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Occurrence of 16:2(n-4) and 18:2(n-4) fatty acids in the lipids of the hydrothermal vent shrimps *Rimicaris exoculata* and *Alvinocaris markensis*: nutritional and trophic implications

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ABSTRACT: Adults of 2 species of vent shrimp, *Rimicaris exoculata* and *Alvinocaris markensis*, were sampled from the Snake Pit and TAG hydrothermal vent sites on the mid-Atlantic ridge. Fatty acid analyses indicated high abundances of 16:2(n-4) and 18:2(n-4) in the lipids of *R. exoculata*, with the highest proportions of these fatty acids detected in the digestive gland [14.5% 16:2(n-4) and 23.8% 18:2(n-4)]. Lipid extracted from abdominal muscle of *R. exoculata* also contained these fatty acids, although in lower proportions [2.0% 16:2(n-4) and 14.5% 18:2(n-4)]. By contrast, lipid extracted from the same tissues in *A. markensis* contained relatively low proportions of (n-4) fatty acids (1.9 to 3.0%), but was substantially enriched in the phototrophic, microplanktonic biomarkers 20:5(n-3) and 22:6(n-3). GC-IRMS (gas chromatography with isotope ratio mass spectrometry) analysis of the (n-4) dienoic fatty acids established $\delta^{13}C$ values of −11.0 to −11.4‰ which is consistent with the fatty acids being derived from chemosynthetically fixed carbon. By contrast, those fatty acids which are characteristic of phototrophic microplankton were isotopically lighter, i.e. −17.1, −17.3 and −15.8‰ for 20:4(n-6), 20:5(n-3) and 22:6(n-3) respectively. The non-methylene interrupted dienes (NMIDs) 20:2Δ5,13 and 22:2Δ7,15 were detected in all samples although in small amounts with *R. exoculata* containing the highest amounts. The (n-4) dienes and NMIDs are consistent with a substantial dietary input of bacterially derived 16:1(n-7) and 18:1(n-7) fatty acids for this species. Thus, these results are consistent with *R. exoculata* adopting a bacterivorous mode of nutrition whilst *A. markensis* is essentially a scavenger (necrophagous) and suggest that the 2 species are not in direct competition for their food supply.

KEY WORDS: Hydrothermal vent shrimp · Fatty acids · Nutrition · Trophic ecology Stable carbon isotope · Δ12 desaturase (n-4) PUFA

INTRODUCTION

The deep-sea ecosystem localised around the hydrothermal vent sites on the mid-Atlantic ridge (MAR) is considered to be predominantly based on carbon fixed by chemoautolithotrophic bacteria (Van Dover 1995). However, although numerous studies have indicated that the bulk of productivity at the vent sites is of bacterial origin, the quantitative significance and contribution of photosynthetically derived material for the nutrition of these communities has not been established (Rieley et al. 1995).

Caridean shrimp belonging to the family Alvinocarididae are the dominant megafauna at MAR vent sites (Segonzac 1992, Van Dover 1995) and *Rimicaris exoculata* is particularly abundant, forming characteristic, dense feeding swarms around hydrothermal chimneys and vents (Rona 1986, Gebruk et al. 1993). A further species, *Alvinocaris markensis*, is generally less abundant and tends to be located away from the vent source

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(Segonzac et al. 1993). *R. exoculata* supports a rich bacterial epibiosis on its gills and within its gill chambers (Casanova et al. 1993) and its mouthparts are hypertrophied and densely covered with setae, which substantially increases the surface area for attachment and growth of bacteria (Gebruk et al. 1993), or is an adaptation for bacterial feeding (Segonzac 1992). Van Dover et al. (1988) suggested that the primary source of nutrition for *R. exoculata* was bacteria living on the surface of chimney sulphides, but that epibiotic bacteria may also contribute to its diet. More recently Gebruk et al. (1993) argued that the levels of epibiotic bacteria on *R. exoculata* are sufficient to meet its entire dietary requirements and that the shrimps maintain close proximity to hydrothermal fluid to 'farm' their bacterial symbionts.

