon and nitrogen are Pee Dee Belemnite and atmospheric N₂, respectively (Michener and Schell 1994).

Statistical comparisons of isotopic data across species, *Uristes* sp. nov. size classes, sites, or depths were first performed through one-way analysis of variance, followed by the posthoc Games-Howell test for subsequent pair-wise comparisons. Size-related correlations for *S. schellenbergi*,

E. gryllus, or *H. dubia* were investigated by Pearson's correlation.

Two-source mixing models: Estimating the importance of carrion—Surface-derived nutrient input to the deep-sea benthos can be dichotomized into two components: detritus (dead organic matter including fecal pellets), which provides the main carbon source for deep-sea benthos, and larger carrion that experiences negligible bacterial degradation before reaching the seafloor bottom (Sokolova 1986). Pelagic organisms living near the ocean's surface exhibit comparable $\delta^{13}\text{C}$ signatures, but microbes continually metabolize sinking organic matter (Sokolova 1986), depleting the heavier ^{13}C isotope. Consequently, trench floor detritus and its consumers should be isotopically lighter than large carrion (Fry and Sherr 1984). We applied a single isotope, two-source mixing model to investigate whether large carrion is the sole source of carbon in the diets of scavenging amphipods (McCutchan et al. 2003). $\delta^{13}\text{C}$ values from the top 1.25 cm of trench sediment samples provided a representative endpoint for detritus and its microbial consumers. Carrion originates from the ocean surface and also deeper pelagic waters. The $\delta^{13}\text{C}$ signatures of local *T. obesus* (bait) and a midwater shrimp collected from 1,000 m in the adjacent Lau Basin (R. Lee unpubl. data) provide a lower and upper estimate of carrion contribution to amphipod diets in the following model: $F_{\text{Carrion}} = 1 - [(\delta_{\text{C}} - \delta_{\text{A}} + 0.4)/(\delta_{\text{C}} - \delta_{\text{Sed}})]$, where δ_{C} , δ_{A} , and δ_{Sed} are the $\delta^{13}\text{C}$ signatures for carrion, hadal amphipods, and trench sediment organic matter, respectively; 0.4 is the average $\delta^{13}\text{C}$ shift for one trophic level as calculated by McCutchan et al. (2003).

DNA-based diet analysis—To investigate how amphipod diets might differ between the two trenches, four *H. dubia* specimens each from F/V 12 (8,723 m, Tonga Trench) and F/V 9 (9,104 m, Kermadec Trench) were selected for a DNA-based diet analysis. The protocol was adapted from Blankenship and Yayanos (2005). The hindguts of *H. dubia* specimens were dissected with ultraviolet-sterilized equipment, and their contents (including small amounts of *H. dubia* and/or *T. obesus* tissue) were removed into DNA-free tubes. Whole genomic DNA was isolated via a forensic DNA kit (MoBio). PCR amplification of a 530–700 base pair (bp) segment of the 18S rRNA gene was performed by combining 3 μL each of the universal metazoan primers 18S E (5'CTGGTTGATCCTGCCAGT 3') and 18S L (5'GAATTACCGCGGCTGCTGGCACC 3') (10 $\mu\text{mol L}^{-1}$) (Hillis and Dixon 1991) with 25 μL HotStar Taq polymerase (Qiagen), 3 μL of gut DNA elute, and 16 μL of PCR-grade water. The PCR profile commenced with a 15-min Hotstart (95°C), one cycle of 80°C for 5 min, followed by 38 amplification cycles of 94°C for 1.5 min, 47°C for 1 min, 72°C for 2 min, and a final 10-min extension at 72°C. Controls for the PCR amplification and DNA extraction were routinely negative. *H. dubia* gut PCR products were subsequently digested with the enzyme Eco 47III to digest both host and tuna (bait) amplicons. Uncut PCR products (presumably enriched for natural diet items) were separated from digested amplicons on a 1% agarose

gel, eluted via a gel extraction kit (Qiagen), and subsequently cloned with the TOPO-TA cloning kit (Invitrogen).

Between 15 and 25 colonies were selected from each *H. dubia* gut library for sequencing. To elucidate plasmids from bacteria and create PCR templates for DNA sequencing, bacteria from each colony were picked with sterile pipette tips and added to PCR reactions containing 5 μL Lyse-N-Go (Pierce), 25 μL HotStar Taq polymerase (Qiagen), 3 μL each of M13F and M13R primers (TOPO-TA cloning kit, Invitrogen), and 14 μL sterile water. After the PCR profile (as described previously), PCR products were purified with an UltraClean PCR clean-up kit (MoBio) and sequenced using the MegaBACE 500 (Amersham). Resultant 18S ribosomal DNA (rDNA) haplotypes (unique sequences) were referenced against GenBank via BLAST to determine their approximate phylogenetic association. Any 18S rDNA sequences with 98% or more similarity to *H. dubia* or *T. obesus* (bait) were discarded. Remaining gut haplotypes were aligned with supporting 18S rDNA sequences obtained from GenBank, including those that appeared in BLAST searches, with CLUSTAL X (default settings), and the alignment was manually refined with MACCLADE. A phylogenetic tree of gut sequences was created by distance analysis (neighbor-joining) based on an alignment of 698 bp (Paup 4.0), and the taxonomic origin of each haplotype was resolved from its topological positioning within the tree.

Blankenship and Yayanos (2005) published a similar molecular dietary study using mitochondrial cytochrome oxidase I (*COI*) as the molecular marker to analyze gut contents of one *E. gryllus* (6.3 km) and two *S. schellenbergi* (7.3 and 8.7 km) from the Tonga Trench. At the time of publication, DNA belonging to at least one species of teleost and several amphipods were discerned, but the remaining 14 haplotypes were taxonomically unresolved. Here, these ambiguous sequences were re-referenced in BLAST in search of finer taxonomic resolution. Several gut haplotypes returned matches that provided further taxonomic insight and thus were combined in a new *COI* phylogenetic tree as described above (based on predicted amino acid translations).

