

A methane seep area in the Skagerrak, the habitat of the pogonophore *Siboglinum poseidoni* and the bivalve mollusc *Thyasira sarsi*

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ABSTRACT: On the southern slope of the central Skagerrak there are locally abundant populations of the pogonophore *Siboglinum poseidoni*, which lives on symbiotic methanotrophic bacteria, and the bivalve *Thyasira sarsi*, which lives on symbiotic sulphur-oxidising bacteria. Investigations have been made on the environmental factors influencing these populations, notably the methane content of the sediment, the rate of sulphate reduction and the distribution of reduced sulphur species. The sub-surface gas distribution was investigated using a deep-tow boomer and sediment cores were collected from both seep and non-seep areas. Methane-saturated sediment, detected by acoustic blanking, was found within 3 m of the sediment surface in water depths from <55 m to 360 m. At greater water depths the blanking was less shallow, sharply declining to 100 ms 2-way travel time at 390 m. There was no evidence for outgassing over most of the survey area. Seeps and their specialised fauna were found on a steeper section of the slope where slumping of the sediment exposed underlying gas-saturated layers. The seeps were small and localised and were estimated to occupy 2.3% of the area of this zone. In seep sediments the potential aerobic methane oxidation rates were 40-fold higher than in non-seep sediments, while the actual rates, measured by a radiotracer technique, were up to 460-fold higher in the surface sediment at the seeps. Within the seep sediments there was a greatly reduced sulphate concentration, down to $6 \times 10^3 \mu\text{g-at. S l}^{-1}$, in the upper 10 cm, compared with $30 \times 10^3 \mu\text{g-at. S l}^{-1}$ at a distance of 30 cm from the seep. Correspondingly, rates of sulphate reduction were very much greater at the seeps, 500 to 2000 $\mu\text{g-at. S reduced dm}^{-3} \text{d}^{-1}$, compared with approximately 50 $\mu\text{g-at. S reduced dm}^{-3} \text{d}^{-1}$ in non-seep sediment. Very high methane concentrations were found where *S. poseidoni* was abundant, up to $3.4 \times 10^3 \mu\text{mol dm}^{-3}$ in the upper 10 cm of the sediment. Dissolved sulphide was also much enriched at the seeps, to 500 $\mu\text{g-at. S l}^{-1}$ pore water, explaining the abundant populations of *T. sarsi*. These 2 species made up the bulk of the infaunal biomass at the seeps, whereas polychaetes dominated in non-seep sediments. The biomass of the macrofauna was 3-fold higher in seep than in non-seep sediment.

KEY WORDS: Methane seeps · Skagerrak · Sulphide · *Siboglinum poseidoni* · *Thyasira sarsi* · Sulphate reduction · Methane oxidation

INTRODUCTION

The southern part of the Skagerrak, from the Danish coast to the midline, is an area which has been subject to heavy sedimentation and contains a large area of gas-saturated sediment (van Weering et al. 1973, van Weering 1975, Hovland 1991). *Siboglinum poseidoni*, a

perviate pogonophore which lives on endosymbiotic methane-oxidising bacteria, has been described from this area (Southward et al. 1981, Flügel & Langhof 1983, Schmaljohann & Flügel 1987) and has recently been reported to be located at small methane seeps (Flügel & Callsen-Cencic 1992). With the exception of some vent and seep mytilids (Cavanaugh et al. 1987,

Childress et al. 1986, Cavanaugh et al. 1992), these pogonophores are the only animals currently known to derive most of their nutrition from methane. They have tissue carbon heavily depleted in ^{13}C , with $\delta^{13}\text{C} = -73.6$ to -78.3% (Schmaljohann et al. 1990). Associated with *S. poseidoni* is the bivalve mollusc *Thyasira sarsi* (Schmaljohann et al. 1990), a species normally living on endosymbiotic sulphur-oxidising bacteria (Dando & Southward 1986, Dando & Spiro 1993) and which has been shown to be associated with methane seep sites in the North Sea (Dando et al. 1991).

The objectives of this study were to investigate more fully the habitat of these animals in the Skagerrak and to define the conditions which give rise to this ecosystem.

METHODS

The area surveyed and sampled on RRS 'Challenger' cruise 82, from 20 to 29 July 1991, is shown in Fig. 1. It included the position $58^{\circ} 1.3' \text{ N}$, $9^{\circ} 34.6' \text{ E}$, in which the pogonophores had previously been located (Flügel & Langhof 1983). Acoustic surveys were carried out using a Simrad EA500 echo sounder with 11 and 12.5 kHz transducers and the British Geological Survey's (BGS) deep-tow boomer. The track plots for the 53 survey lines are shown in Fig. 1.

Sediment samples were collected in a $0.5 \times 0.5 \text{ m}$ box corer. PVC tubes of 100 mm i.d. were used to take subcores from the box for sediment chemistry. Subsamples

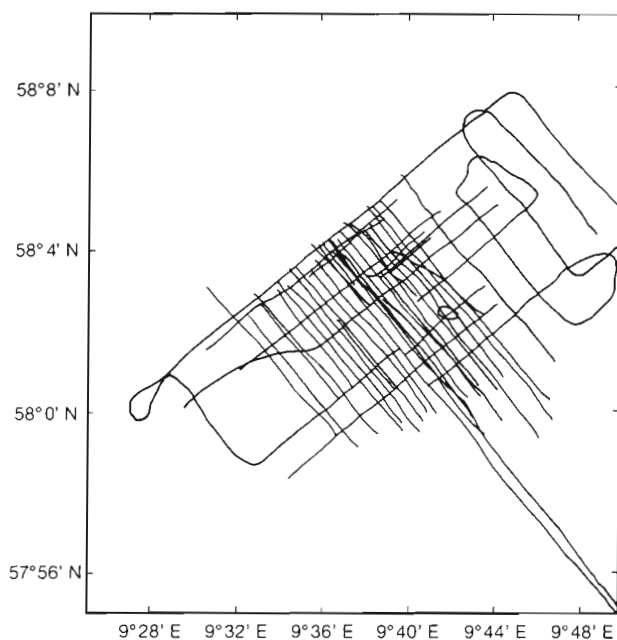


Fig. 1. Survey lines on 'Challenger' cruise 82 in the Skagerrak

were removed at 2, 5 or 10 cm intervals down the depth of the core using cut-off polystyrene syringes. After the subcores had been positioned, the side of the box was removed to allow examination of the sediment structure and to enable samples for macrofaunal determination to be removed from different horizons. Sediment subsamples were also taken from different horizons after sediment lying near to the removed side had been scraped away. Additional sediment was collected using a multiple corer to examine undisturbed sediment surface and a Kasten corer to look at deeper horizons. Sediment temperatures were in the range 6.3 to 6.6°C .