In contrast, *Alvinocaris markensis* does not appear to support epibiotic bacteria, nor does it actively swim on the periphery of the vent plumes (Casanova et al. 1993). Observations of the morphology and *in situ* behaviour of these 2 species led Segonzac et al. (1993) to speculate that they occupy different ecological niches, i.e. *Rimicaris exoculata* was considered to be a primary consumer, whilst *A. markensis* was considered exclusively necrophagous.

Fatty acid biomarker compounds have previously been utilised to provide useful insights into the trophic interactions between marine consumers and their food supply (Sargent et al. 1987, Fullarton et al. 1995, Pranal et al. 1996, St. John & Lund 1996). More recently, conventional gas chromatography has been coupled with isotope ratio mass spectrometry (GC-IRMS) to determine the $^{13}C/^{12}C$ isotopic composition of individual fatty acids (Fang et al. 1993a, Abrajano et al. 1994). The isotopic signature of compounds within an organism results from the combined effects of dietary sources and metabolic isotopic fractionations and can therefore provide useful insights into marine food webs (Hayes 1993).

In this study we analysed the fatty acid compositions of the vent shrimp *Rimicaris exoculata* and *Alvinocaris markensis* to improve our understanding of their sources of nutrition and trophic ecology.

METHODS

Station locations and description. Adult *Rimicaris exoculata* were obtained from collections made during the NERC-BRIDGE funded Anglo-Russian cruise (BRAVEX) to the TAG vent site (26°N, MAR), in the summer of 1994, whilst *Alvinocaris markensis* and a further specimen of *R. exoculata* were obtained from a collection made from the adjacent Snake Pit site (23°N) during the French HYDROSNAKE cruise in 1988. All specimens were deep frozen (-70°C) shortly after collection and, after dissection, the tissues were transported to the laboratory either in BLB buffer (5% sodium dodecylsulphate, 250 mM EDTA, 50 mM Tris-HCl, pH 8) or chloroform:methanol 2:1. Parallel DNA studies showed the tissue samples to be in an excellent state of preservation, based on the recovery of high-molecular-weight genomic DNA with virtually no evidence of enzymatic degradation (Dixon et al. unpubl.).

Fatty acid analysis. Tissue samples were homogenised in chloroform:methanol 2:1 (v/v) before filtering through a prewashed (chloroform:methanol 2:1 v/v) Whatman No. 1 paper filter. Total lipid was then extracted following Folch et al. (1957) and dried under nitrogen before transmethylation in absolute methanol containing 1.5% (v/v) sulphuric acid for 16 h at 50°C (Christie 1982). After extraction and purification, component fatty acids were identified by chromatography on a Canberra 436 GC fitted with a BP20 fused silica capillary column (50 m × 0.32 mm i.d., SGE) using hydrogen as carrier gas (Henderson et al. 1994). Peaks were identified by reference to samples of known composition and by GC-MS using a Fisons MD 800 fitted with a DB-5MS column (15 m × 0.25 mm i.d., J & W Scientific) using helium as carrier gas. The running order of fatty acids is as shown in Table 1.

Diene characterisation. To reduce co-elution and improve GC-MS, the total fatty acid methyl esters were separated into saturated, monounsaturated and polyunsaturated fatty acid fractions by argentation high-performance thin-layer chromatography (HPTLC) using hexane:diethyl ether (90:10 v/v) (Wilson & Sargent 1992). Fatty acid diethylamides were then prepared by the method of Nilsson & Liljenberg (1991) and identified by GC-MS operated in EI negative mode. Diethylamide derivatives of the (n-4) dienes and nonmethylene interrupted dienes (NMIDs) were characterised by GC-MS as follows: 16:2(n-4) m/z [M^+] 307 and 26 m/z gaps at m/z 264 to 238 and 224 to 198 indicated Δ12 and Δ9 double bonds; 18:2(n-4) m/z [M^+] 335, and 26 m/z gaps at m/z 292 to 260 and 252 to 226 indicated Δ14 and Δ11 double bonds; 20:2(n-4) m/z [M^+] 363, and 26 m/z gaps at m/z 320 to 294 and 280 to 254 indicated Δ16 and Δ13 double bonds; 20:2Δ5,13 m/z [M^+] 363 and 26 m/z gaps at m/z 278 to 252 and 168 to 142 indicated Δ13 and Δ5 double bonds: 22:2Δ7,15 m/z [M^+] 391, and 26 m/z gaps at m/z 306 to 280 and 196 to 170 indicated Δ7 and Δ15 double bonds.