Results

Isotopic evidence for nutritional sources and trophic position—Collectively, amphipod $\delta^{13}\text{C}$ values ranged from -11.4‰ to -23.7‰ (Table 2), suggesting variability in diet. Most amphipod signatures were between those of the ^{13}C -depleted sediments (-21.4‰ , -22.3‰ , and -22.8‰ for Tonga Trench 6,205 m and 10,158 m, and Kermadec Trench 9,941 m, respectively) and ^{13}C -enriched bait tuna (-16.3‰). Results from single isotope, two-source mixing models indicated that large carrion is not the exclusive source of nutrition for any of the scavenging species (Table 3). Nutritional sources that would lighten amphipod $\delta^{13}\text{C}$ signatures include surface-derived detritus and its consumers (as indicated by the isotopically light-trench sediments) (Fry and Sherr 1984) and possibly sulfide-based chemosynthesis (chemoautotrophic bacterial $\delta^{13}\text{C}$ signa-

Table 2. The average $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ isotope values for each amphipod species categorized by trench, depth, and size.

Species	Trench*	n	Size range (mm)	$\delta^{13}\text{C}$ (‰)±SE	$\delta^{15}\text{N}$ (‰)±SE
<i>Eurythenes gryllus</i>					
6.3–6.8 km	TT	5	25–35	−19.42±0.92	12.02±0.15
<i>Scopelocheirus schellenbergi</i>					
6.3–6.8 km	TT	3	21–36	−19.04±0.71	10.65±0.75
8.6–8.7 km	TT	37	26–41	−19.69±0.20	11.02±0.19
9.1 km	KT	1	31	−20.5	9.7
Species average		41		−19.67±0.19	10.96±0.18
<i>Uristes</i> sp. nov.					
8.6–8.7 km	TT				
Juveniles		4	3–4	−19.29±0.41	10.21±0.87
Medium-sized adults		6	5–8	−19.46±0.27	11.06±0.59
Large adults		9	9–10	−18.93±0.24	12.36±0.40
Species average		19		−19.18±0.13	11.45±0.37
<i>Hirondellea dubia</i>					
8.6–8.7 km	TT				
Juveniles		12	4–9	−18.11±0.53	10.04±0.26
Medium-sized adults		11	11–15	−17.45±0.70	11.29±0.37
Large adults		13	17–21	−18.39±0.47	11.73±0.29
Average		36		−18.02±0.32	11.09±0.21
9.1 km	KT	19	13–19	−19.89±0.26	11.69±0.21
10.8 km	TT	20	11–17	−19.85±0.34	11.98±0.15
Species average		75		−18.98±0.22	11.48±0.13

* TT, Tonga Trench; KT, Kermadec Trench.

tures can be as heavy as -27% , but are usually lighter; Robinson and Cavanaugh 1995). As a caveat, the same trends were not apparent for the $\delta^{15}\text{N}$ data. Thus, integration of $\delta^{15}\text{N}$ values for a two-isotope mixing model did not improve the resolution.

The amphipod $\delta^{15}\text{N}$ values ranged from 7.9‰ to 13.8‰. This suggests the hadal scavenging community spans several trophic levels (Table 2). Most amphipod $\delta^{15}\text{N}$ signatures were considerably enriched compared to the bait (8.3‰) and trench sediment (5.8‰, 5.0‰, and 6.0‰ for Tonga Trench 6,205 m and 10,158 m, and Kermadec Trench 9,941 m, respectively). Notably, average $\delta^{15}\text{N}$ amphipod signatures are at least one trophic step above the $\delta^{15}\text{N}$ value reported for *T. obesus* (8.32‰), which is the epitome of a large nekton carcass. However, the analyzed *T. obesus* specimen was a juvenile, and Allain et al. (pers. comm.) reported a large ontogenetic $\delta^{15}\text{N}$ shift for this species; adult *T. obesus* $\delta^{15}\text{N}$ signatures from nearby water masses (New Caledonia) average 12.41‰.

Isotopic differences among species, size classes, depths, and trenches—Depth-integrated $\delta^{13}\text{C}$ signatures were not significantly different among species ($F_{3,135} = 1.696$, $p = 0.171$) (Fig. 2A). However, interspecific $\delta^{13}\text{C}$ differences were observed between the three species coexisting at 8.6–8.7 km within the Tonga Trench ($F_{2,73} = 12.362$, $p < 0.001$) (Fig. 2B). At these depths, adult *H. dubia* were enriched in $\delta^{13}\text{C}$ as compared to *Uristes* sp. nov. and *S. schellenbergi* ($p = 0.029$ and $p = 0.001$, respectively), suggesting that large carrion was a more significant component of the *H. dubia* diet than either *Uristes* sp. nov. or *S. schellenbergi* diets (Table 3). Moreover, the $\delta^{13}\text{C}$ signatures of *H. dubia*

caught in the deepest depths of the Tonga Trench (10.8 km) were also significantly lighter than their shallower counterparts ($F_{1,42} = 3.381$, $p = 0.002$) (Fig. 2B). This presumably reflects a reduction of carrion in the diets of *H. dubia* inhabiting the deepest depth zones of the Tonga Trench.

Traps attract a localized population of amphipods. It is possible that heavy $\delta^{13}\text{C}$ signatures simply reflect a random recent event of carrion fall in the area. Thus, it is important to elucidate whether the $\delta^{13}\text{C}$ differences between traps is the result of depth differences or random carrion flux into the area before trapping. F/V 6 and F/V 12 are considered replicates for the intermediate depths, because they are separated by >500 miles but depths differ by only 150 m. A Kolmogorov-Smirnoff comparison of *H. dubia* and *S. schellenbergi* $\delta^{13}\text{C}$ data from these two traps revealed the $\delta^{13}\text{C}$ for these populations were not significantly different (both $p > 0.1$). This suggests that depth is a pervasive factor in specific $\delta^{13}\text{C}$ signatures.

No interspecific $\delta^{13}\text{C}$ differences were observed for coexisting *E. gryllus* and *S. schellenbergi* species collected from the shallower hadal depths of the Tonga Trench (6.3–6.8 km), although the sample size was small ($n = 5$ and $n = 3$, respectively). *S. schellenbergi* captured between 6.3 km and 6.8 km exhibited heavier $\delta^{13}\text{C}$ values than those trapped between 8.6 km and 8.7 km (Fig. 2B), but the difference was not statistically supported ($p = 0.46$). However, statistical sensitivity was compromised by the extremely small sample size ($n = 3$) from 6.3–6.8 km.

