

# Changes in the lipid composition of Antarctic sea-ice diatom communities during a spring bloom: an indication of community physiological status

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**Abstract:** The lipid composition of natural populations of diatoms in the sea ice at McMurdo Sound was determined during the austral spring bloom of 1985, using an Iatroscan TLC-FID system. The major lipid classes in all samples were polar lipids (including phospholipid, glycolipid and chlorophyll) and triacylglycerol, with lesser proportions of free fatty acids. Total lipid increased through November and early December, reaching a maximum (3300 mg m<sup>-2</sup> at Cape Armitage and 1800 mg m<sup>-2</sup> at Erebus Ice Tongue) *c.* one week after the chlorophyll *a* maxima. This increase was largely attributable to a corresponding increase in triacylglycerol. At the lipid maxima, triacylglycerol/polar lipid ratios in the range 1.0 to 2.5 were observed. The dynamic variations in lipid class abundances indicate that profound changes in the physiology of sea-ice diatoms are occurring throughout the spring bloom. A range of sterols (C<sub>26</sub>–C<sub>30</sub>) were detected; 24-methylenecholesterol, brassicasterol and 24-ethylcholesterol were the major sterols at the Cape Armitage and Erebus sites. The similarity of the sterol profiles to those of Antarctic freshwater algal communities strongly indicates diatoms as a more probable source of C<sub>29</sub> sterols in the freshwater lakes than cyanobacteria or other algal groups. The hydrocarbons isolated from sea-ice diatoms at all sites were dominated by two unsaturated components, *n*-C<sub>21:6</sub> and a diunsaturated isoprenoid C<sub>25</sub> alkene. Until this study, no biological source had been validated for the isoprenoid C<sub>25:2</sub> diene, even though it has been detected in many estuarine and coastal sediments.

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**Key words:** hydrocarbons, lipids, sterols.

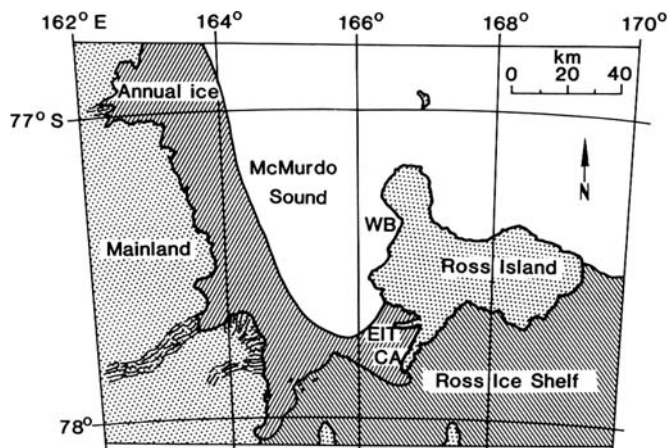
## Introduction

During the austral spring and summer at McMurdo Sound, Antarctica, microalgal communities composed almost exclusively of diatoms are found near the bottom of the hard congelation ice and in the underlying platelet ice when it is present (Bunt 1963, Palmisano & Sullivan 1983, 1985). As sea-ice diatoms are important components of the carbon and energy flux in the polar oceans, their development, physiology and ecology are being studied intensively.

Our group has examined changes in photosynthetic metabolism of sea-ice microalgae during a spring bloom at McMurdo Sound (Palmisano & Sullivan 1985, Palmisano *et al.* 1988). Smith & Morris (1980) showed that phytoplankton of the Southern Ocean, under low light and low temperature conditions, incorporated as much as 80% of the carbon fixed during photosynthesis into lipid. In contrast, our studies of Antarctic sea-ice communities and Li & Platt's (1982) studies of arctic phytoplankton, indicated that incorporation of <sup>14</sup>C into lipids was less than 30% of the total <sup>14</sup>C uptake. Assimilation of <sup>14</sup>C into specific lipid fractions (neutral,

glycolipid and phospholipid) changed dramatically, however, over the course of the bloom and its decline. The most dramatic changes in <sup>14</sup>C incorporation were found in the neutral lipid fraction (Palmisano *et al.* 1988). In a related study of the lipid distribution in the sea-ice algal communities, high neutral lipid content was observed in communities at two study sites shortly after the chlorophyll maximum (Nichols *et al.* 1988).