$^{13}C/^{12}C$ analyses. Carbon stable isotope ratios ($^{13}C/^{12}C$) were measured by GC-IRMS using a VG Isochrom II instrument whose performance has been described by Eakin et al. (1992). A fused silica capillary column (SGE: BP20, 50 m × 0.32 mm, i.d. 0.5 µm) was used with helium as the carrier gas and a splitless injection mode was employed. The temperature pro-

gram was: 50°C (0.1 min) rising to 190°C with a 40°C min^{-1} gradient, then rising to 230°C (50 min) with a 2.5°C min^{-1} gradient. The dienoic fatty acids samples were injected in duplicate and compared with a reference CO_2 source which was calibrated for $\delta^{13}C$ (‰) by conventional dual-inlet mass spectrometry with reference to Peedee Belemnite (PDB). Replicate analyses suggest a precision of ±0.4‰. The $\delta^{13}C$ composition of the methanol derivatization reagent was determined as −41.8‰ by the quartz closed tube combustion technique of Sofer (1980). The contribution of the derivatized carbon was calculated by rearranging the equation of Abrajano at al. (1994) and assumes that there is no isotopic fractionation during derivatization.

$$\delta^{13}C_{FA} = \frac{\delta^{13}C_{FAME} - (1-x)\delta^{13}C_{CH_3OH}}{x}$$

where $\delta^{13}C_{FA}$ is the isotopic composition of the free fatty acid, $\delta^{13}C_{FAME}$ is the isotopic composition of the fatty acid methyl ester, x is the fractional carbon contribution of the free fatty acid to the ester and $\delta^{13}C_{CH_3OH}$ is the isotopic composition of the methanol derivatization reagent.

RESULTS

Bulk fatty acid compositions of total lipid extracted from *Rimicaris exoculata* collected from the TAG site and *Alvinocaris markensis* from the Snake Pit site were essentially similar, comprising approximately 36 to 40% saturated fatty acids (SFA), 14 to 20% monounsaturated fatty acids (MUFA) and 40 to 50% polyunsaturated fatty acids (PUFA) (Table 1). The fatty acid composition of a single specimen of *R. exoculata* from the Snake Pit also fitted this pattern (Table 1). However, detailed fatty acid analyses indicated substantial differences between the 2 species.

As the MUFA 18:1(n-7) is generally an abundant constituent of bacterial lipids, the ratio of 18:1(n-9)/18:1(n-7) can give a useful indication of the contribu-

Table 1. *Alvinocaris markensis*, *Rimicaris exoculata*. Fatty acid composition of total lipid from various tissues of vent shrimp from the Snake Pit and TAG hydrothermal vent sites. (Only mean values are presented for clarity; tr = <0.1%)