No intra- or interspecific correlations between $\delta^{13}\text{C}$ values and amphipod size were discerned (all $p > 0.05$), and thus we found no evidence from the $\delta^{13}\text{C}$ data that the

Table 3. Estimated contribution of large carrion (F_{Carrion}) based on $\delta^{13}\text{C}$ isotopic two-source mixing models assuming a 0.4‰ trophic shift. Lower and upper estimates were calculated by substituting *T. obesus* (tuna) $\delta^{13}\text{C}$ value of -16.3‰ and the midwater shrimp value of $\delta^{13}\text{C} -17.8\text{‰}$ for δ_{C} , respectively. TT and denotes the Tonga Trench and KT denotes the Kermadec Trench.

Location and species	Amphipod Avg. $\delta^{13}\text{C}$ values (‰)	Sediment sample and depth (m)	Sediment $\delta^{13}\text{C}$ (‰)	Average F_{Carrion}	
				Lower estimate	Upper estimate
Tonga Trench					
Shallow: 6.3–6.8 km					
<i>Eurythrenes gryllus</i>	-19.42	TT 6,205	-21.4	0.31	0.66
<i>Scopelocheirus schellenbergi</i>	-19.04	TT 6,205	-21.4	0.38	0.76
Intermediate: 8.6–8.7 km					
<i>S. schellenbergi</i>	-19.69	TT 10,158	-22.3	0.37	0.66
<i>Uristes sp. nov.</i>	-19.18	TT 10,158	-22.3	0.45	0.78
<i>Hirondellea dubia</i>	-18.02	TT 10,158	-22.3	0.64	>1.0
Deep: 10.8 km					
<i>H. dubia</i>	-19.85	TT 10,158	-22.3	0.34	0.63
Kermadec Trench					
Intermediate: 9.1 km					
<i>S. schellenbergi</i> *	-20.5	KT 9,941	-22.8	0.29	0.54
<i>H. dubia</i>	-19.89	KT 9,941	-22.8	0.39	0.66

* Based on one specimen.

amphipod guild partitions nutritional sources (e.g., detrital-based food chain or large carrion) based on size.

Average $\delta^{15}\text{N}$ comparisons revealed significant differences among amphipod species ($F_{3,132} = 2.443$, $p = 0.067$), due mainly to the interspecific difference between *S. schellenbergi* and *E. gryllus* ($p = 0.002$) (Table 2; Fig. 2A). In fact, *S. schellenbergi* had the lowest average $\delta^{15}\text{N}$ values of all analyzed species (10.96‰), indicating this species tends to feed at about half a trophic level below juvenile *E. gryllus*, which had the highest average $\delta^{15}\text{N}$ values (12.02‰). Moreover, *S. schellenbergi* exhibited a much

wider range of $\delta^{15}\text{N}$ values ($\sim 5.1\text{‰}$) than *E. gryllus* (range of 1‰), although the *E. gryllus* sample size was considerably smaller.

H. dubia captured from 9.1 km in the Kermadec Trench were significantly depleted in ^{13}C as compared to those specimens caught from 8.6 km to 8.7 km in the Tonga Trench ($F_{1,42} = 3.693$, $p = 0.001$). These data suggest that carrion is of greater importance in the diet of the Tonga Trench *H. dubia* population than the Kermadec Trench population at comparable depths (Table 3).

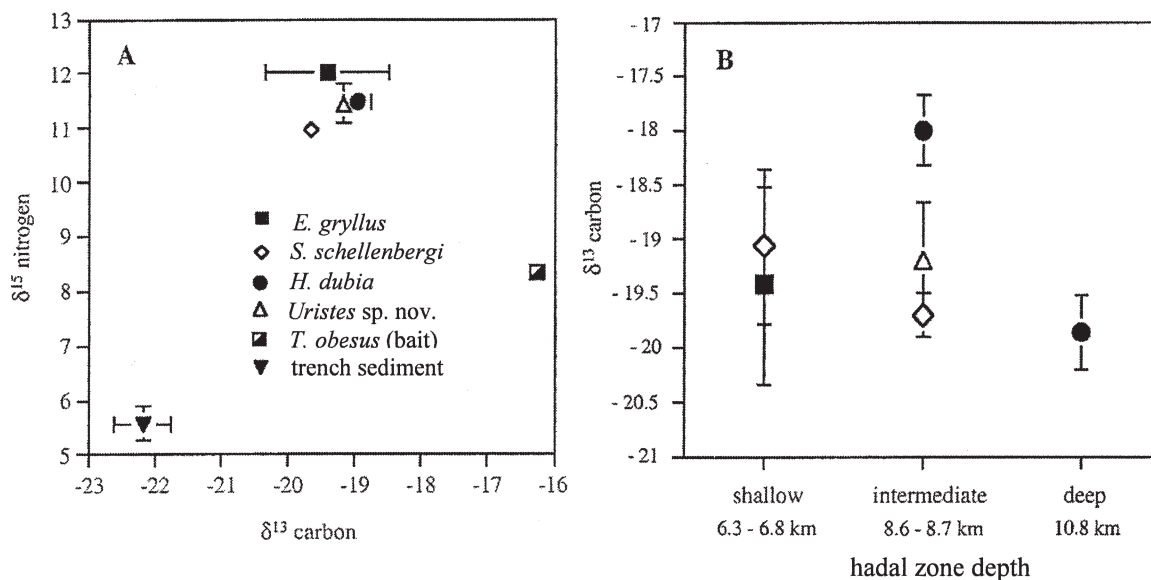


Fig. 2. Specific average stable isotope values of the hadal amphipod community. Bars represent standard errors; some standard errors are too small to visualize. (A) Dual isotope plot depicting average $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ signatures for four amphipod species, tuna (bait), and trench sediment. (B) Average $\delta^{13}\text{C}$ signatures for each amphipod species versus depth in the Tonga Trench. The specific $\delta^{13}\text{C}$ values of coexisting amphipods trapped between 8.6 km and 8.7 km are significantly different ($p < 0.0001$).