Sediment subsampling and analytical methods for gases, iron sulphides, elemental sulphur, sulphate and sulphate reduction rates followed those previously described (Dando et al. 1991). Dissolved sulphide, sulphite and thiosulphate were derivatised with monobromobimane under argon and the derivatives separated by HPLC and measured by fluorescence (Newton et al. 1981).

Sealed 4 ml syringe cores were injected with $5 \mu\text{l}$ of $\text{K}_2^{35}\text{SO}_4$ along the centre of the core and incubated in the dark for 12 to 15 h at 6°C . At the end of the incubation the sediment was extruded into glass vials containing 4 ml 5% neutralised zinc acetate solution. This precipitated any hydrogen sulphide and stopped further bacterial activity. The samples were then frozen until analysis when reduced sulphur species were all converted to hydrogen sulphide by reaction with acidified chromous chloride solution (Zhabina & Volkov 1978). The H_2S released was trapped in zinc acetate solution, mixed with scintillant (Packard Instagel) and the amount of $^{35}\text{SO}_4^{=}$ reduced determined by scintillation counting, using the external standard channel ratios method for quench correction. The radioactivity of the $^{35}\text{SO}_4^{=}$ remaining was determined by scintillation counting of sediment washings. Sulphate concentrations in the interstitial water were determined by ion chromatography and the water content of the sediment was measured by drying the sediment to constant weight at 60°C . Sulphate reduction rates were calculated by multiplying the proportion of the total ^{35}S reduced by the fractionation factor and by the dissolved sulphate dm^{-3} sediment (Jørgensen 1978).

Methane oxidation rates were determined by 2 methods. In near-surface layers the potential rate of aerobic oxidation was determined by incubating a sediment suspension under a methane/air gas phase which gave a final concentration in the aqueous phase of $1 \mu\text{M}$ methane. The consumption of methane was followed by gas chromatography.

The rate of methane oxidation under *in situ* oxygen and methane concentrations was determined by injection of ^{14}C -labelled methane (prepared from meth-

anogen cultures by U. Schuster, Univ. of Essex, UK) into intact sediment subsamples (Iversen & Jørgensen 1985) contained in 5 ml polystyrene syringes (Sabre) which were sealed with butyl rubber bungs. 20 to 50 μl of an anoxic solution of artificial seawater (30‰), containing 2 μl methane ml^{-1} , was injected through the rubber plunger seal of the syringe along the length of the sediment core. The syringe samples were then incubated at 6°C in the dark for 12 h. Incubation was stopped by pushing the sediment into gas-tight glass vials prefilled with 5 ml 2 N NaOH. This stopped the bacterial activity and trapped the ^{14}C -bicarbonate. The amounts of $^{14}\text{CO}_2$ and $^{14}\text{CH}_4$ in the vials were determined according to Iversen & Jørgensen (1985). Carbon dioxide was trapped after acidification of the sediment. Additionally, the amount of radioactivity fixed as organic carbon in the sediment was determined in triplicate by combusting the dried sediment with a Packard sample oxidiser and determining the activity of the CO_2 released by scintillation counting. Radioactivity in the different fractions was estimated using a Beckman LS scintillation counter with quench corrections based on external standards and channel ratios. Rates of methane oxidation in the sediment were calculated from the amount of $^{14}\text{CO}_2$ and $^{14}\text{C}_{\text{assimilated}}$ formed in the sediment and from the concentration and radioactivity of the methane. The rates of oxidation were shown to be linear with time in pilot studies up to 24 h and therefore only end-point determinations were applied to most samples.

Chemical concentrations and rate measurements are reported per dm^3 sediment, or per litre of extracted interstitial water.

Direct counts of bacterial numbers were made using epifluorescence microscopy of sonicated, diluted sediment samples stained with acridine orange. The fauna was collected by sieving the sediment through a mesh of 500 μm diameter, fixed in buffered 2% formalin in seawater and transferred to 75% ethanol.

RESULTS

The survey area covered water depths from 55 to 425 m; the bathymetry is plotted in Fig. 2. The deep-tow boomer records showed acoustic blanking, regions of sediment through which sound waves would not penetrate, over most of the area. From shallower than 55 m water depth (the shallowest part of the slope investigated) downslope to approximately 360 m the blanking started within the upper 3 m of the sedimentary column (Figs. 3 & 4). This blanking also showed as a strong reflector on the Simrad EA500 echo sounder records. The positions of sediment cores taken are

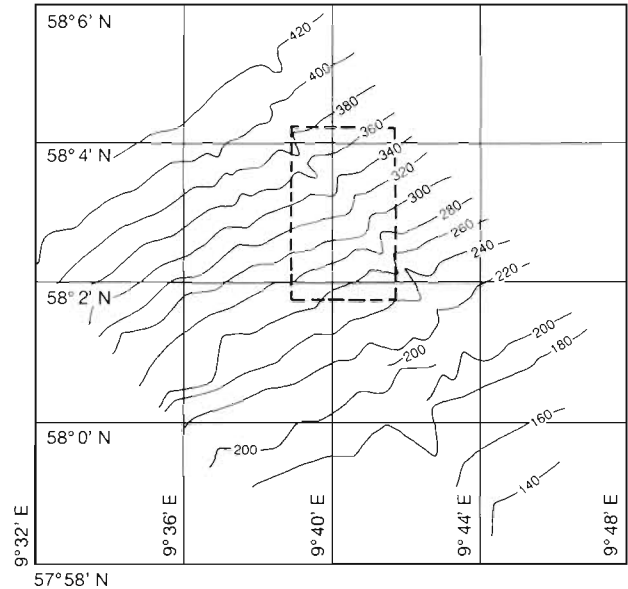


Fig. 2. Bathymetry of survey area. Rectangle shows the larger area (shown in Fig. 5) studied by means of sediment cores

shown in Fig. 5. Confirmation that the acoustic blanking was due to gas bubbles was obtained from 4 Kasten core samples taken in positions where the acoustic blanking approached the sediment surface, Stns 45, 94, 112 and 114 (Fig. 5) in water depths of 321, 305, 358 and 366 m respectively. Gas bubbles were observed in these cores at sub-bottom depths of, respectively, 1.55, 0.83, 1.35 and 0.62 m. In addition box cores at Stns 61, 71 and 105 (Fig. 5) showed gas bubbles venting from part of the core surface and the core at Stn 106 showed a dome of sulphide-rich and methane-rich mud rising from part of the base of the core to within 20 cm of the sediment surface.