Fatty acid	Snake Pit			TAG			
	A. markensis		*R. exoculata*	*R. exoculata*			
	Digestive gland (n = 3)	Tail (n = 3)	Digestive gland (n = 1)	Digestive gland (n = 3)	Tail (n = 3)	Carapace (n = 3)	Gills (n = 3)
14:0	1.7	0.8	5.2	4.7	0.7	2.1	2.3
14:1	1.2	2.0	1.4	tr	tr	2.0	3.3
15:0	0.5	0.2	0.1	0.1	0.2	0.1	0.2
16:0	13.3	9.4	6.4	7.2	9.7	7.2	9.4
16:1(n-7)	9.0	5.5	22.3	19.7	11.1	19.0	19.4
16:2(n-3)	0.8	0.4	0.8	0.6	1.6	0.2	0.2
16:2(n-4)	0.4	1.6	10.7	14.5	2.0	7.0	9.4
17:0	0.5	0.3	0.2	0.3	1.8	0.2	0.2
16:3(n-3)	0.5	0.3	0.3	0.2	0.4	0.8	0.5
16:4(n-3)	tr	tr	tr	tr	tr	1.9	1.2
18:0	3.8	3.5	1.4	1.5	3.7	1.9	1.9
18:1(n-9)	16.8	12.5	3.6	2.5	8.4	6.6	4.9
18:1(n-7)	12.5	18.0	14.3	12.8	16.6	13.0	11.9
18:2(n-6)	1.5	1.0	tr	0.1	0.5	0.4	0.5
18:2(n-4)	1.5	1.4	19.9	23.8	13.0	15.9	17.5
18:3(n-3)	0.6	0.5	0.1	0.1	0.3	1.3	1.2
18:4(n-3)	1.1	1.0	0.1	tr	0.3	0.3	tr
20:0	0.2	0.2	0.1	0.1	0.4	2.1	0.2
20:1(n-9)	1.5	1.0	0.7	0.6	1.1	0.5	0.7
20:2 NMID	0.2	0.2	1.3	1.1	0.3	1.8	1.4
20:2(n-4)	tr	tr	tr	tr	tr	tr	tr
20:4(n-6)	2.5	3.6	0.9	1.0	2.6	2.7	1.9
20:4(n-3)	0.6	0.7	0.1	tr	0.1	tr	tr
20:5(n-3)	7.9	16.2	2.6	2.9	9.7	6.1	4.7
22:0	0.2	0.4	0.4	0.2	0.8	0.2	tr
22:1(n-9)	0.4	0.6	0.3	0.3	0.8	0.7	0.5
22:2 NMID	0.7	0.5	1.5	1.2	1.3	1.3	1.1
22:2(n-4)	tr	tr	tr	tr	tr	tr	tr
22:5(n-3)	1.4	0.6	tr	tr	0.5	0.2	0.3
22:6(n-3)	18.1	17.1	4.8	3.9	11.4	3.9	4.5
24:1(n-9)	0.6	0.5	0.5	0.4	0.7	0.5	0.7