In this study, temporal analyses of the lipid composition of natural populations of sea-ice diatom communities were undertaken to determine physiological and biochemical changes occurring during a spring bloom of sea-ice microalgae. The distribution and significance of hydrocarbon and sterol profiles obtained for the sea-ice diatom communities are also presented. Few reports of the sterol and hydrocarbon distributions of Antarctic freshwater algal communities, including stromatolites, are available (Matsumoto *et al.* 1982, 1983, Orcutt *et al.* 1986, Volkman *et al.* 1986a). The data presented here provides new information on the possible origins of hydrocarbons and sterols in Antarctic sediments and food webs.



**Fig. 1.** McMurdo Sound, Antarctica. Sampling sites: WB = Wohlschlag Bay, EIT = Erebus Ice Tongue, CA = Cape Armitage. Cape Evans (CE) site located approximately 5 km north-east of EIT on Ross Island.

## Materials and methods

Sea-ice communities were sampled during December 1985 at three sites: Cape Armitage, the Erebus Ice Tongue and Wohlschlag Bay in McMurdo Sound, Antarctica (Fig. 1). The Cape Armitage site has been described in detail elsewhere (e.g. Grossi *et al.* 1984, Palmisano *et al.* 1985).

Congelation ice samples (2–3 cores) at each site were collected with a SIPRE ice auger (7-cm core diameter) from annual sea ice with thicknesses between 1.6 and 2.5 m. The lower 20-cm sections were cut from each core with a stainless-steel handsaw. Samples were wrapped in opaque black plastic and placed in Freezefast styrofoam containers for transport to the laboratory. It has previously been demonstrated that, in McMurdo Sound, 99% of the chlorophyll *a* is found in the bottom 20 cm of the congelation ice (Palmisano & Sullivan 1983). Each 20-cm section was melted at less than 5°C in 1.2 l of filtered sea-water.

Platelet ice samples were collected by SCUBA divers and were melted at less than 5°C without addition of sea-water. Freshly collected, unpreserved, sea-ice microalgae samples were examined by phase-contrast microscopy (Zeiss) to determine species composition. Sea-ice algal cells were prepared for lipid analysis by filtration onto glass-fibre filters (Whatman GF/C) that had been prewashed with  $\text{CHCl}_3$ -MeOH.

Samples were quantitatively extracted at the Eklund Biological Laboratory at McMurdo station with the modified one-phase  $\text{CHCl}_3$ -MeOH Bligh and Dyer extraction (White *et al.* 1979). After phase separation, the lipids were recovered in the lower  $\text{CHCl}_3$  layer (solvents were removed *in vacuo*)

and were stored sealed under nitrogen at  $-20^\circ\text{C}$ .

Subsequent lipid fractionation and analyses were performed at the CSIRO Marine Laboratories in Hobart. A portion of the total lipid extract was analysed with an Iatroscan MK III TH10 TLC-FID analyser (Iatron Laboratories, Japan) as described in detail elsewhere (Volkman *et al.* 1986b). Briefly, samples were applied to silica gel SII chromarods (5  $\mu\text{m}$  particle size) using 1- $\mu\text{l}$  disposable micropipettes (Drummond). Chromarods were developed in a glass tank lined with pre-extracted filter paper. The solvent system used for the lipid separation was hexane-diethyl ether-acetic acid (60/17/0.5; v/v/v). This solvent system was designed to clearly resolve triacylglycerols and free fatty acids as well as other common neutral lipid classes. Polar lipids, containing predominantly phospholipid, glycolipid and chlorophyll (Harwood & Russell 1984), remained at the origin and were not further separated. After development, the chromarods were oven dried for 10 min at  $100^\circ\text{C}$  and analysed immediately to minimize adsorption of atmospheric contaminants. The FID was calibrated for each compound class (0.1–20  $\mu\text{g}$  range). Total lipid content represents the sum of the individual lipid classes determined using the Iatroscan TLC-FID.

A second portion of the total lipid extract was separated into individual lipid classes by column chromatography on silicic acid (3 g) deactivated with 5% distilled water. Eight lipid fractions were obtained. The hydrocarbon fraction was eluted with 4 ml hexane and 7 ml hexane-toluene (1/1; v/v), and sterols were eluted with 15 ml hexane-ethylacetate (85/15; v/v).

Gas chromatographic (GC) analyses were performed with a Hewlett Packard 5890 GC equipped with a 50 m  $\times$  0.2 mm internal diameter cross-linked methyl silicone fused-silica capillary column and a flame ionization detector. Details are provided in Nichols *et al.* (1988). Lipid, hydrocarbon and sterol compositional data reported for these samples are the means of 2–3 replicate analyses. Hydrogenations were carried out by adding the hydrocarbons to a suspension of  $\text{PtO}_2$  (Adams catalyst) in purified iso-octane and bubbling  $\text{H}_2$  through the suspension for 3 h.