The sediment depth at which acoustic blanking occurred increased to 20 ms at 360 m water depth and then sharply dropped below the depth of acoustic penetration by the boomer, 100 ms 2-way travel time, at approximately 390 m water depth (Fig. 3). North of this deeper boundary a few 'plumes' of gas-charged sediment were observed (Figs. 4 & 6).

An area of hummocks and sediment slumps (Figs. 3 & 4) was associated with a slight increase in the slope of the seabed to $1.61 \pm 0.18^\circ$, starting at approximately 280 m water depth and continuing to the region where the depth of sediment gas saturation, as determined by acoustic blanking on the boomer records, fell below 20 ms at 360 m water depth. Sediment core sampling was concentrated in this zone of sediment slumps. Within this region 44 box core samples were collected, only 3 of which had high methane concentrations in the surface sediment.

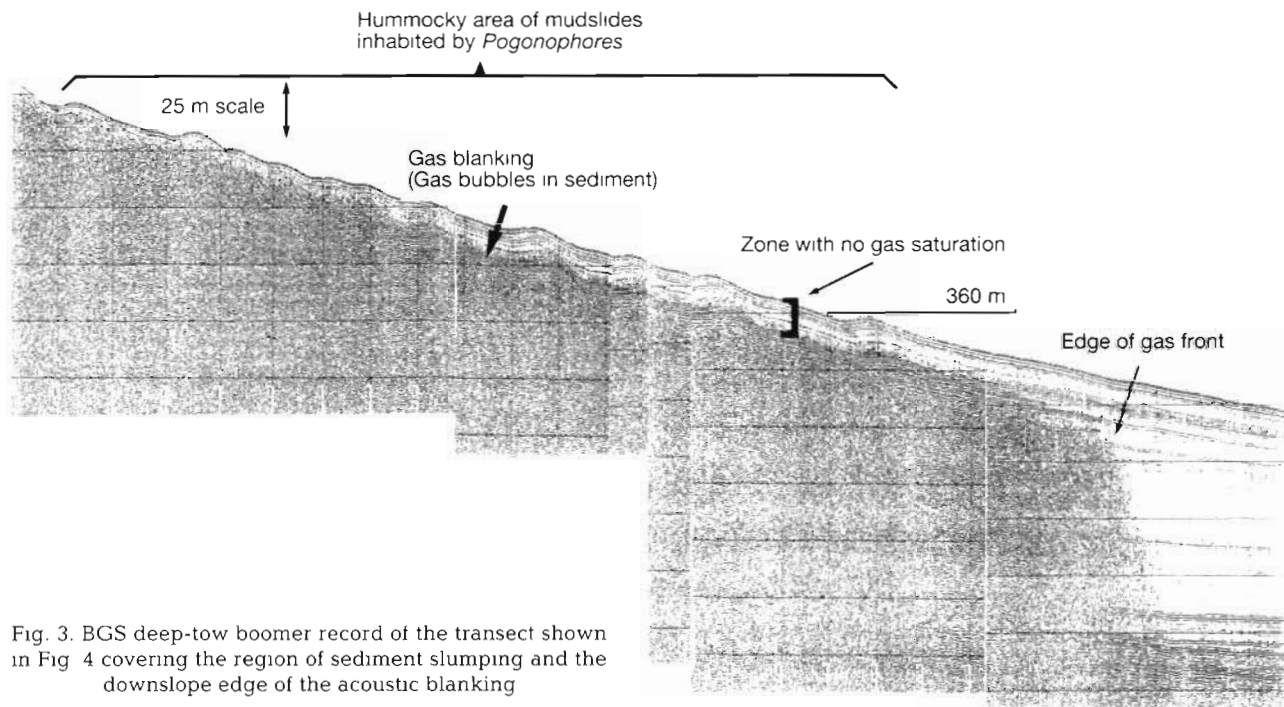


Fig. 3. BGS deep-tow boomer record of the transect shown in Fig. 4 covering the region of sediment slumping and the downslope edge of the acoustic blanking

Geochemical profiles were determined on 14 cores. Typical sediment profiles for methane, methane oxidation rates, sulphate, sulphate reduction, soluble and insoluble sulphur species, ammonia, pH and water content are shown for a core from Stn 35 at 323 m water depth (Fig. 7). In most cores sampled the

methane concentrations in the upper 10 cm of sediment were $< 1 \mu\text{mol dm}^{-3}$ sediment. The depth profiles indicate that, in most cases, methane is consumed within the upper 10 cm of the sediment and does not diffuse into the overlying seawater. Below approximately 10 cm sediment depth the methane concentrations increased with depth until visible bubble formation occurred at concentrations of approximately $3 \times 10^3 \mu\text{mol dm}^{-3}$ sediment. Aerobic methane oxidation rates in sediment samples exposed to air were normally below $2.4 \mu\text{mol dm}^{-3} \text{d}^{-1}$ in the upper 10 cm of sediment, with an integrated rate of 17.5 ± 4.7 (SD) $\mu\text{mol m}^{-2} \text{d}^{-1}$ for the upper 8 cm of sediment. The tracer injection method gave oxidation rates of $1.6 \mu\text{mol dm}^{-3} \text{d}^{-1}$ at the surface, decreasing with depth to at least 42 cm, e.g. the box core at Stn 81 (Fig. 8). Deeper in the sediment rates increased again, for example in the methane-saturated sediment from the Kasten core at Stn 45 samples from 140 to 320 cm depth sub-bottom showed a mean oxidation rate of $12.0 \mu\text{mol dm}^{-3} \text{d}^{-1}$ (Fig. 9a).

