# Novel Brominated Phospholipid Fatty Acids from the Caribbean Sponge Petrosia Sp.

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# NOVEL BROMINATED PHOSPHOLIPID FATTY ACIDS FROM THE CARIBBEAN SPONGE *PETROSIA* SP.[1]

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ABSTRACT.—The long-chain fatty acids (5*E*,9*Z*)-6-bromo-5,9-heptacosadienoic acid [**2a**], (5*E*,9*Z*)-6-bromo-5.9-octacosadienoic [**3a**], (*Z*)-19-heptacosenoic acid [**4**], and the novel aldehydes 17-pentacosenal [**5**] and 17-hexacosenal [**6**] were identified in the phospholipids (mainly phosphatidylethanolamine and phosphatidylcholine) of the sponge *Petrosia* sp. Structural elucidation was accomplished by means of mass spectrometry and chemical transformations, including deuteration with Wilkinson's catalyst. The dimethyldisulfide derivatization of $\Delta^{5,9}$ fatty acids is also presented.

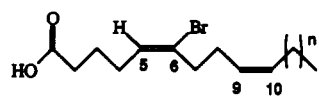
Phospholipids with brominated fatty acids are rare in nature. Only two reports of naturally occurring phospholipid brominated fatty acids have been documented. Two heptacosadienoic acids with the 5,9-diene pattern, *iso/anteiso* methyl branching, and the bromovinyl functionality were isolated from the sponges *Petrosia ficiformis* and *Petrosia hebes* (1). The long-chain fatty acid (5*E*,9*Z*)-6-bromo-5,9-hexacosadienoic acid [**1a**] was also isolated from the phospholipids of a Hymeniacidonid sponge (1).

Other free halogenated long-chain fatty acids have been reported in the marine environment. Six fatty acid chlorohydrins of palmitic and stearic acids (with the combinations 9-chloro-10-hydroxy and 10-chloro-9-hydroxy) were isolated from the jellyfish *Aurita aurita* (3). Schmitz and Gopichand (4) isolated (7*E*,13ξ,15*Z*)-14,16-dibromo-7,13,15-hexadecatrien-5-ynoic acid from the sponge *Xestospongia muta*. Other examples include a monobrominated straight-chain $C_{18}$ bisacetylenic acid from *Xestospongia testudinaria* (5) and six mono- and dibrominated straight-chain unsaturated $C_9$, $C_{16}$, and $C_{18}$ acids from the same genus (6).

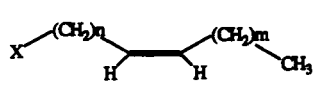
In this work we report the isolation and structural elucidation of two new brominated phospholipid fatty acids, (5*E*,9*Z*)-6-bromo-5,9-heptacosadienoic acid [**2a**], and (5*E*,9*Z*)-6-bromo-5,9-octacosadienoic acid [**3a**], from the phospholipids of a *Petrosia* sp. de Laubenfels (family Petrosiidae, order Haplosclerida), which was recently collected in an expedition to Mona Island, Puerto Rico. We also report the characterization of (*Z*)-19-heptacosenoic acid [**4**], and the novel aldehydes 17-pentacosenal [**5**] and 17-hexacosenal [**6**] which were also found in the phospholipids of the same sponge.

## RESULTS AND DISCUSSION

The principal phospholipids from *Petrosia* sp. were identified by tlc as phosphatidylethanolamine and phosphatidylcholine, but small amounts of phosphatidylserine




**1a** n=15
**2a** n=16
**3a** n=17




**4** X=$HO_2C$, n=17, m=6
**5** X=OHC, n=15, m=6
**6** X=OHC, n=15, m=7

[1]Dedicated to Prof. Carl Djerassi, a teacher and a friend, on the occasion of his 70th birthday.

and phosphatidylinositol were also detected. The major fatty acids and aldehydes from the phospholipids were identified from the gc equivalent chain length values (ECL) and eims of the methyl esters, the *N*-acylpyrrolidides of the acids, and the dimethyl acetals

TABLE 1. Principal Phospholipid Fatty Acids and Aldehydes from *Petrosia* sp.