Gas chromatography-mass spectrometric (GC-MS) analyses of hydrocarbon and sterol (as OTMSi ethers) samples were performed on a Hewlett Packard (HP) 5890 GC and 5970 Mass Selective Detector (MSD) fitted with a direct capillary inlet. Operating conditions have been described in detail in Nichols *et al.* (1988). Identifications of individual sterol and hydrocarbon components were confirmed by comparing mass spectra with those of previously reported spectra (Brooks *et al.* 1968, de Leeuw *et al.* 1983, Albaiges *et al.* 1984, Rowland *et al.* 1985, Robson & Rowland 1986), and by comparing retention data (both before and after hydrogenation) with data for commercial and previously identified laboratory standards.

## Results and discussion

### Species composition

The composition of sea-ice algal communities of Cape Armitage, the Erebus Ice Tongue and Wohlschlag Bay have been previously reported (Nichols *et al.* 1988, Palmisano *et al.* 1988). The sea-ice diatom communities in mid-December were dominated by *Amphiprora* sp., *Nitzschia stellata* and *Berkeleya* sp. at Cape Armitage; *N. stellata*, *Amphipora*, *Pleurosigma*, *N. kerguelensis* and some small centrics at the Erebus Ice Tongue site; and *Porosira pseudodenticulata* at Wohlschlag Bay.

### Lipid-class distribution

Lipid compositional data obtained for congelation and platelet sea-ice diatom communities isolated from McMurdo Sound are presented in Table I and Fig. 2. Polar lipids (containing predominantly phospholipids, glycolipids and chlorophyll; Harwood & Russell (1984)) and triacylglycerols were the major lipid classes detected in all congelation ice diatom samples, with lesser amounts of free fatty acids, sterols and hydrocarbons also present. The Wohlschlag Bay community contained predominantly polar lipids, with only small amounts

of neutral lipid present (Table I); the difference is probably due to the markedly different species composition at this site.

Seasonal data were obtained for the congelation sea-ice communities from Cape Armitage and the site adjacent to the Erebus Ice Tongue (Table I, Figs 2, 3). Total lipid increased at the two sites through November and early December, peaking approximately one week after the observed chlorophyll *a* maxima. Total lipid abundance reached 3300 mg m<sup>-2</sup> at Cape Armitage and 1800 mg m<sup>-2</sup> at the Erebus Ice Tongue site. At both sites, the increase in total lipid was largely due to an increase in the energy-rich storage component, triacylglycerol. At the lipid maxima, triacylglycerol/polar lipid ratios of 1.0 (Erebus Ice Tongue) and 2.5 (Cape Armitage) were observed (Table I, Fig. 3). At the Erebus site, a second, smaller increase in the triacylglycerol/polar lipid ratio occurred at the end of the study period, whilst at Cape Armitage the amount of triacylglycerol declined steadily after the lipid maximum.

These data can be related to changes in the assimilation of <sup>14</sup>C into specific lipid fractions observed in these communities (Palmisano *et al.* 1988). Neutral lipid fractions accounted for c. 11% of the total carbon assimilated during late November–early December, followed by a decline in mid-December to 0.1% of carbon assimilated. During late December, assimilation into neutral lipids increased again, leveling off

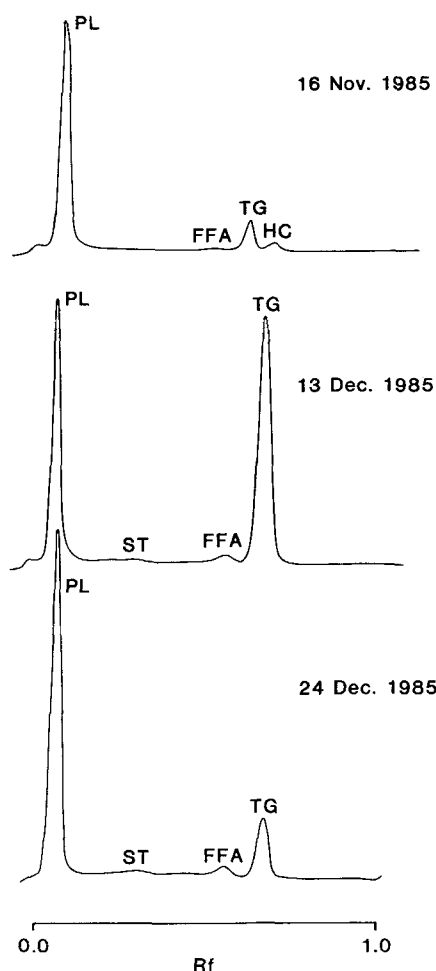
Table I. Lipid abundance and composition in sea-ice algal communities from McMurdo Sound, Antarctica.