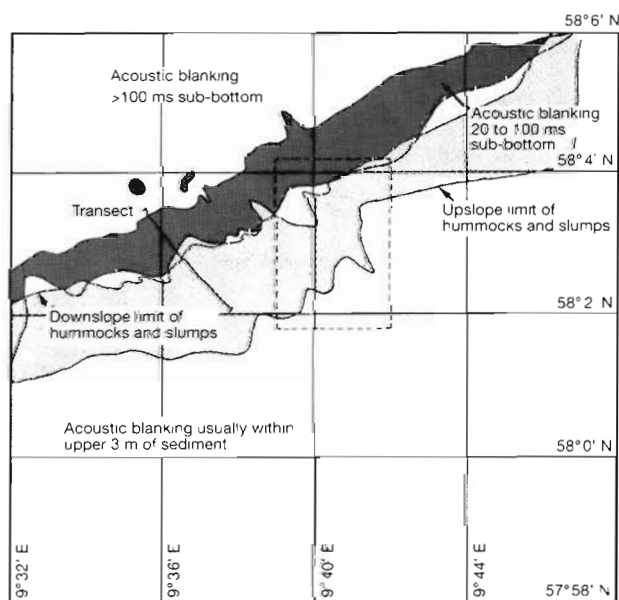


Fig. 4 Map of the survey area showing region of shallow gas-saturated sediment and area of sediment hummocks and slumps. The rectangle shows the larger area (shown in Fig. 5) studied by means of sediment cores

The sulphate concentrations in the interstitial water of most cores did not fall below $20 \times 10^3 \mu\text{g-at. S l}^{-1}$ in the upper 50 cm of the sediment (Figs. 7b & 8b). Integrated sulphate reduction rates for the upper 40 cm of sediment were in the range 4.5×10^3 to 15.1×10^3 , mean $7.2 \times 10^3 \pm 3.1 \times 10^3$ (SD) $\mu\text{g-at. sulphate S reduced m}^{-2} \text{d}^{-1}$. Acid-labile sulphide concentrations and chromous-reducible sulphide concentrations in the sediment were high, both reaching up to $48 \times 10^3 \mu\text{g-at. S dm}^{-3}$ in the upper 50 cm of sediment. Sulphide

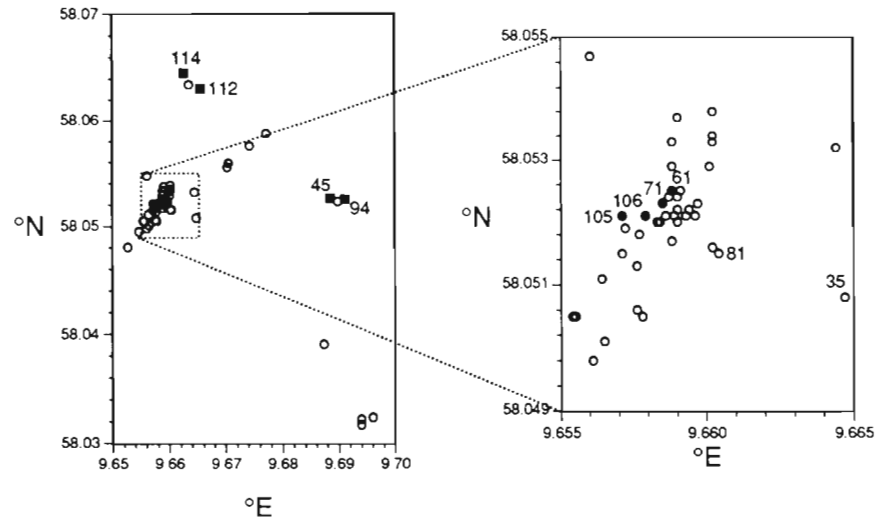


Fig. 5. Positions of sediment cores taken. Expanded section shows the detail of core positions in the seep area; co-ordinates are shown as decimal degrees. The position of Kastenlot cores (■) and box cores containing seep sediment (●) or non-seep sediment (○) are marked

oxidation rates appeared to be high with very low concentrations of dissolved sulphide in most cores, $\leq 3 \mu\text{g-at. S l}^{-1}$, and high elemental sulphur concentrations, up to $7 \times 10^3 \mu\text{g-at. S dm}^{-3}$ sediment. Sulphate reduction rates increased with sediment depth until approximately 10 cm depth, reaching a maximum rate in most cores of 46 to 53 $\mu\text{g-at. sulphate S reduced dm}^{-3} \text{ d}^{-1}$, and then remained at a rate of 25 to 50 $\mu\text{g-at. sulphate S reduced dm}^{-3} \text{ d}^{-1}$ until at least 50 cm depth. These rates increased below 1 m sediment depth, in the 1 Kasten core studied for sulphate reduction rates these were 64 to 1054 $\mu\text{g-at. sulphate S reduced dm}^{-3} \text{ d}^{-1}$ in the region of low sulphate concentration and methane saturation (Fig. 9b).

In most cores, e.g. Fig. 7, the pH of the interstitial water ranged between 7.20 and 7.65 in the upper 2 cm of sediment to between 7.49 and 8.05 at 50 cm depth. Interstitial ammonia concentrations varied from between 26 and 500 μM in the upper 2 cm to between 300 and 1400 μM at 50 cm depth. The organic carbon content was variable but normally in the range 1.8 to 2.8% of dry weight in the upper 50 cm of sediment.

The major differences from the above results were in cores where methane bubbles were being vented from the surface. One of these cores, from Stn 61 at 333 m water depth, was venting methane bubbles from approximately 8 outlets on one-half of the box core. Escaping gas had a strong hydrogen sulphide smell. The bubbles brought to the surface black, sulphide-rich mud which flowed over the 1 mm thick light-brown oxidised sediment surface. None of the bubbles had formed a depression in the sediment surface, as was described by Flügel & Callsen-Cencic