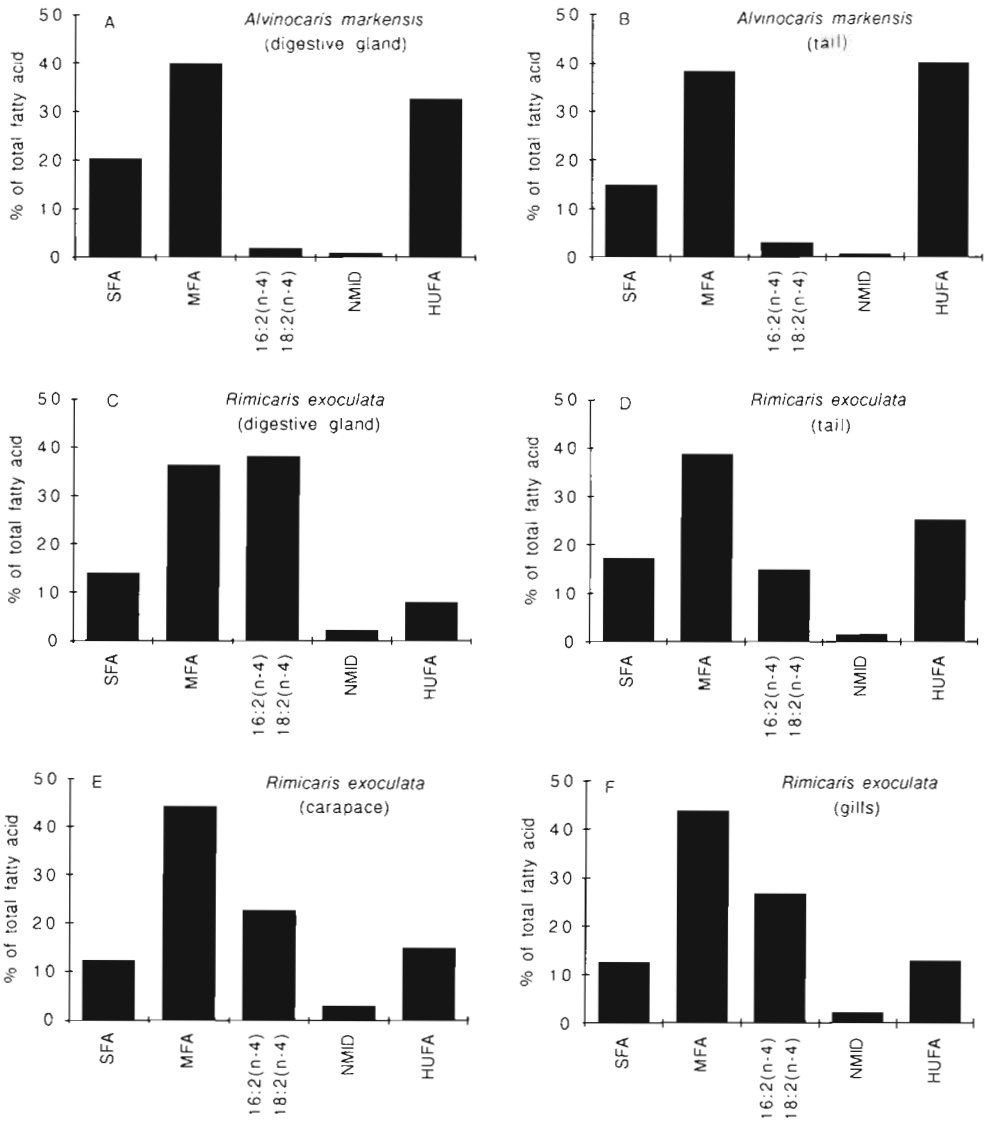

Fig. 1. Proportions of different groups of fatty acids from various tissues in *Alvinocaris markensis* and *Rimicaris exoculata* sampled from the Snake pit and TAG vent sites respectively. SFA = saturated fatty acid, MFA = monounsaturated fatty acid, NMID = non-methylene interrupted diunsaturated fatty acid, HUFA = highly unsaturated fatty acids, i.e. 3 or more double bonds

tion of bacteria to the nutrition of marine organisms (Sargent et al. 1987). The lowest ratio for these fatty acids was in the digestive gland of *Rimicaris exoculata* (0.2) whilst abdominal muscle, carapace and gills gave values of ca 0.4 to 0.5. However, the much larger amounts of 18:1(n-9) in *Alvinocaris markensis* gave a much higher ratio of 18:1(n-9)/18:1(n-7), with values of 0.7 and 1.3 for abdominal muscle and digestive gland respectively. These results suggest a higher dietary input of bacteria for *R. exoculata* than *A. markensis*. The MUFA 16:1(n-7), which is also abundant in bacteria, was also a major fatty acid in the shrimps and particularly the digestive gland, carapace and gills of *R. exoculata* where epibiotic bacteria were likely to contribute to the analyses. 16:1(n-7) was detected in lower amounts in *A. markensis* and comprised only 9.0 and 5.5% of total fatty acid for digestive gland and abdominal respectively.

Rimicaris exoculata contained high levels of the diunsaturated fatty acids 16:2(n-4) and 18:2(n-4) (Fig. 1, Table 1). The digestive gland was particularly rich in these compounds where they comprised 14.5 and 23.8% of total fatty acids respectively. The fatty acid composition of the single specimen of *R. exoculata* from the Snake Pit site was very similar to those from the TAG site and also contained high levels of (n-4) fatty acids. Other tissues in *R. exoculata* contained substantial proportions of these fatty acids, i.e. in rank order of compositional abun-

dance: gills > carapace > abdominal muscle (Table 1). Although (n-4) fatty acids were detected in *Alvinocaris markensis*, they comprised a much smaller proportion of the fatty acid pool and, when combined, only constituted 1.9 and 3.0% for digestive gland and abdominal muscle respectively. GC-IRMS analysis of these dienoic fatty acids from the gills of *R. exoculata* indicated that the $\delta^{13}C$ compositions were similar, i.e. −11.0 and −11.4‰ for 16:2(n-4) and 18:2(n-4) respectively (Table 2). By contrast, the fatty acids which are characteristic of phototrophic microplankton were isotopically lighter, i.e. −17.1, −17.3 and −15.8‰ for 20:4(n-6), 20:5(n-3) and 22:6(n-3) respectively (Table 2).