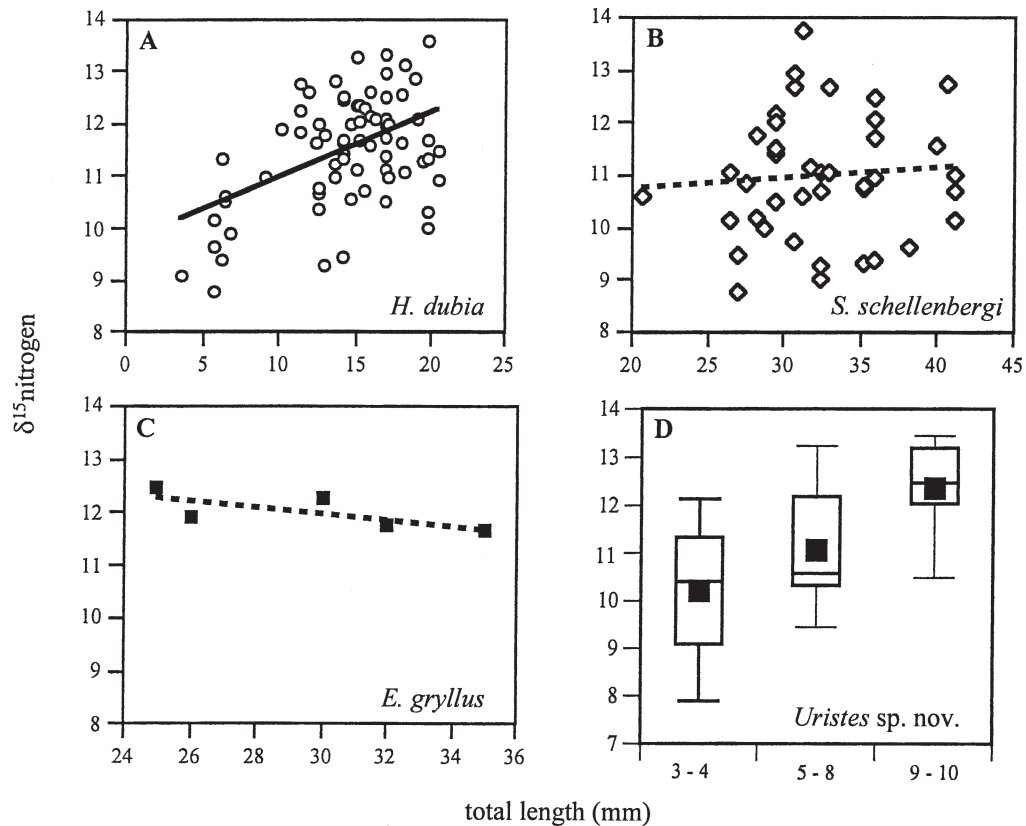


Fig. 3. The $\delta^{15}\text{N}$ values of *H. dubia*, *S. schellenbergi*, *E. gryllus*, and *Uristes* sp. nov. as a function of body size. (A–C) Simple linear regression trends are displayed; solid lines indicate the correlation is significant ($p < 0.05$) whereas dashed lines indicate the correlation is not significant ($p > 0.05$). (D) Means are represented by filled squares and the *Uristes* sp. nov. $\delta^{15}\text{N}$ signatures increase with size ($p = 0.05$).

Positive correlations between size and $\delta^{15}\text{N}$ signatures were observed for the two smaller and demersal amphipod species, *Uristes* sp. nov. ($F_{2,15} = 3.574$, $p = 0.054$) and *H. dubia* ($r = 0.474$, $p < 0.001$) (Fig. 3). These data suggest an ontogenetic shift in diet; larger dietary items become increasingly important with age. The $\delta^{15}\text{N}$ signatures of the larger species, *E. gryllus* and *S. schellenbergi*, did not show any correlation with size (all $p > 0.05$). However, only juvenile *E. gryllus* and subadult or adult *S. schellenbergi* were available for isotope analyses, so their full range of developmental stages was not analyzed (Fig. 3).

H. dubia DNA-based diet analyses—Between zero and four gut haplotypes were discerned from each clone library, with *H. dubia* 18S rDNA amplicons constituting between 15% and 100% of the clones selected from each library. Collectively, 17 sequences were determined from *H. dubia* gut contents, and at least 14 of these were unique haplotypes (GenBank Accession Nos. DQ177348–DQ177364). The gut sequences assembled into five topological groups within the phylogenetic tree (Fig. 4), and there was a surprising similarity between sequences detected from the Tonga Trench and Kermadec Trench amphipod guts. Animal 18S rDNA haplotypes constituted the minority and included only DNA from fish and amphipods (Fig. 4). Clone KT3-B was the sole fish haplotype, and its high degree of similarity to *Auxis rochei*

(mackerel) and *T. obesus* classify this clone as a perciform. The amphipod sequences TT5-C and KT2-B exhibited 99.7% and 99.4% sequence similarity, respectively, with *S. schellenbergi* (captured from the Tonga Trench), indicating that *H. dubia* consumes *S. schellenbergi*. Clone TT5-B likely originated from another species of gammarid amphipod not captured during the Cook Expedition.

The remaining nonanimal gut sequences topologically associate with fungi, diatoms, or alveolates (Fig. 4). Fungi haplotype KT2-C is probably red yeast, possibly a member of the genus *Rhodotorula*, a ubiquitous deep-sea fungus previously isolated from the Japan, Palau-Yap, and Mariana Trenches (Nagahama et al. 2001, 2003). Fungal clone TT1-C exhibited 98% sequence similarity with the ascomycete *Peziza phyllogena*, and most top BLAST returns belonged to this genus. At least three species of diatoms are represented by sequences KT1-B, TT1-A, TT5-A, and KT4-A. Sequences KT1-B and TT1-A are 99% identical to one another, perhaps denoting the same species of diatom. Amplified 18S rDNA sequences clustering within the Alveolate lineage were abundant, and these haplotypes clustered into two distinct groups. Identical sequences KT3-A and TT1-B are probably ciliates. Their nearest topological neighbor (clone AT7-37) is also a ciliate isolated from a hydrothermal area (Lopez-Garcia et al. 2003), and the majority of the BLAST returns also belonged to Ciliophora (data not shown). A more precise