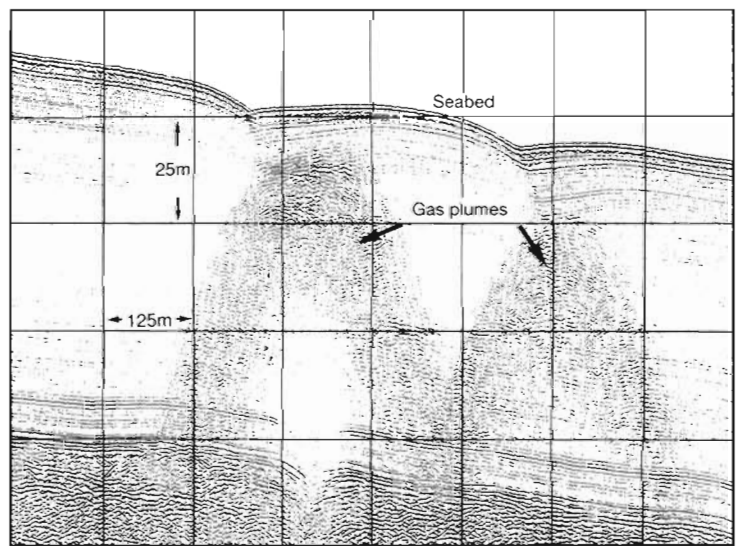


Fig. 6. 'Plumes' of acoustic blanking observed at 380 m water depth

(1992). A fine brown mat of tubes of the pogonophore *Siboglinum poseidoni* was exposed at the surface on the seeping half of the box, but none were visible on the non-seeping half. Methane concentrations were in the range 0.7×10^3 to $3.4 \times 10^3 \mu\text{mol dm}^{-3}$ in the upper 10 cm of sediment from the seeping part of the core but were $< 10 \mu\text{mol dm}^{-3}$ in the corresponding horizon in the non-seeping part of the core (Fig. 10a). A transect away from a gas outlet showed that the methane concentration in 5 ml sediment syringe subcores, taken from the sediment surface, decreased from $2.16 \times 10^3 \mu\text{mol dm}^{-3}$ to $42 \mu\text{mol dm}^{-3}$ within 5 cm (Fig. 11). Methane oxidation rates, measured by the radiotracer method, similarly decreased from 740 to $8 \mu\text{mol dm}^{-3} \text{ d}^{-1}$ over the same distance (Fig. 11).

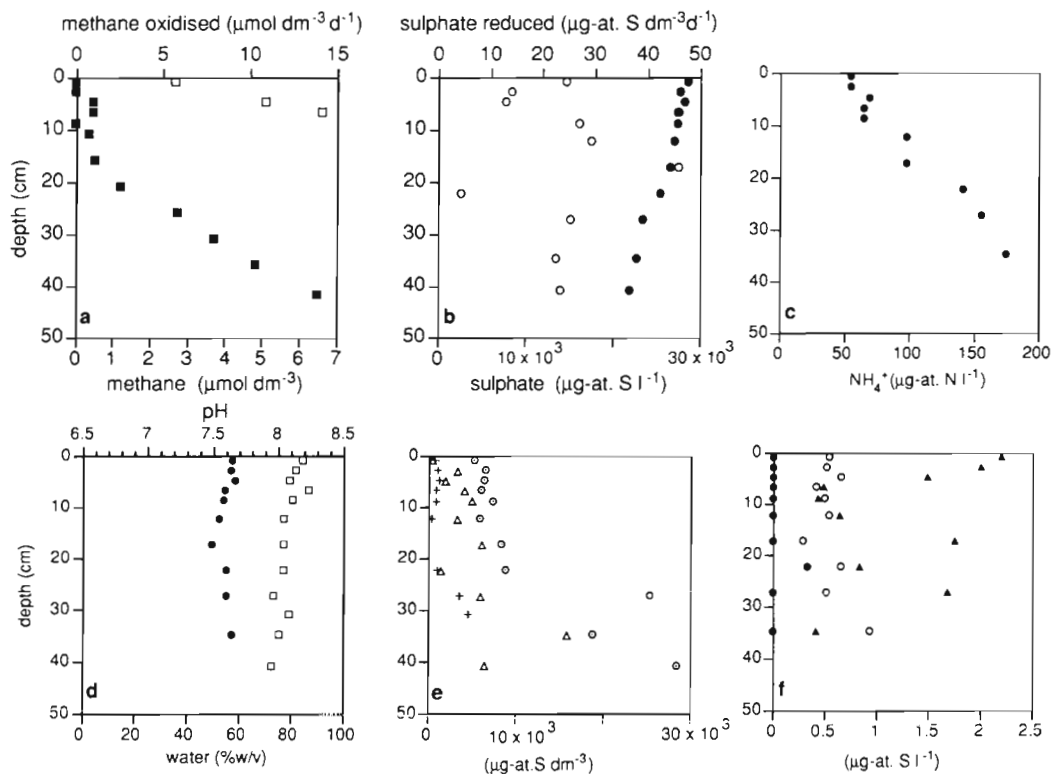


Fig. 7. Sediment profiles at Stn 35 (Fig. 5) for (a) methane (■) and aerobic methane oxidation rate (□); (b) sulphate (●) and sulphate reduction rate (○); (c) NH_4^+ ; (d) pH (●) and water content (□); (e) S^0 (+), acid-labile sulphide (Δ) and chromous reducible sulphide (○); (f) sulphite (●), thiosulphate (○) and sulphide (\blacktriangle) in pore water

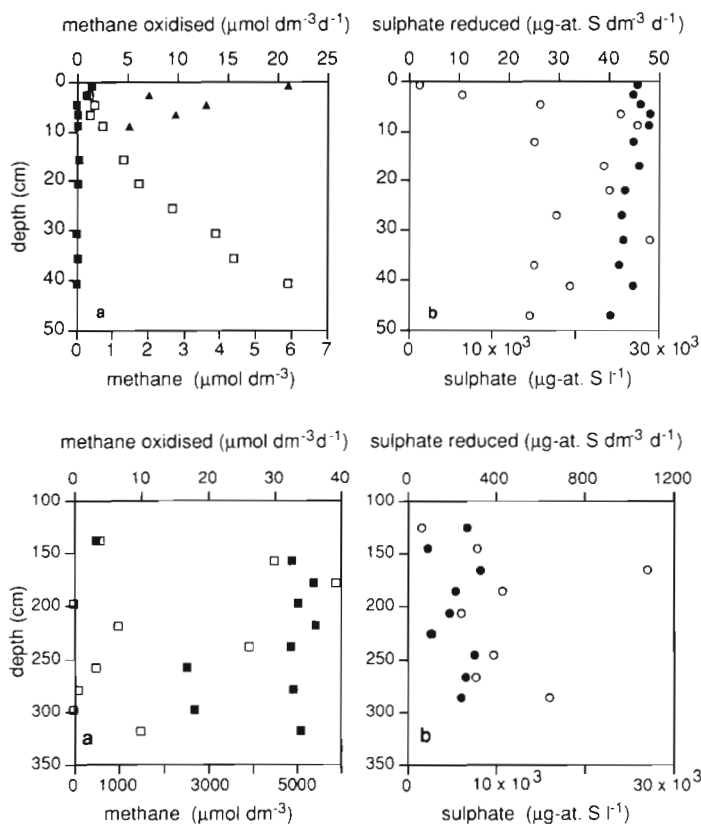


Fig. 8. Profiles at Stn 81 for (a) methane (□), aerobic methane oxidation rate (\blacktriangle) and the methane oxidation rate using the radiotracer method (■), (b) sulphate (●) and sulphate reduction rate (○)

Fig. 9. Sediment profiles in a Kasten core taken at Stn 45 for (a) methane (■) and methane oxidation rate using the radiotracer method (□), (b) sulphate (●) and sulphate reduction rate (○)