Location	Date (1985)	Mean Chl. <i>a</i> (mg m <sup>-2</sup> ) <sup>a</sup>	Total lipid (mg m <sup>-2</sup> )	Lipid composition (%) <sup>b</sup>					Lipid/Chl. <i>a</i>	TG/PL
				HC	TG	FFA	ST	PL		
<b>A. Congelation ice</b>										
Cape Armitage (CA)	17/11	13.5 (7.7) <sup>c</sup>	290	7	4	6	3	80	21.5	0.05
	25/11	135 (42)	1810	1	43	9	1	46	13	0.93
	30/11	—	2350	1	48	9	3	40	—	1.2
	7/12	150 (69)	1730	1	33	16	4	46	11.5	0.72
	13/12	144 (28)	3330	1	68	3	1	27	23	2.5
	18/12	110 (42)	2577	2	52	9	2	35	23	1.5
	24/12	88 (10)	1610	1	34	4	1	61	18	0.55
Erebus Ice Tongue (EIT)	16/11	—	540	4	11	4	2	79	—	0.14
	25/11	83 (12)	870	2	25	4	1	68	10.5	0.37
	30/11	108 (12)	1200	1	29	6	1	62	11.1	0.47
	13/12	92 (21)	1590	1	43	3	1	53	17.3	0.81
	18/12	54 (18)	1830	2	44	6	2	46	33.1	0.96
	24/12	46 (31)	990	2	16	6	2	74	21.5	0.22
	28/12	15 (6)	1290	1	32	7	5	56	86	0.57
Cape Evans (CE)	24/11	—	6034	1	56	4	1	38	—	1.5
Wohlschlag Bay (WB)	12/12	—	1200	2	7	6	2	81	—	0.09
<b>B. Platelet ice</b>										
Cape Armitage	17/11	—	3140	2	5	2	1	90	—	0.06
Erebus Ice Tongue	16/11	—	6380	1	6	3	1	89	—	0.07
	7/12	—	9650	1	10	2	1	86	—	0.12

<sup>a</sup> Data from Palmisano *et al.* 1988

<sup>b</sup> Percentage composition of total lipids; abbreviations: HC, hydrocarbon; TG, triacylglycerols; FFA, free fatty acids; ST, sterols; PL, polar lipids

<sup>c</sup> Standard deviation



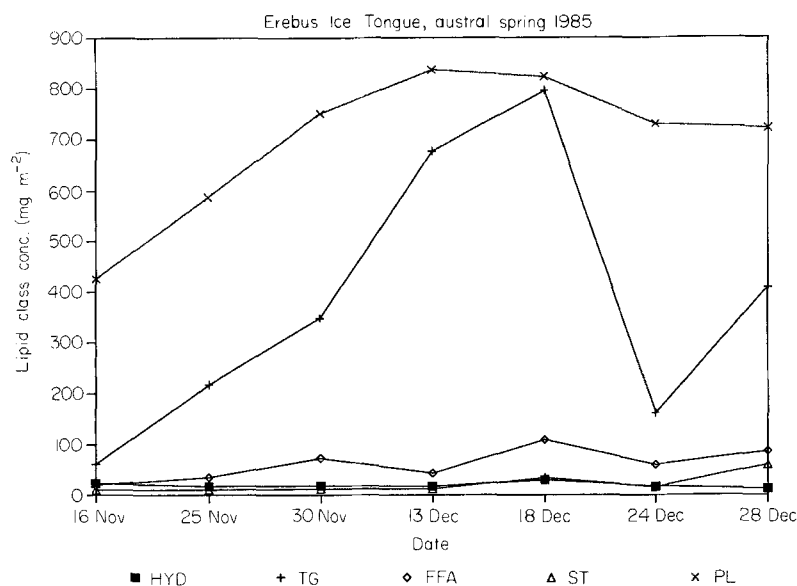
**Fig. 2.** Introscan TLC-FID chromatograms of total lipids from Erebus Ice Tongue sea-ice diatom communities. HC = hydrocarbons, TG = triacylglycerols, FFA = free fatty acids, ST = sterols, PL = polar lipid.