| ECL[a] | Compound | Abundance (wt %) |
|---|---|---|
| | Aldehydes | |
| 16.49 | Hexadecanal (16:0) | 0.6 |
| 17.43 | Heptadecanal (17:0) | 0.8 |
| 18.55 | Octadecanal (18:0) | 4.1 |
| 24.28 | 17-Tetracosenal (24:1) | 2.1 |
| 24.80 | 17-Pentacosenal (25:1)[b] | 3.2 |
| 26.16 | 17-Hexacosenal (26:1)[b] | 0.5 |
| | Fatty Acids | |
| 13.68 | 12-Methyltridecanoic (*i*- 14:0) | 0.6 |
| 14.00 | Tetradecanoic (14:0) | 2.2 |
| 14.52 | 4,8,12-Trimethyltridecanoic (16:0) | 0.5 |
| 14.68 | 13-Methyltetradecanoic (*i*- 15:0) | 9.8 |
| 14.74 | 12-Methyltetradecanoic (*ai*- 15:0) | 1.7 |
| 15.00 | Pentadecanoic (15.0) | 0.4 |
| 15.61 | 14-Methylpentadecanoic (*i*- 16:0) | 1.5 |
| 15.73 | 9-Hexadecenoic (16:1) | 4.2 |
| 15.80 | 11-Hexadecenoic (16:1) | 0.8 |
| 16.00 | Hexadecanoic (16:0) | 9.5 |
| 16.36 | 15-Methyl-9-hexadecenoic (17:1) | 7.7 |
| 16.43 | Methylhexadecanoic (17:0) | 0.2 |
| 16.64 | 15-Methylhexadecanoic (*i*- 17:0) | 2.0 |
| 16.72 | 14-Methylhexadecanoic (*ai*- 17:0) | 1.8 |
| 16.82 | 2-Methoxy-5-hexadecenoic (16:1) | 1.6 |
| 16.80 | 9-Heptadecenoic (17:1) | 0.3 |
| 16.89 | 11-Heptadecenoic (17:1) | 0.3 |
| 17.00 | Heptadecanoic (17:0) | 1.6 |
| 17.76 | 11-Octadecenoic (18:1) | 4.3 |
| 18.00 | Octadecanoic (18:0) | 3.9 |
| 18.38 | Methyloctadecanoic (19:0) | 1.4 |
| 18.63 | 17-Methyloctadecanoic (*i*- 19:0) | 0.6 |
| 18.80 | 11-Nonadecenoic (19:1) | 3.8 |
| 19.00 | Nonadecanoic (19:0) | 0.2 |
| 19.19 | 5,8,11,14-Eicosatetraenoic (20:4) | 2.0 |
| 19.24 | 8,11,14,17-Eicosatetraenoic (20:4) | 1.3 |
| 19.96 | Methylnonadecanoic (20:0) | 0.5 |
| 20.00 | Eicosanoic (20:0) | 0.3 |
| 20.59 | 19-Methyleicosanoic (*i*- 21:0) | 0.6 |
| 20.71 | 18-Methyleicosanoic (*ai*- 21:0) | 0.6 |
| 20.80 | 13-Heneicosenoic (21:1) | 0.5 |
| 21.09 | 4,7,10,13,16,19-Docosahexaenoic (22:6) | 1.8 |
| 21.20 | 7,10,13,16-Docosatetraenoic (22:4) | 1.2 |
| 22.00 | Docosanoic (22:0) | 0.2 |
| 22.65 | 21-Methyldocosanoic (*i*- 23:0) | 0.3 |
| 23.00 | Tricosanoic (23:0) | 0.2 |
| 21.43 | 2-Hydroxydocosanoic (*h*- 22:0) | 0.6 |
| 23.75 | 16-Tetracosenoic (24:1) | 0.3 |
| 23.81 | 17-Tetracosenoic (24:1) | 0.8 |
| 23.87 | 19-Tetracosenoic (24:1) | 0.2 |
| 24.00 | Tetracosanoic (24:0) | 0.2 |
| 24.35 | 17-Pentacosenoic (25:1) | 1.0 |
| 25.37 | 5,9-Hexacosadienoic (26:2) | 0.7 |

TABLE 1. Continued.

| ECL[a] | Compound | Abundance (wt %) |
|---|---|---|
| 25.63 | 17-Hexacosenoic (26:1) | 1.0 |
| 25.73 | 19-Hexacosenoic (26:1) | 1.3 |
| 26.00 | Hexacosanoic (26:0) | 0.2 |
| 26.46 | 5,9-Heptacosadienoic (27:2) | 0.6 |
| 26.90 | 19-Heptacosenoic (27:1)[b] | 0.3 |
| 27.00 | Heptacosanoic (27:0) | 1.6 |
| 27.54 | 5,9-Octacosadienoic (28:2) | 4.9 |
| 27.89 | 6-Bromo-5,9-hexacosadienoic (26:2) | 0.6 |
| 29.01 | 6-Bromo-5,9-heptacosadienoic (27:2)[b] | 0.7 |
| 29.30 | 5,9,23-Tricontatrienoic (30:3) | 0.3 |
| 30.05 | 6-Bromo-5,9-octacosadienoic (28:2)[b] | 3.0 |