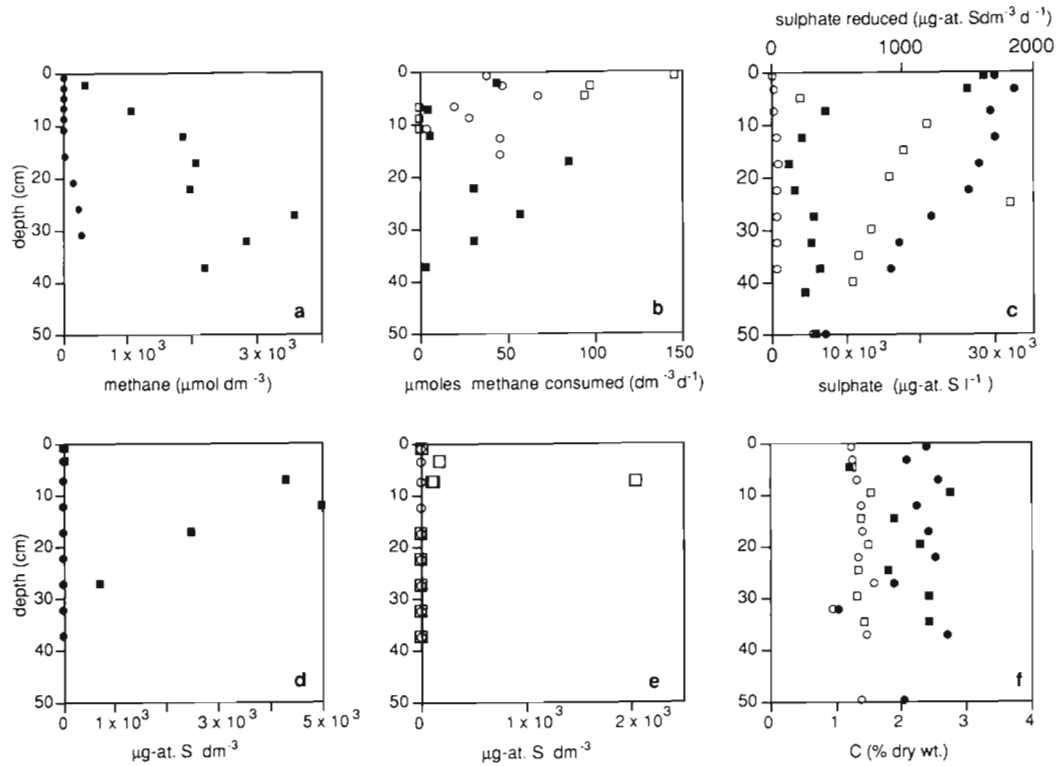


Fig. 10. Sediment profiles in 2 subcores taken 30 cm apart from the seep box core at Stn 61. Profiles from the seeping part of the core are shown by squares and profiles from the non-seeping part are shown by circles: (a) methane (●, ■); (b) aerobic methane oxidation rate (○, □) and methane oxidation using the radiotracer method (■); (c) sulphate (●, ■) and sulphate reduction rate (○, □); (d) dissolved sulphide (●, ■); (e) thiosulphate (○, □); (f) organic (●, ■) and inorganic carbon (○, □)

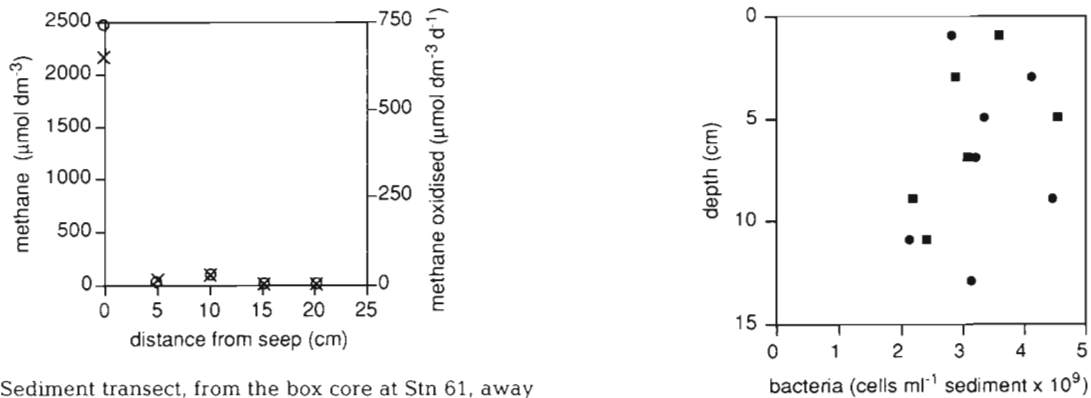


Fig. 11. Sediment transect, from the box core at Stn 61, away from a gas outlet showing methane (○) and methane oxidation rate by the radiotracer method (×) in the upper 5 cm of sediment

Fig. 12. Sediment profile of bacterial numbers in the seep (■) and non-seep (●) parts of the box core from Stn 61

Total non-volatile organic carbon contents were similar for both seep and non-seep sections (Fig. 10f) as were total bacterial counts (Fig. 12). Aerobic methane oxidation rates in sediment suspensions from the seep were high (94 to 146 $\mu\text{mol dm}^{-3} \text{d}^{-1}$) in the upper 6 cm of sediment in the seep area, whereas no activity was found below (Fig. 10b). In the non-seeping part of the core aerobic methane oxidation was lower (4 to

67 $\mu\text{mol dm}^{-3} \text{d}^{-1}$), but was present to at least 16 cm sediment depth. Measurements of the methane oxidation rate by the tracer injection method showed a surface maximum close to the seep (Fig. 11) and a sub-surface maximum at approximately 15 cm depth reaching 85 $\mu\text{mol dm}^{-3} \text{d}^{-1}$ (Fig. 10b). Turnover times, calculated on the integrated values of methane concentration and oxidation rates over the depth sampled,

Table 1. Species composition of seep and non-seep areas at Stn 61

Species	Seep part of core		Non-seep part of core	
	no. m ⁻²	g m ⁻²	no. m ⁻²	g m ⁻²
<i>Paramphinone jeffreysii</i>	2767		926	
Other polychaetes	299		574	
Total polychaete biomass		9.2		29.2
<i>Siboglinum poseidoni</i>	17600	76.6	50	0.2
<i>Abra nitida</i>	567		303	
<i>Thyasira sarsi</i>	133		0	
<i>Thyasira equalis</i>	300		750	
<i>Thyasira eumyaria</i>	0		83	
<i>Nucula</i> sp.	33		0	
Total bivalve biomass		36.5		9.3
<i>Amphiura</i> sp.	33	2.8	0	
Total biomass		125.1		38.7

ranged between 42 d in a non-seep core (Stn 81) to 6 d in a seep core (Stn 105). Sediment from Stn 61 had a turnover time for methane of 63 d.