at *c.* 13% of the total carbon fixed. Carbon assimilation into glycolipid and phospholipid was relatively constant during the bloom. The initial period of neutral lipid synthesis coincided with increasing triacylglycerol concentration, and the decline in neutral lipid synthesis occurred when triacylglycerol reached its maximum value (Fig. 3, Table I). The final period of increased assimilation of  $^{14}\text{C}$  into neutral lipid is reflected by an increase in triacylglycerol at the Erebus Ice Tongue site, but not at Cape Armitage.

Changes observed in carbon assimilation patterns and resultant lipid distribution profiles may reflect changes in the physiology of the respective algal communities or variations in the species composition. However, concurrent microscopic studies suggest that changes in species composition at the Erebus Ice Tongue and Cape Armitage sites were relatively minor (Lizotte & Sullivan, unpublished data), and were believed to be unrelated to the profound changes observed in  $^{14}\text{C}$  assimilation and lipid distribution throughout the season. We consider, therefore, that the changes predominantly reflect differences in the physiology of the sea-ice diatom communities.

A number of comparisons can be made that provide further insight into the changing status of the Cape Armitage and Erebus communities. Growth conditions can influence algal physiological status. The most widely studied factor has been the influence of nitrogen; nitrogen limitation may result in increased lipid synthesis in unicellular algae (Borowitzka 1988). A sea-ice diatom community obtained from the Erebus Ice Tongue site and cultured in the laboratory under conditions of nitrogen limitation also showed high triacylglycerol content (triacylglycerol/polar lipid = 0.62).

Comparison of data obtained for platelet communities underlying the congelation sea-ice diatoms also provides additional information. Platelet communities analysed at



**Fig. 3.** Distribution of individual lipid classes in Erebus Ice Tongue sea-ice diatom communities during the austral spring, 16 November to 28 December 1985.



early stages of the congelation ice algal bloom (mid-November) showed lipid distribution profiles similar to those observed in the overlying congelation ice communities. However, the platelet community sampled at the Erebus site during the period of high  $^{14}\text{C}$  assimilation into neutral lipid and high triacylglycerol content in the congelation ice community, did not contain high levels of triacylglycerol. As the two community types (congelation and platelet ice) had a similar species composition, the differences observed in the lipid distribution of the two communities are probably the result of differences in algal growth conditions, and therefore physiological status, at the time of sampling. In early December, high algal densities occur in the congelation ice (chlorophyll *a* = 150 mg m<sup>-2</sup> at Cape Armitage and 100 mg m<sup>-2</sup> at the Erebus Ice Tongue site; Table I). Algae isolated in the minute brine pockets that occur in the congelation ice may, therefore, have depleted the available nutrients, and replenishment of nutrients from the nutrient-rich sea-water below the sea ice may have not been substantial enough to maintain the algal community. Grainger (1977) calculated ratios of P to N consumption for an arctic ice algal bloom, and suggested that nitrogen limitation was possible. The dynamic changes in lipid class abundances observed in this study in natural sea-ice diatom populations support this view, and provide a direct measure of the profound changes occurring in the physiological status of the communities.

In this study of Antarctic sea-ice algal communities, single communities were followed through bloom development and senescence, since interfering processes such as vertical mixing and horizontal advection are absent. Similarly,

losses from grazing and sinking were minor. These data, therefore, not only provide information on sea-ice algal communities, but also serve as a model for studies of physiological changes occurring during estuarine, coastal or open water algal blooms.

### Sterols

A wide range of C<sub>26</sub>–C<sub>30</sub> sterols were detected in the congelation sea-ice diatom communities (Table II, Fig. 4). At the Cape Armitage and Erebus Ice Tongue sites C<sub>28</sub> (mainly 24-methylenecholesterol and brassicasterol) and C<sub>29</sub> (mainly 24-ethylcholesterol) sterols were the major components. Brassicasterol and 24-methylenecholesterol were the dominant components at Cape Evans and Wohlschlag Bay, respectively. The similar profiles at the Cape Armitage, Erebus Ice Tongue and Cape Evans sites reflect the similarity of the species composition at these three sites. Differences in the relative proportions of individual sterols reflect the different proportions of the species. Likewise, the vastly different species composition of the Wohlschlag Bay sea-ice diatom community, compared with the other three sites, is reflected in the sterol profiles (Table II).