The NMIDs 20:2Δ5,13 and 22:2Δ7,15 were detected in both *Rimicaris exoculata* and *Alvinocaris markensis*, although they were only relatively minor components of their fatty acid pools (Fig. 1, Table 1). NMIDs were most abundant in *R. exoculata* and when combined, comprised totals of 1.6, 2.3, 2.5 and 3.1% of fatty acids in the abdominal muscle, digestive gland, gills and carapace respectively. In *A. markensis* these fatty acids were present in lower proportions and did not exceed 0.9% of total fatty acid (Fig. 1, Table 1).

The PUFA 20:5(n-3) and 22:6(n-3) were particularly abundant in *Alvinocaris markensis* (Fig. 1, Table 1). The abdominal and digestive gland of *A. markensis* both contained similar proportions of 22:6(n-3), i.e. 17.1 and 18.1%, whilst the abdominal muscle contained approximately twice the levels of 20:5(n-3) as the digestive gland (16.2 and 7.9% respectively). Substantially smaller amounts of these PUFA were present in *Rimicaris exoculata*, particularly the digestive gland, carapace and gills, where they each comprised only 2.9 to 6.1% of total fatty acids. However, abdominal muscle contained relatively high amounts of 20:5(n-3) and 22:6(n-3) (9.7% and 11.4% respectively).

Table 2. *Rimicaris exoculata*. Carbon stable isotope measurements of various tissues from the TAG vent site

Reference	Tissue	$\delta^{13}C$ (‰)
Van Dover et al. (1988) (total carbon)	Abdominal muscle	−11.6 to −12.1
Gebruk et al. (1993) (total carbon)	Whole animals	−10.5 to −12.1
	Mouthparts	−16.0
	Abdominal muscle + gills	−13.1
	Abdominal muscle + mouthparts	−15.1
Present study[a]	Gills	
14:0		−12.7
16:0		−13.9
16:1(n-7)		−13.1
16:2(n-4)		−11.0
18:2(n-4)		−11.4
20:4(n-6)		−17.1
20:5(n-3)		−17.3
22:6(n-3)		−15.8

[a]Determined by means of duplicate injections

DISCUSSION

Trophic ecology

The striking differences between the fatty acid compositions of *Rimicaris exoculata* and *Alvinocaris markensis* support published observations on the trophic ecology of these 2 species, which suggested that they occupy different ecological niches in the vicinity of hydrothermal vents (Segonzac et al. 1993). *R. exoculata* tends to swarm around the venting chimneys and is thought to derive nutrition both from bacterial production associated with the surface of the vent mounds and the ectosymbiotic filamentous bacteria which proliferate on the animal's hard surfaces, particularly within the enlarged gill chambers which are characteristic of this species (Van Dover et al. 1988, Gebruk et al. 1993). The relatively low proportions of (n-3) PUFA coupled with the high levels of monounsaturated fatty acids and the low 18:1(n-9)/18:1(n-7) ratio confirms that bacteria are a major source of nutrition for *R. exoculata*. By contrast, it has been proposed by Segonzac et al. (1993) that *A. markensis* adopts a more necrophagous, scavenging mode of feeding (which is borne out by the the more typical shrimp-like feeding appendages and the more massive chelae). A higher ratio of 18:1(n-9)/18:1(n-7) and the substantial levels of (n-3) PUFA indicates that the diet of *A. markensis* is relatively rich in such compounds and this is consistent with a necrophagous lifestyle in which feeding on animal tissue may predominate, but the possibility of a phytodetrital contribution in its diet cannot be excluded. The larvae of vent shrimp are also known to adopt a typically bathypelagic lifestyle (Dixon & Dixon 1996, Herring 1996, Pond et al. 1997) and also contain high levels of (n-3) PUFA. As these larvae travel considerable distances from the vent sites, they presumably must rely on phytodetritus for a least at portion of their dietary requirements (Pond et al. 1997).