The differences in dissolved sulphate profiles from subcores taken 30 cm apart from the seeping and non-gas seeping parts of the core were dramatic (Fig. 10c). Sulphate remained above $27 \times 10^3 \mu\text{g-at. S l}^{-1}$ in the upper 20 cm of the non-seep part and then decreased, almost linearly, to $7 \times 10^3 \mu\text{g-at. S l}^{-1}$ at 50 cm sediment depth. In the seep part of the core sulphate rapidly decreased from the surface to $6 \times 10^3 \mu\text{g-at. S l}^{-1}$ at 10 cm sediment depth and then remained at approximately this concentration for the rest of the core. Sulphate reduction rates, below 10 cm sediment depth, were in the range 0.5×10^3 to $2 \times 10^3 \mu\text{g-at. S reduced dm}^{-3} \text{ d}^{-1}$; at this rate, in the absence of re-oxidation of the sulphide or a replacement of the interstitial water, the sulphate would become exhausted within 3 to 6 d. The sulphate reduction rates for the seep part of the core at Stn 61 were underestimated due to the fact that up to 70% of the added $\text{K}_2^{35}\text{SO}_4$ was reduced within the 12 h incubation period, thus sulphate depletion became significant during the incubation period. The integrated rate for the seep part of the core was $147.8 \times 10^3 \mu\text{g-at. sulphate S reduced m}^{-2} \text{ d}^{-1}$ for the upper 40 cm of sediment, compared to $7.1 \times 10^3 \mu\text{g-at. sulphate S}$ for the non-seep part of the core.

Dissolved sulphide concentrations in the methane-saturated part of the core were in the range 0.5 to $500 \mu\text{g-at. S l}^{-1}$, compared to no detectable sulphide in the other half of the box core (Fig. 10d). Thiosulphate concentrations ranged up to $2.05 \times 10^3 \mu\text{g-at. S l}^{-1}$ in the seep half, compared to between 0.0 and $1.0 \mu\text{g-at. S l}^{-1}$ in the non-seep part of the box core (Fig. 10e).

The density of *Siboglinum poseidoni* was 17 600 ind. m⁻² in the half of the core from which gas was venting, compared to 50 m⁻² in the other half of the

core. These pogonophores made up 63% of the wet weight of animals in the seeping part of the core where the total biomass (122 g m^{-2}) was 3-fold higher than that in the non-seeping part of the core (biomass 38.7 g m^{-2}) (Table 1). The bivalve *Thyasira sarsi*, at a density of 133 ind. m⁻², was responsible for most of the remaining biomass in the seeping half of the core but was absent from the non-seeping section. In contrast the other major thyasirid species present, the smaller *T. equalis*, was more common in the non-seep half, 750 m⁻², than in the seep half, 300 m⁻². The in-fauna in the non-seep part of the core was dominated by polychaetes, which at almost 1500 m⁻² made up 75% of the biomass, with almost all the residue being due to the bivalves *T. equalis* and *Abra nitida*.

The box core collected from Stn 71 was similar to this, except that the zone of outgassing sediment with pogonophores extended only 8 cm into the 50 cm box (Fig. 13). In the core from Stn 105 the gas escaped only from the extreme corner, around the edges of a hagfish *Myxine glutinosa* burrow. A sub-core taken alongside the visible gas seep showed the presence of gas bubbles within the sediment at 25 cm below the surface. In the final seep core, Stn 106, the region of methane- and sulphide-rich sediment again only occurred in one corner and reached from the base of the 68 cm deep core to within 20 cm of the sediment surface. More detailed analysis of the fauna collected in the samples will be published elsewhere.

DISCUSSION

The Skagerrak is a sedimentary basin in which post-glacial deposits reach thicknesses up to 90 m (van Weering 1975). Within the study area the rate of sedi-

mentation has been calculated to be in the range 1 to 3 mm yr⁻¹ (van Weering et al. 1987). The resulting rapid carbon burial results in high levels of methane in the sediments. This was first recognised by van Weering et al. (1973) and van Weering (1975) who noted areas of acoustic blanking, 'shadow zones', due to gas bubbles in the sediment. The most recent map of shallow gas occurrence in the Skagerrak is that of Hovland (1991). A zone of acoustic turbidity is seen in the sediments along the southern part of the Skagerrak in water depths from 200 to 300 m and shallower. The area in water shallower than 90 m off the northern coast of Denmark had not been surveyed. The present study found that the zone of acoustic turbidity continues inshore to water depths of less than 50 m off Hirtshals. Within the greater part of this area methane bubbles are present to within 3 m of the sediment surface, as confirmed by core samples. The shallow gas zone is probably continuous with the one running in a NW-SE direction through the northern part of Vendsyssel (Jessen 1936).

Within the shallow gas area the 'seeps', where methane bubbles escape into the water column, are concentrated in the steeper part of the slope. In this region mounds and slumps occur, exposing the methane-saturated sediment horizons which are otherwise 0.5 m or more below the surface. Despite the presence of this shallow methane-saturated sediment there is little evidence of large-scale seepage into the water column. The methane concentration profiles indicate that most of the gas is consumed within the sediment (Figs. 7a & 8a). Within the zone of sediment slumps or hummocks, between 280 and 360 m depth, acoustically turbid plumes, probably due to gas bubbles, rising from the seabed, have been observed on 18 kHz echosounder records (H. Flügel & P. Callsen-Cencic pers. comm.). Flügel & Callsen-Cencic (1992) recently collected a box core from this area which contained numerous *Siboglinum poseidoni* and which vented methane bubbles when brought to the surface. We collected 3 similar venting cores, 2 containing pogonophores, among the 44 taken in this region, i.e. 7% of the total. The seep cores lay in an approximately NE-SW line parallel to the depth contours in this part of the Skagerrak. None of the 0.25 m² box cores was seeping methane from its entire area, suggesting that the areas of methane seepage are very small. On the basis of the area of seeping sediment collected in the cores taken we estimate that a maximum of 2.3% of the region of sediment slumps is seeping gas.