A number of interesting features were observed for the sterol profiles. 24-Ethylcholesterol is the main sterol present in the Erebus Ice Tongue sea-ice diatom community, and is the second most abundant sterol detected in the Cape Armitage and Cape Evans samples. This sterol has been commonly used as a marker for higher plants (Huang & Meinschein

Table II. Sterol composition of congelation sea-ice algal communities from McMurdo Sound, Antarctica.

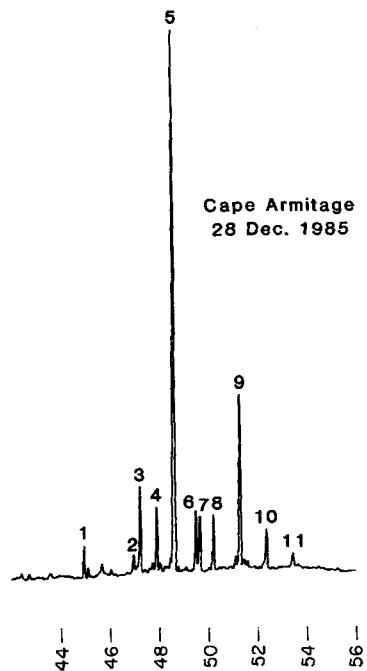
Peak number <sup>a</sup>	Sterol	Common name	EIT <sup>b</sup> 18 Dec	CA 18 Dec	CE 24 Nov	WB 12 Dec
1	24-norcholesta-5, 22-dien-3 $\beta$ -ol		0.2	1.8	3.1	—
2	27-nor-24-methylcholesta-5, 22E-dien-3 $\beta$ -ol		0.5	0.8	0.9	—
3	cholesta-5, 22E-dien-3 $\beta$ -ol	trans-22-de hydrocholesterol	3.8	5.4	3.7	1.2
4	cholest-5-en-3 $\beta$ -ol	cholesterol	5.3	4.9	6.6	4.2
5	24-methylcholesta-5, 22E-dien-3 $\beta$ -ol	brassicasterol <sup>c</sup>	15.6	42.0	57.5	1.4
6	24-methylcholesta-5, 24-dien-3 $\beta$ -ol	24-methylenecholesterol	19.6	3.4	8.2	89.5
7	24-methylcholest-5-en-3 $\beta$ -ol	24-methylcholesterol	4.2	5.3	6.5	0.2
	4-methyl-5 $\alpha$ -cholestan-3 $\beta$ -ol	4-methylcholestanol				
8	24-ethylcholesta-5, 22E-dien-3 $\beta$ -ol		2.9	3.9	0.5	0.2
9	24-ethylcholest-5-en-3 $\beta$ -ol	24-ethylcholesterol	28.8	16.0	12.9	0.2
	4, 23, 24-trimethylcholest-22E-en-3 $\beta$ -ol	dinosterol	3.2	0.1	—	—
10	4-methyl-24-ethylcholest-22E-en-3 $\beta$ -ol <sup>d</sup>		3.8	5.8	—	—
11	C <sub>30</sub> stanol		6.5	3.6	—	0.8
	Others		5.8	6.8	—	2.1
	Sum C <sub>26</sub>		0.2	1.8	3.1	—
	Sum C <sub>27</sub>		9.6	11.2	11.2	5.4
	Sum C <sub>28</sub>		39.2	50.7	72.3	91.2
	Sum C <sub>29</sub>		31.7	19.9	13.4	0.5
	Sum C <sub>30</sub>		13.5	9.6	—	0.9

<sup>a</sup> Peak numbers refer to Fig. 4

<sup>b</sup> Sampling site abbreviations as indicated in Table I

<sup>c</sup> C<sub>24</sub> stereochemistry not determined

<sup>d</sup> Tentative identification



**Fig. 4.** Partial gas chromatogram (42–56 min) illustrating a representative sterol profile obtained for Cape Armitage sea-ice diatom community, 28 December 1985. Peak numbers refer to Table II.

1979). Its presence in Antarctic marine sediments in McMurdo Sound (Venkatesan 1988), in soils associated with the dry valley lakes of Victoria Land (Matsumoto *et al.* 1982) and in cyanobacterial mats (modern stromatolites) from Antarctic oasis lakes (Orcutt *et al.* 1986), where terrestrial input is negligible, has been attributed to cyanobacterial or green algal input. The similarity of the sterol profiles of sea-ice diatom communities (Table II) to those observed in previous Antarctic studies strongly indicates that diatoms, rather than cyanobacteria or other algal groups previously proposed, are a more probable source of  $C_{29}$  sterols, including 24-ethylcholesterol, in these extreme environments.