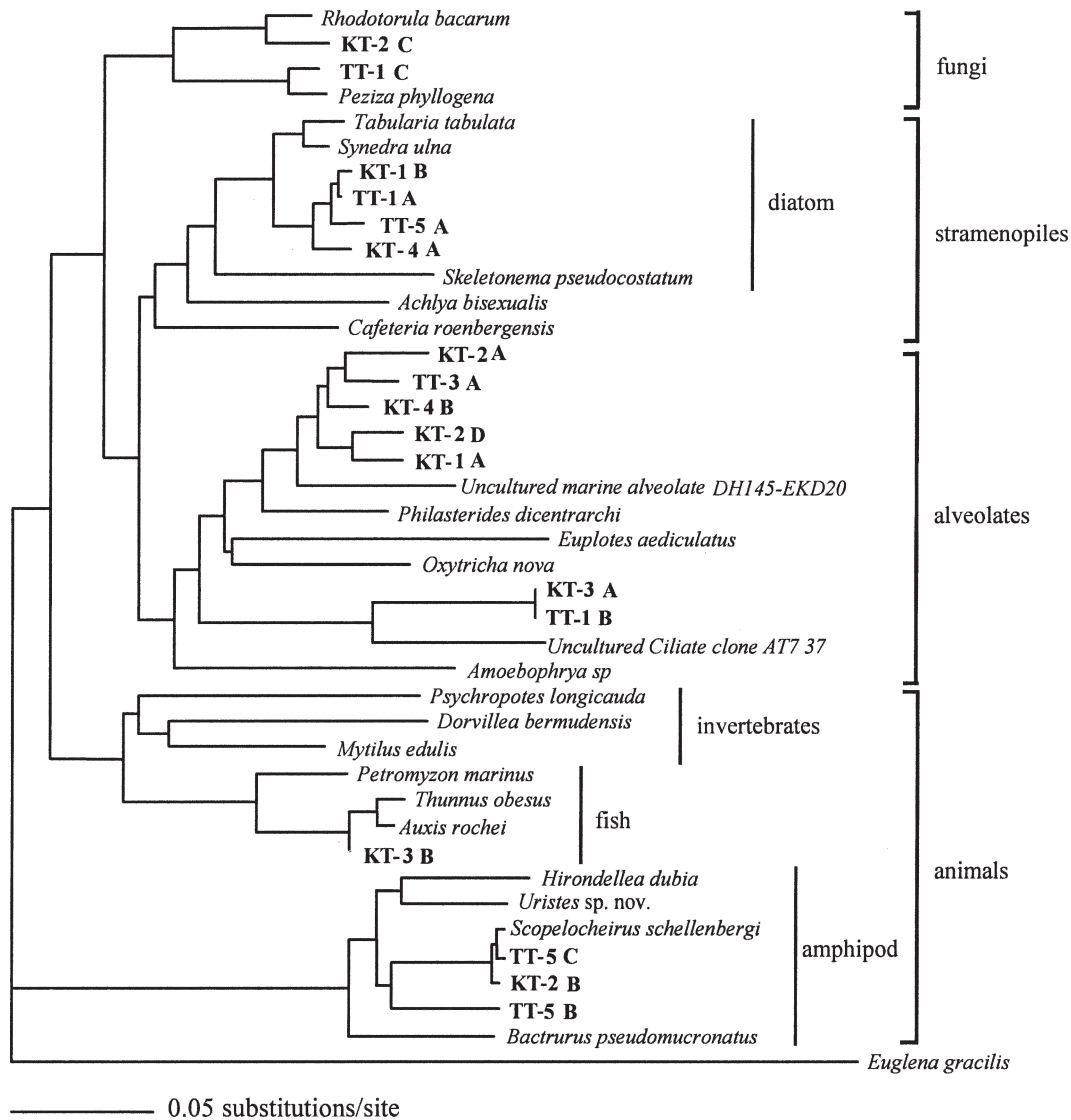


Fig. 4. Neighbor-joining phylogenetic tree of *Hirondellea dubia* gut 18S rDNA sequences based on 678 nucleotide positions. Sequences recovered from *H. dubia* guts are labeled KT or TT (amphipod captured in Kermadec or Tonga Trench, respectively), followed by the sample number and unique haplotype letter. *Euglena gracilis*, which is outside the eukaryotic crown, is the outgroup.

taxonomic identification of the alveolate cluster represented by haplotypes KT2-A, TT3-A, KT4-B, KT2-D, and KT1-A is difficult to ascertain, in part because of the taxonomic ambiguity of the closest neighbor (alveolate clone DH145-EKD20; Lopez-Garcia et al. 2001). Recent 18S rDNA marine surveys have uncovered an abundance of alveolates that do not affiliate with known lineages such as dinoflagellates and ciliates; DH145-EKD20 is one such “unassigned” alveolate (Edgcomb et al. 2001; Lopez-Garcia et al. 2001). Several of these gut haplotypes (e.g., KT4-B) actually attracted *Philasterides dicentrarchi* (a ciliate) as the most relevant match during BLAST searches (~91% sequence similarity), and ciliates also tended to dominate BLAST returns, further confounding the taxonomic resolution of these five gut haplotypes.

Reanalysis of E. gryllus and S. schellenbergi gut DNA—A new analysis of five *E. gryllus* gut haplotypes and one *S.*

schellenbergi gut haplotype suggests these sequences, which were previously unresolved, are actually urochordates (Fig. 5). The phylum Urochordata (tunicates) includes salps, larvaceans, and ascidians. Interestingly, the phylogenetic tree published in Blankenship and Yayanos (2005) included two *COI* ascidian reference sequences (*Ciona intestinalis* and *Halocynthia roretzi*), although these six gut haplotypes did not associate with these two ascidian sequences. Stach and Tuberville (2002) note that the tunicate *COI*-based phylogeny is not congruent with 18S rDNA and morphological phylogenies; our neighbor-joining tree corroborates this and shows that the urochordate *COI* amino acid phylogeny clusters into two distinct groups (Fig. 5). The six amphipod gut haplotypes appear to associate only with the alternate urochordate cluster that was not included in the tree published by Blankenship and Yayanos (2005). Notably, the vast majority of urochordate *COI* sequences available in