Dienoic fatty acid synthesis

A notable finding in the present investigation was the high abundances of 16:2Δ9,12 and 18:2Δ11,14 fatty acids in the lipids of *Rimicaris exoculata* where

they comprised up to 38.3% of total fatty acid. 16:2(n-4) is readily synthesised by some species of marine photosynthetic algae which possess active Δ9 and Δ12 fatty acid desaturases. Diatoms generally contain 16:2(n-4) (Chuecas & Riley 1969, Volkman et al. 1989), although its fatty acids are not elongated further to 18:2(n-4) in these algae, or in animals that ingest it. Material from the euphotic zone can sediment rapidly to the deep ocean (Lampitt 1985) and scanning electron microscopy of particulate matter from the neutrally buoyant plumes of hydrothermal vents, including TAG, has indicated the presence of biological material originating in the euphotic zone (Dixon et al. 1995). However, in the typically oligotrophic mid-Atlantic waters in the vicinity of the TAG and Snake Pit vent sites, it is unlikely that substantial amounts of phytodetritus are sedimenting to the ocean floor, and it is more probable that the (n-4) fatty acids are synthesised by organisms within the vent ecosystem.

We suggest that the 16:2(n-4) detected here is synthesised by a Δ12 desaturase acting on 16:1(n-7), a fatty acid which is generally abundant in bacteria. Until relatively recently it was considered that bacteria are incapable of synthesising PUFA as they lack the necessary fatty acid desaturases. However, it is now firmly established that some deep-sea bacteria can be prolific producers of 20:5(n-3) and 22:6(n-3) PUFA (De Long & Yayanos 1986), as can some bacterial strains isolated from fish intestines (Yazawa et al. 1988). We found the highest levels of (n-4) dienoic fatty acids, both 16:2(n-4) and 18:2(n-4), in the digestive gland of *Rimicaris exoculata* and this could be associated with the abundances of bacteria in this organ which serves, amongst other things, a digestive function. Van Dover et al. (1988) noted high bacterial densities in the gut of *R. exoculata* (10^9 cells ml^{-1}) although they did not observe any morphological characteristics of the digestive system which suggested the presence of endosymbiotic bacteria. However, *R. exoculata* appears to ingest substantial amounts of metallic sulphide crystals which could potentially be utilised by gut bacteria as an energy source. It is possible that these gut bacteria could contribute to the animal's diet, not in a bulk energy sense, but by the provision of essential dietary components. Jacq et al. (1989) detected small amounts C18 and C20 PUFA in a strain of filamentous bacteria (*Thiothrix* sp.) sampled from a coastal hydrothermal vent, which is similar to the bacteria found at deep-sea vents. It is also significant that *Thiothrix* sp. can be a nutritious food source and supports high growth rates in subtidal gastropods at coastal hydrothermal vents (Stein 1984).

Δ12 desaturases have not previously been detected in marine animals, and it is therefore very unlikely that the vent shrimp itself forms 16:2(n-4). It is notable that photosynthetic prokaryotes, e.g. *Synechococcus* sp., readily produce 18:2(n-6) as they contain a Δ12 desaturase (Goodloe & Light 1982). However, as far as we know, a Δ12 desaturase has not been found in a chemoautotrophic prokaryote. The supposition that 16:2(n-4) and 18:2(n-4) are derived from bacteria is supported by the stable isotope analyses of the 16:2(n-4) and 18:2(n-4) fatty acids in *Rimicaris exoculata*. Previous carbon stable isotope measurements of total carbon from *R. exoculata* vary from −10.5 to −16.0‰ which suggests that the majority of carbon in this shrimp is of chemosynthetic origin (Van Dover et al. 1988, Gebruk et al. 1993; Table 2). As bulk carbon isotopic compositions of phytoplanktonic material from the shallow ocean are typically in the range −17 to −28‰ (Goericke et al. 1994) and given that the (n-4) dienoic acids here had isotopic values of −11.0 to −11.4‰, this supports the notion that these fatty acids are synthesised within the vent ecosystem and that they are of bacterial origin. By contrast, 20:4(n-6), 20:5(n-3) and 22:6(n-3) were comparatively isotopically light (−17.1, −17.3 and −15.8‰ respectively) which is consistent with a photic origin for these fatty acids.