The occurrence of 24-ethylcholesterol at high relative abundance in the sea-ice diatom communities extends the number of marine algal classes in which this sterol is found as a major component. This result is relevant to studies of ancient sediments. Sterols are converted to steranes in ancient sediments, and the ratio of  $C_{27}$ : $C_{29}$  steranes is often used as an index of the relative amounts of marine and terrigenous organic matter. The finding of  $C_{29}$  sterols at high abundance in marine algae, and in this instance sea-ice diatoms, supports the view that organic and petroleum geochemists should not assume that  $C_{29}$  sterols or steranes are necessarily derived from terrigenous sources (Jones *et al.* 1987, Volkman 1986). In Antarctic studies, variations in the ratio of  $C_{27}$ : $C_{29}$  sterols or steranes are more likely to reflect changes in species composition, rather than variations in the proportions of marine and terrestrial input.

The occurrence of a  $C_{26}$  sterol as a minor component in the

sea-ice diatom communities is also noteworthy. A  $C_{26}$  sterol was previously noted as a minor constituent of the diatom *Thalassionema nitzschioides* (Ballantine *et al.* 1979). A number of studies have used 24-norcholesta-5, 22-dien-3 $\beta$ -ol as an invertebrate marker (Gillan 1981, Volkman *et al.* 1981). The discovery of  $C_{26}$  sterols in diatoms suggests their use as invertebrate markers should be qualified with additional data.

Three  $C_{30}$  sterols were also detected in the sea-ice diatom communities. Dinosterol, identified from mass spectral data and coinjection with the authentic sterol, was present as a minor component (3.2% of total sterol, Table II) in the Erebus Ice Tongue samples. Dinosterol has to date only been reported in dinoflagellates and has therefore been used as a biomarker for this algal group (de Leeuw *et al.* 1983). However, in a recent study of the sterol distribution found in microbial mats in Solar Lake adjacent to the Gulf of Aqaba in north-east Sinai, it was suggested that dinosterol and related sterols are not exclusive markers for dinoflagellates, but may be produced by other organisms (Edmunds & Eglinton 1984). Two other  $C_{30}$  sterols (sterols nos 10 and 11; Table II, Fig. 4) were detected in both the Cape Armitage and Erebus Ice Tongue samples. Although standards are not presently available, the mass spectra of sterol no. 10 was identical to that observed for a  $C_{30}$  sterol identified as 4-methyl-24-ethylcholest-22E-en-3 $\beta$ -ol found in the prymnesiophyte *Pavlova lutheri* (Ballantine *et al.* 1979). Sterol no. 10 eluted, however, just after the sterol isolated from *Pavlova*. Based on the data presently available, sterol no. 10 is structurally very similar to 4-methyl-24-ethylcholest-22E-en-3 $\beta$ -ol, but may differ in the position or stereochemistry of a side-chain substituent group. The mass spectrum of sterol no. 10 (and 4-methyl-24-ethylcholest-22E-en-3 $\beta$ -ol) are distinguished from that of dinosterol based on their higher  $m/z$  83 to  $m/z$  69 ratio. Sterol no. 11 was identified as a  $C_{30}$  stanol from GC-MS data, and eluted just after authentic dinostanol (unpublished data). It is presumed to be derived from sterol no. 10.

Whilst further work is needed to determine the precise structures of the  $C_{30}$  sterols (nos 10 and 11) in these Antarctic sea-ice communities, their occurrence raises a number of interesting points.  $C_{30}$  sterols have not been previously reported in diatoms. Their occurrence in natural sea-ice diatoms suggests that other flagellates, not observed by microscopy, may be present or, more probably, that sea-ice diatoms may biosynthesize  $C_{30}$  sterols containing a methyl group at the  $C_4$  position. Small amounts of  $C_{28}$  and  $C_{29}$  sterols containing a methyl group at the  $C_4$  position were also detected in the Cape Armitage and Erebus Ice Tongue sea-ice diatom communities; these sterols are most likely precursors to the  $C_{30}$  sterols.