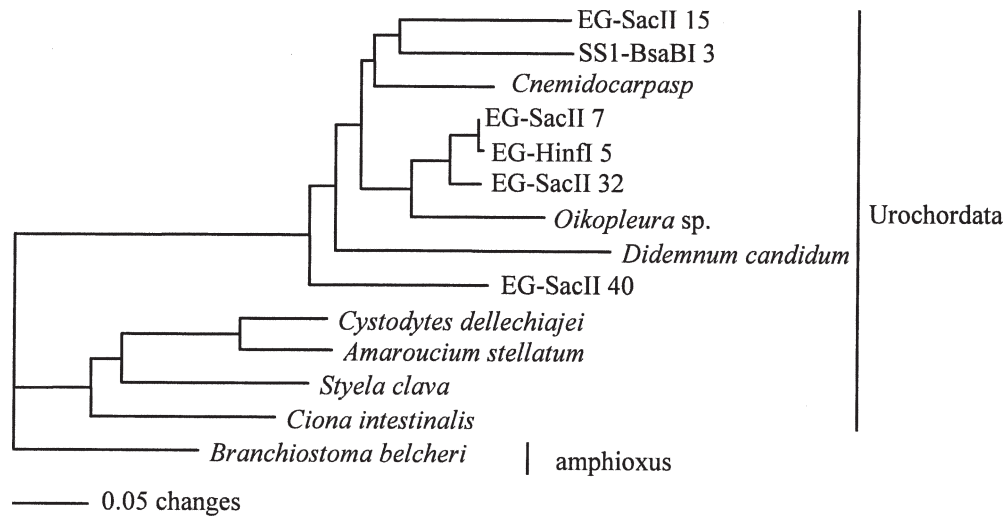


Fig. 5. Neighbor-joining phylogenetic tree of six *COI* gut haplotypes from *E. gryllus* and *S. schellenbergi* based on 230 predicted amino acid positions. All supporting sequences are ascidians with the exception of *Oikopleura sp.*, which is a larvacean, and the outgroup *Branchiostoma belcheri*, which belongs to amphioxus.

GenBank are ascidians; the lack of adequate larvacean and salp representatives precludes further resolution of these gut sequences.

Discussion

Is nekton carrion the sole source of nutrition for the scavenging guild?—Carrion does not appear to be the exclusive food source for any of the scavenging amphipod species. Thus, contrary to some current views, there are no obligate necrophagous species in this hadal scavenging guild. The true importance of carrion is impossible to quantify, however, because the proportion of carrion originating from the surface versus deeper waters and the isotopic range of possible carrion items is unknown. Molecular dietary analyses support the theory of facultative scavengers as DNA sequences belonging to probable nekton carcasses (e.g., fish) constituted the unexpected minority of dietary DNA detected in every examined amphipod gut (this study and Blankenship and Yayanos 2005).

Alternative nutritional strategies and diets of lysianassoids—Deep-sea scavenging lysianassoids are divided into the benthopelagic and demersal guilds (Sainte-Marie 1992). The larger benthopelagic *S. schellenbergi* and *E. gryllus* are thought to rely heavily, if not exclusively, on nekton carrion (Thurston 1990; Kaiser and Moore 1999; Dauby et al. 2001) because of the efficiency with which they can locate, ingest, and store large amounts of carrion (Sainte-Marie 1992). The smaller demersal species *Uristes sp. nov.* and *H. dubia* do not possess the size or extensive gut capacity common in benthopelagic lysianassoids (Dahl 1979; Blankenship et al. 2006) and are generally considered facultative scavengers (Dahl 1979; Sainte-Marie 1992).

We propose that the benthopelagic *S. schellenbergi* and *E. gryllus* populations employ predation, and possibly

detritivory, when nekton carrion is unavailable. The presence of dietary amphipod DNA in their guts (Blankenship and Yayanos 2005) is suggestive of predation because trophically diverse amphipods are ubiquitous in the hadal zone (Kamenskaya 1995). Predation on other amphipods is common; gut content analyses of carnivorous lysianassoids inhabiting shallow waters often reveal amphipod remains (Sainte-Marie 1986), and deep-sea lysianassoids are known to cannibalize other lysianassoids in artificial settings (Ingram and Hessler 1983). Urochordates could be taken by either predation or necrophagy. Sessile ascidians are abundant in the abyss and shallow hadal zone (Belyaev 1989) and would be a natural prey choice for predatory amphipods inhabiting the shallower hadal depths (i.e., juvenile *E. gryllus*). Pelagic salps or larvaceans could be obtained either via predation in the water column or as carrion on the seafloor, and urochordate remains are thought to be a significant carbon sink to the seafloor (Deevey and Brooks 1971; Robison et al. 2005).

S. schellenbergi and *E. gryllus* may also consume microeukaryotes, either purposely or accidentally, while feeding at the bottom. One DNA haplotype recovered from a *S. schellenbergi* gut may represent a stramenopile (a protist group containing diatoms and brown algae) (Blankenship and Yayanos 2005). Moreover, Kamenskaya (pers. comm.) found fragments of bottom animals together with detritus and mineral grains in the stomach contents of some *S. schellenbergi* from other trenches and also found large mineral grains mixed with animal tissues in the guts of four *E. gryllus* specimens.

Surprisingly, $\delta^{15}\text{N}$ data suggest that juvenile *E. gryllus* feed at a higher trophic level than adult *S. schellenbergi* of similar size. This result was unanticipated since these amphipod species are similar in size, belong to the benthopelagic guild, and the results of their DNA-based analysis of gut contents imply congruence in diets (Blankenship and Yayanos 2005). If the frequency and

type of carrion does not change substantially with depth, then the $\delta^{15}\text{N}$ data imply interspecific divergence of nonscavenging foraging strategies.

One possible explanation is that *S. schellenbergi* supplements necrophagy mainly by preying on smaller benthic organisms. Antarctic amphipods that derived a significant portion of their diet from preying on benthic fauna, such as holothurians and polychaetes, exhibited lower average $\delta^{15}\text{N}$ signatures as well as more intraspecific variation than amphipod species that relied mostly on large carrion (Nyssen et al. 2002). In contrast, *E. gryllus* may rely on the predation of larger organisms at higher trophic levels. This aggressive species certainly appears capable of taking larger prey. They are known to exhibit cannibalistic behavior when maintained in crowded aquaria (Dauby et al. 2001) and have been observed attacking live fish that are restrained on a hook and line (Smith and Baldwin 1984; Thurston 1990). Alternatively, the interspecific $\delta^{15}\text{N}$ signatures may reflect the changing availability of food with depth, as both detrital quality and hadal biodiversity are thought to decrease with depth (Belyaev 1989; Sokolova 1994). Notably, the apparent discrepancy between the interspecific diet congruence reported in Blankenship and Yayanos (2005) and the interspecific $\delta^{15}\text{N}$ differences reported here may stem from the limitations and biases inherent in each type of analysis. For example, highly degraded dietary DNA in the hindgut of the amphipods, small sample sizes, and biases inherent in the DNA-based technique almost certainly restricted the scope of dietary organisms that were detected by this method (see Blankenship and Yayanos 2005 for discussion).