If it is indeed true that 16:2(n-4) is of bacterial origin, what then is its functional/physiological significance? De Long & Yayanos (1985, 1986) have established that the fatty acid composition of deep-sea bacteria responded to increasing pressure by synthesising greater amounts of unsaturated fatty acids in order to optimise membrane fluidity and function. It is therefore plausible that the bacteria associated with the Snake Pit and TAG hydrothermal vent sites synthesise 16:2(n-4) as an adaptation to the temperature extremes and high hydrostatic pressures which are a characteristic of this bathypelagic environment.

Converting 16:2(n-4) to 18:2(n-4) requires an elongase which is an abundant enzyme in all marine organisms. It is therefore plausible that *Rimicaris exoculata*, having derived 16:2(n-4) from its diet, then elongates it to 18:2(n-4). It is well established that C16 PUFA are generally very minor components of animal cell membranes, possibly because they are of insufficient length for correct membrane functioning. *Alvinocaris markensis* also contains these fatty acids but in much lower levels, and as this species is not directly a bacterial feeder, it is consistent with the (n-4) fatty acids being originally of bacterial origin. The low levels of (n-4) fatty acids in *A. markensis* probably originate from necrophagy on other vent organisms essentially lacking these fatty acids and suggest that dead *R. exoculata* are not a major component of its diet. However, the possibility that these fatty acids are efficiently catabolised by *A. markensis* cannot be excluded.

The NMIDs 20:2Δ5,13 and 22:2Δ7,15 detected in the vent shrimp are thought to be derived from bacterial 18:1(n-7) by chain elongation to 20:1(n-7) and desaturation by a Δ5 desaturase to produce 20:2Δ5,13, followed by further chain elongation to 22:2Δ7,15 (Ackman & Hooper 1973, Zhukova 1991, Fullarton et al. 1995). Animals which synthesise NMIDs typically have diets which are rich in 16:0, 16:1(n-7) and 18:1(n-7) with a relative deficiency of PUFA, i.e. typically bacterial. We detected higher proportions of NMIDs in *Rimicaris exoculata* than *Alvinocaris markensis* which is consistent with *R. exoculata* deriving a greater proportion of its diet from bacterial sources. However, even in *R. exoculata* the amounts of these NMIDs did not exceed 1 to 2% of total fatty acids, which is considerably lower than the value of 15 to 20% reported for the littoral symbiotic bivalve *Lucinoma borealis* (Fullarton et al. 1995) and 12.2 to 17.7% for the hydrocarbon seep mussel (Fang et al. 1993b). It is probable that a high dietary input of (n-4) PUFA precludes the necessity of *R. exoculata* to synthesise NMIDs from 18:1(n-7).

It is possible that the differences in fatty acid composition between the 2 species could result from the different vent sites from which the animals were collected, i.e. *Rimicaris exoculata* was from the TAG and *Alvinocaris markensis* from the Snake Pit site. However, the *R. exoculata* from the Snake Pit site, although only a single specimen, did contain high levels of (n-4) fatty acids which suggests that the observed differences between the 2 shrimp species result from their trophic ecology rather than differences in nutritional environment.

In summary, fatty acids analyses of adult *Rimicaris exoculata* and *Alvinocaris markensis* have clearly indicated that these species occupy different ecological niches in the MAR vent environment, which confirms the observations of Segonzac et al. (1993). There is strong evidence to suggest that the filamentous chemoautotrophic bacteria associated with the vent ecosystem contain a Δ12 desaturase. This supposition, if correct, may have considerable implications for our understanding of the nutrition of vent communities in particular and deep sea organisms in general.

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