There has been increased interest recently in the occurrence of dinosterane and other 4-methyl steranes in ancient sediments and petroleum (Wolff *et al.* 1986a, b, Summons *et al.* 1987). In this study, we have identified a third 4-methyl  $C_{30}$

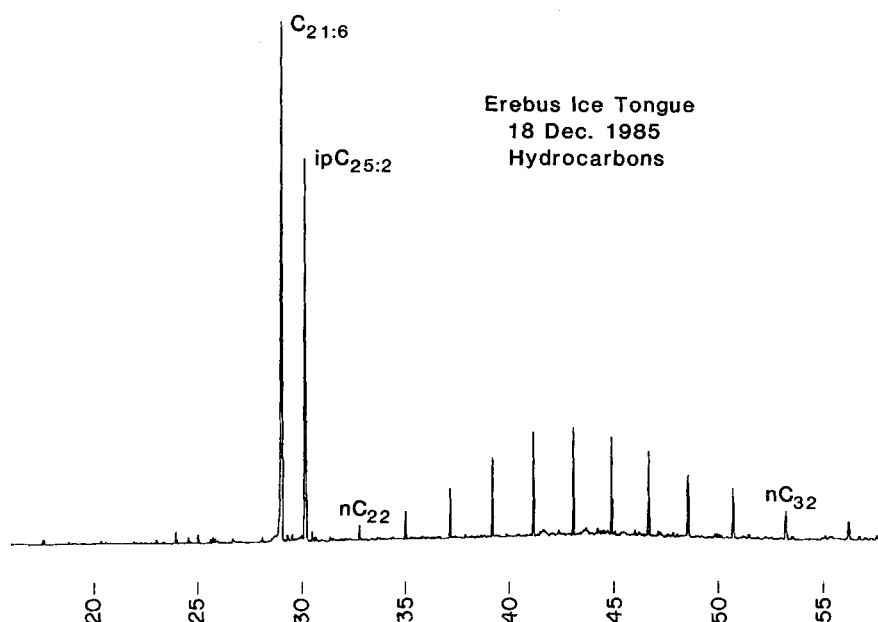


Fig. 5. Partial gas chromatogram (16–58 min) illustrating representative hydrocarbon profile obtained for Erebus Ice Tongue sea-ice diatom community, 18 December 1985.

sterol (in addition to dinosterol and 4-methyl-24-ethylcholesterol-22-en-3 $\beta$ -ol) in sea-ice diatoms that may be a potential precursor to 4-methyl cyclic hydrocarbons found in ancient sediments. Further studies of the occurrence of these biomarkers will enhance our knowledge of the biology of fossil algae as well as providing an additional tool for investigating the relationships between petroleum and their source rocks.

### Hydrocarbons

A representative sea-ice diatom hydrocarbon profile obtained for the Erebus Ice Tongue community is shown in Fig. 5. The hydrocarbons were dominated by two unsaturated components:  $n$ - $C_{21:6}$  and a diunsaturated isoprenoid  $C_{25}$  alkene ( $ipC_{25:2}$ ). The hydrogenation product of the diene was identified as 2,6,10,14-tetramethyl-7-(3-methylpentyl)pentadecane (Nichols *et al.* 1988).  $n$ - $C_{21:6}$  is often the major hydrocarbon in photosynthetic diatoms (Lee & Loeblich 1971). Until the present study, no biological source had been recorded for the  $C_{25:2}$  isoprenoid diene, even though there are numerous reports of this and structurally similar alkenes in estuarine and coastal sediments (see Robson & Rowland 1986).

The hydrocarbon distribution of McMurdo Sound sediments was recently reported in detail (Venkatesan 1988). The  $C_{25}$  alkene and related hydrocarbons were major components in the hydrocarbon fraction; however, a precise source for these alkenes was not determined, although the high primary productivity during the austral summer was recognized. Our observation of the  $C_{25}$  diene in sea-ice diatoms indicates that unicellular algae can synthesize these hydrocarbons, and it seems likely that diatoms are a major source of these alkenes which are widely distributed in particulate matter and sediments.

Sea-ice diatoms are important constituents of the carbon and energy flux in polar regions. The lipid class data presented in this report have provided an insight into changes occurring in the physiological status of sea-ice algal communities during a spring bloom. Analysis of individual lipid classes (sterols and hydrocarbons) at the molecular level has also yielded information important to studies of chemotaxonomy, dietary transfer of lipids in polar marine food-webs, and aspects of the microbial ecology, oceanography and organic geochemistry of diatom communities and associated sediments. Together, the biochemical analyses undertaken in this study provide information not readily available using classical techniques on the development, physiology, and ecology of sea-ice microbial communities.

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