H. dubia uses an impressive spectrum of diet and foraging modes. Microeukaryotes appear pervasive in the gut contents of this species, indicating that adults ingest sediment. Diatoms are likely surface-derived detritus, although the fungi and some alveolate species (particularly the predatory ciliates) may be autochthonous (Takami et al. 1997; Lopez-Garcia et al. 2003; Nagahama et al. 2003). The detection of *S. schellenbergi* DNA from the guts of two *H. dubia* specimens was an extraordinary result. We interpret this finding, in conjunction with another unresolved amphipod DNA sequence, as evidence that *H. dubia* preys on other amphipods. *S. schellenbergi* is at least twice the size of *H. dubia*, but large size does not appear to offer sanctuary from becoming potential prey. Ingram and Hessler (1983) report that scavenging amphipods commonly prey on other amphipods within a baited trap; larger individuals, regardless of species, appear to be preferentially preyed upon. Whether this intrataxocene predation is an artifact of trapping or representative of natural events remained an open question (Ingram and Hessler 1983). We argue that *H. dubia* feeding on *S. schellenbergi* is a natural event. The trap deployed at 9.1 km in the Kermadec Trench captured >9,000 *H. dubia*, but only two *S. schellenbergi* individuals; both *S. schellenbergi* specimens were intact (Blankenship et al. 2006). Thus, *H. dubia* feeding on *S. schellenbergi* in this trap is unlikely. Moreover, *S. schellenbergi* DNA was detected in one-fourth of the analyzed *H. dubia* guts. Adult *H. dubia* $\delta^{15}\text{N}$ signatures were also consistently enriched relative to adult

S. schellenbergi $\delta^{15}\text{N}$ values, further supporting this contention.

Is there evidence of competition among coexisting amphipod species?—Average depth-integrated isotopic ratios are startlingly comparable across species, superficially suggesting that species occupy similar trophic niches (Hobson and Welch 1992). However, the significant $\delta^{13}\text{C}$ differences observed among three species coexisting at 8.6–8.7 km in the Tonga Trench indicate that species foraging roles change with depth (Fig. 2B). We propose that this depth-related $\delta^{13}\text{C}$ shift reflects a form of character displacement (diet partitioning, sensu Fenchel 1975) that may be driven by competition among coexisting amphipod species. We believe this to be the first evidence of competition in the hadal zone.

The larger benthopelagic *S. schellenbergi* would be expected to consume more carrion than the smaller demersal *H. dubia* and *Uristes* sp. nov. species. This does not appear to be the case. *S. schellenbergi* actually exhibits the lowest $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of the scavenging guild coexisting at 8.6–8.7 km. Instead, *H. dubia* dominates the carrion consumption (Table 3). Moreover, *H. dubia* appears to prey on *S. schellenbergi*, which may further deter *S. schellenbergi* from approaching carrion in this depth range (Thurston and Bett 1995). *Uristes* sp. nov. is probably best suited to capitalize on smaller carcasses (e.g., copepods) along with detritus and small fauna in the sediment (Sainte-Marie 1986; Dauby et al. 2001; Klages et al. 2001).

Do specific foraging strategies shift with depth or age?—Detritivores (including deposit and suspension feeders) often exhibit lighter $\delta^{15}\text{N}$ signatures than carnivores (Nyssen et al. 2002). Thus, we interpret the ontogenetic $\delta^{15}\text{N}$ pattern observed for both *H. dubia* and *Uristes* sp. nov. (Fig. 3) as evidence that juveniles rely on detritivory to a greater degree than adults. Similar ontogenetic shifts from mainly detritivory to carnivory have been noted in other lysianasoid species based on gut content analyses (Christiansen and Diel-Christiansen 1993), including *Hirondellea gigas* from the Philippine Trench (Hessler et al. 1978). Adults have the capacity to withstand long periods between meals because of their lipids stockpiled from previous nutritious meals, but recently hatched individuals may not have this metabolic advantage (Ingram and Hessler 1987). Thus, detritivory or feeding on small carcasses offers a means to sustain energetic requirements between episodes of large carrion fall (Dahl 1979). Notably, chitin tends to be significantly impoverished in $\delta^{15}\text{N}$ as compared to muscle tissue (Cherel and Hobson 2005). Since juvenile amphipods tend to exhibit a higher chitin to muscle mass ratio than adults, it is possible that chitin additively biases this ontogenetic pattern to some degree.

In the deepest depths of the Tonga Trench (10.8 km), the depleted $\delta^{13}\text{C}$ signatures of *H. dubia* imply that their nutrition is mostly derived from a detrital-based food chain. Since larger carrion should be preferentially consumed (Stockton and DeLaca 1982; Solokova 1986), this finding suggests a trophic shift necessitated by the lack of