The seeping part of the cores was characterised not only by high methane concentrations in the upper

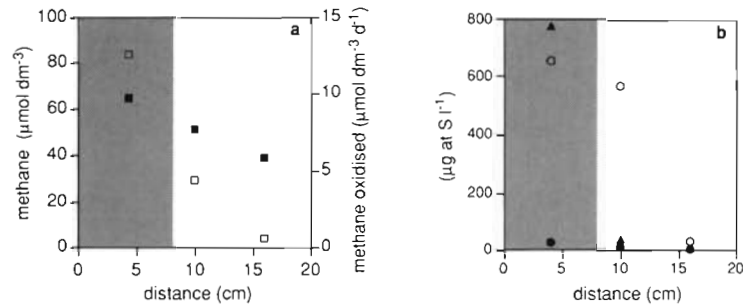


Fig. 13. Transect, at 35 cm sediment depth, across part of the box core from Stn 71. The shaded area shows the region of dark mud containing some pogonophores *Siboglinum poseidoni*. (a) Methane (■) and methane oxidation rate by the radiotracer method (□); (b) sulphite (●), thiosulphate (○) and sulphide (▲) in pore water

10 cm of sediment but also by high dissolved sulphide and by high sulphate reduction rates. This accounted for the co-distribution of both *Siboglinum poseidoni*, a gutless pogonophore which derives its nutrition from symbiotic methane-oxidising bacteria (Schmaljohann et al. 1990), and *Thyasira sarsi*, a bivalve which may obtain the majority of its carbon from symbiotic sulphur-oxidising bacteria in its gills (Dando & Southward 1986, Dando & Spiro 1993). *T. equalis*, the most common thyasirid bivalve in this part of the Skagerrak, was less numerous at the gas seeps. Although *T. equalis* does contain symbiotic sulphur-oxidising bacteria in its gills this species has fewer bacteria than *T. sarsi* (Southward 1986) and appears to be less dependent on the bacterial carbon for its nutrition (Dando & Spiro 1993).

The 2 methods used for measuring methane oxidation rates are not directly comparable. The suspension method, using identical oxygen and methane concentrations in samples from different sediment layers, allows a comparison of potential rates, corresponding to the distribution of aerobic methane oxidising bacteria. The tracer injection method gives rates close to the actual rates of methane oxidation in the sediment where rates depend on the number of methane-oxidising bacteria and the concentrations of methane and of suitable electron acceptors. It should be noted that these incubations were carried out at *in situ* temperatures but not at *in situ* pressures and therefore outgassing of methane in the seep sediments would have occurred. In general the suspension method showed higher rates in surface sediments, corresponding to the expected distribution of aerobic methanotrophs which would be depth limited by oxygen penetration. This in turn would be controlled by macrofaunal burrowing activity. In addition, close to sites of escaping gas bubbles, overlying water is drawn down into the sediment to replace the interstitial water 'pumped' out by the ris-

ing gas bubbles (Dando et al. 1994, O'Hara et al. 1994). The water draw-down close to gas seeps would allow oxygen to penetrate the sediment deeper than would otherwise be the case. This explains the presence of aerobic methane oxidation in deeper sediment horizons (≥ 16 cm) alongside the gas seep in the core from Stn 61 (Fig. 10b) and also accounts for the sharp reduction in methane concentrations in the sediment alongside the seeps since the rate of diffusion is lower than the water inflow to the gas channels (Dando et al. 1994, O'Hara et al. 1994).

The tracer injection method revealed a second (anaerobic) methane oxidation maximum in deeper horizons. This had previously been reported from the Skagerrak (Iversen & Jørgensen 1985). The maximum rate occurred at 15 cm, identical to that reported for sediment in Saanich Inlet, Canada (Devol et al. 1984).

The very high sulphate reduction rates, 0.5×10^3 to 1.0×10^3 $\mu\text{g-at. sulphate S reduced dm}^{-3}$ sediment d^{-1} , clearly associated with the gas seepage, were unexpected. They have not been observed at shallower methane seep sites in the North Sea and Kattegat (Dando et al. 1991, 1994, Jensen et al. 1992). The high rates in the Skagerrak were found both in the surface sediments of seeps, where methane was outgassing and the concentration of methane was $\geq 2 \times 10^3$ $\mu\text{mol dm}^{-3}$ sediment, and also in methane-saturated sediment in deeper sediment. However the rates are an order of magnitude higher than those observed for methane oxidation by the radiotracer method. It is possible that organic material fixed by the dense pogonophore/bacterial associations helps to fuel the sulphate reduction. The high sulphate reduction rates are believed to be sustained despite low sulphate concentrations due to the gas-flow induced interstitial water circulation within the sediment (Dando et al. 1994) which replenishes the sulphate.

The pogonophore *Siboglinum poseidoni* has a length of up to 19 cm (Flügel & Callsen-Cencic 1992). This suggests that its distribution will be limited to regions where substantial methane concentrations are found within the upper 20 cm of sediment. Since there is no mechanism for active uptake of methane, high concentrations would be needed for methanotrophy by bacteria contained within internal tissues. In the 2 cores in which *S. poseidoni* was found maximum methane concentrations in the upper 20 cm of the sediment were 280 to 3300 $\mu\text{mol dm}^{-3}$. Earlier reports of concentrations of as low as 1 to 44 $\mu\text{mol kg}^{-1}$ sediment (Schmaljohann & Flügel 1987, Schmaljohann et al. 1990) may have referred to sediment near to the seeps but in which pogonophores were not found. Over most of the survey area the zone of methane saturation was below 60 cm with < 1 $\mu\text{mol methane dm}^{-3}$ sediment in the upper 10 cm. This sharp

gradient, which is maintained by anaerobic methane oxidation, would prevent juvenile *S. poseidoni* from reaching the extensive underlying biogenic methane deposits. In the fine-grained Skagerrak sediments methane only reaches the upper layers in places where downslope slumping on the steeper part of the slope exposes the gas-saturated sediment or where bioturbation, such as that caused by the burrowing of the hagfish at Stn 105, provides a channel for the underlying gas to escape. This will also release hydrogen sulphide produced in the zone of anaerobic methane oxidation. Methanotrophic activity, both free living and symbiotic, should also provide an input of readily metabolisable organic matter into the sediment and hence increase the sulphate reduction rate. Hydrogen sulphide concentrations in the interstitial water of sediment samples collected away from the seeps were ≤ 3 μM . The high sulphide concentrations at the seeps will enhance the growth of the bivalve mollusc *Thyasira sarsi* which is largely dependent on endosymbiotic sulphur-oxidising bacteria for its nutrition.

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