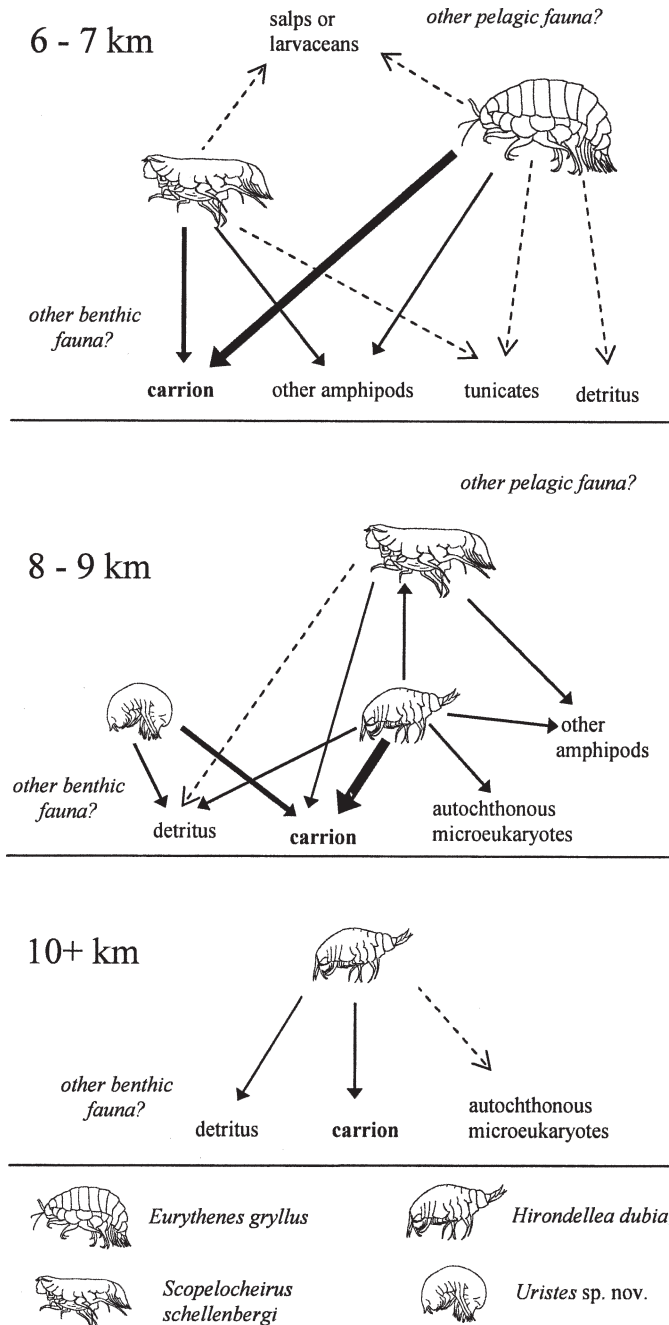


Fig. 6. A preliminary hadal food web in the Tonga Trench. Solid lines indicate dietary items supported by molecular diet analyses or stable isotope ratios, whereas dashed lines are potential food items suggested by unresolved or indirect evidence. The estimated degree of carrion partitioning among coexisting species is displayed by the thickness of the solid line. Many other benthic and pelagic fauna are likely to be important components of this food web, but have not been detected in these studies.

carrion at these depths. Likewise, several deep-sea crustaceans are known to exhibit trophic shifts within their vertical range, presumably reflecting shifting food availability (Cartes et al 1998; Carney 2005). However, this

conclusion is based on one trapping at these great depths, and is thus subject to caution.

Notably, mitochondrial DNA-based analyses (16s rDNA and COI) of several *H. dubia* specimens (whole specimens with guts) from 10.8 km uncovered bovine DNA (L. Blankenship, unpubl. data). This enlightening discovery suggests that anthropogenic input (e.g., ship galley discards) influences the ocean's deepest food web.

Implications for hadal food webs—A comparison of *H. dubia* diets from each trench provides insight into how the amphipod food webs differ between the Tonga and Kermadec Trenches. Both trenches exceed 10 km in depth and contain similar scavenging assemblages (Blankenship et al. 2006). Because of increased primary productivity at higher latitudes, detritus reaching the Kermadec Trench floor is probably greater in quality and quantity than detrital deposition in the Tonga Trench (Sokolova 1986). As a result, the Kermadec Trench benthic community should also exhibit higher biomass. *H. dubia*, which appears to be a highly opportunistic feeder, may take advantage of these improved feeding conditions in the Kermadec Trench by foraging to a greater extent on detritus and its consumers. Accordingly, Blankenship et al. (2006) found evidence that *H. dubia* females were sexually maturing at a smaller size in the Kermadec Trench, an observation that implied a more frequent feeding routine. Thus, our results suggest that the importance of large carrion to the hadal scavenging guild is influenced by the nutritional properties of the trench. Specifically, carrion appears to be a more important dietary component when other sources of food are lower in quality or quantity (see Table 3).

The hadal food web also changes with depth due to changing faunal composition and nutrient flux (Fig. 6). Consequently, hadal food webs are a property of both the trench and depth; they are influenced by overlying surface production, community composition, depth, and proximity to land masses and thus terrestrial input. For example, eutrophic trenches tend to support higher benthic biomass (Jumars and Hessler 1976) and, in some cases, a more speciose scavenging assemblage (Belyaev 1989; Kamenskaya 1995; Perrone et al. 2002). The degree of similarity in food-web structure, and the importance of carrion to the scavenging guild, between oligotrophic and eutrophic trenches remains to be addressed.

Does trophic plasticity confer success in the hadal zone?—Amphipods are one of the most trophically diverse taxa in the marine environment (Nyssen et al. 2002). Feeding ecology studies from shallower depths reveal that both the taxocene and individual species often comprise multiple trophic types; this diversity appears to contribute to their adaptive radiation in some marine environments (Sainte-Marie 1986; Dauby et al. 2001; Nyssen et al. 2002). For example, the Weddell Sea benthic amphipod community includes almost all known feeding types (e.g., suspension feeder, scavenger, and browser) (Dauby et al. 2001). The stable isotope range of the entire amphipod community is quite broad ($\delta^{13}\text{C} \sim 9\%$ and $\delta^{15}\text{N} \sim 7.5\%$), but the range observed for the resident scavenging guild is more restricted

($\delta^{13}\text{C} \sim 5\text{‰}$ and $\delta^{15}\text{N} \sim 3\text{‰}$ based on three species, 15 individuals) (Nysse et al. 2002). Similarly, the carnivorous amphipod guild from the Porcupine Abyssal Plain (north-east Atlantic abyss) also revealed a more restricted isotopic range of approximately 4‰ for $\delta^{13}\text{C}$ and 5‰ for $\delta^{15}\text{N}$ (Iken et al. 2001). In comparison, the isotopic range exhibited by the Tonga and Kermadec Trenches hadal scavenging amphipod guild is quite large ($\delta^{13}\text{C} \sim 12\text{‰}$ and $\delta^{15}\text{N} \sim 6\text{‰}$), particularly for $\delta^{13}\text{C}$. The range observed for individual species is equally as impressive; *H. dubia* $\delta^{13}\text{C}$ values spanned more than 11.5‰, whereas *S. schellenbergi* and *Uristes* sp. nov. $\delta^{15}\text{N}$ signatures covered a range of 5.0‰ and 5.6‰, respectively. Hence, the hadal scavenging amphipods appear to express extraordinary trophic diversity as a guild and considerable elasticity within individual species.

In oligotrophic trenches, conditions of extreme food limitation would certainly favor animals capable of a diverse diet to ensure acquisition of enough nutrients for survival. Indeed, obligate necrophagy does not appear to be a feasible foraging strategy in the Tonga and Kermadec Trenches. Instead, these hadal scavengers exhibit impressive trophic plasticity that enables them to assume a variety of roles in hadal food webs and optimize the poor or sporadic feeding conditions. Moreover, the ease with which these scavenging amphipods shift from one trophic type to another enables them to reduce the rate of competition to facilitate coexistence (Huston 1979); this may ultimately be an important mechanism in maintaining hadal Lysianassoid biodiversity.

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Received: 22 January 2006

Accepted: 17 January 2007

Amended: 8 January 2007