[a]Equivalent chain length (ECL) values are those of the methyl esters of these acids.
[b]These acids have not been described before in nature.

of the aldehydes. The complete list of fatty acids and aldehydes is shown in Table 1. *Petrosia* sp. had six aldehydes, probably arising from 1-*O*-alk-1-enyl-2-acyl-*sn*-glycero-3-phosphoethanolamine, a common plasmalogen in sponges (7). Two of these aldehydes have not been previously described in nature, namely (*Z*)-17-pentacosenal [**5**] and (*Z*)-17-hexacosenal [**6**]. As dimethyl acetals these aldehydes normally do not afford a molecular ion upon eims, but an ion equivalent to $[M-31]^+$ (loss of a methoxy group) permits the determination of the mol wt (8). The dimethyl acetals displayed a base peak at *m/z* 75 due to the $[C_3H_7O_2]^+$ fragment. For example, (*Z*)-17-pentacosenal dimethyl acetal presented an $[M-31]^+$ peak at *m/z* 379 and a base peak at *m/z* 75, while 17-hexacosenal dimethyl acetal had an $[M-31]^+$ peak at *m/z* 393 and also a base peak at *m/z* 75. The double bond positions in the dimethyl acetals were initially determined by ms, using the corresponding dimethyl disulfide derivatives (9), and further confirmed by oxidative cleavage with $KMnO_4/NaIO_4$. For example, (*Z*)-17-pentacosenal dimethyl acetal afforded, upon reaction with dimethyl disulfide and iodine in $Et_2O$, 17,18-bis(methylthio)pentacosanal (during workup the dimethyl acetal was transformed into the aldehyde), which upon eims cleaves efficiently between C-17 and C-18, affording fragments at *m/z* 299 $[C_{18}H_{35}SO]^+$ and at *m/z* 159 $[C_9H_{19}S]^+$, allowing unambiguous determination of the double bond position at C-17. These results were further confirmed by $KMnO_4/NaIO_4$ oxidation of (*Z*)-17-pentacosenal dimethyl acetal. which gave methyl 17,17-bis(methoxy)heptadecanoate and methyl octanoate. Aldehyde **6** was characterized in a similar way. The 17,18-bis(methylthio)hexacosanal provided major fragments at *m/z* 299 $[C_{18}H_{35}SO]^+$ and at *m/z* 173 $[C_{10}H_{21}S]^+$, while $KMnO_4/NaIO_4$ oxidation of (*Z*)-17-hexacosenal dimethyl acetal afforded methyl 17,17-bis(methoxy)heptadecanoate and methyl nonanoate. A careful gc Ft-ir of the mixture presented no absorption in the 960–980 $cm^{-1}$ region, indicating cis rather than trans unsaturation.

The previously unreported (*Z*)-19-heptacosenoic acid [**4**] was also characterized in the phospholipid mixture. Compound **4** afforded, after hydrogenation in MeOH with $PtO_2$ as catalyst, methyl heptacosanoate as confirmed by gc co-injection with an authentic sample. This experiment excludes the possibility of any methyl branching. The double bond position was determined by derivatization to methyl 19,20-bis(methylthio)heptacosanoate. The double bond was found to be at C-19 from the ms fragments at *m/z* 357 $[C_{21}H_{41}SO_2]^+$ and *m/z* 159 $[C_9H_{19}S]^+$. The structure was confirmed

by $KMnO_4/NaIO_4$ oxidation, since methyl 19-heptacosanoate cleaved to nonadecanedioic acid dimethyl ester and methyl octanoate.

The $\Delta^{5,9}$ demospongic acids isolated from *Petrosia* sp. were characterized as 5,9-hexacosadienoic acid, 5.9-heptacosadienoic acid, and 5,9-octacosadienoic acid (Table 1). The complete characterization of all of these fatty acids has been previously described (10). However, in this work we investigated the reaction of $\Delta^{5,9}$ fatty acid methyl esters with dimethyldisulfide (DMDS) and iodine in $Et_2O$. The reaction with DMDS gave a five-membered cyclic thioether substituted with two alkyl chains, each containing a

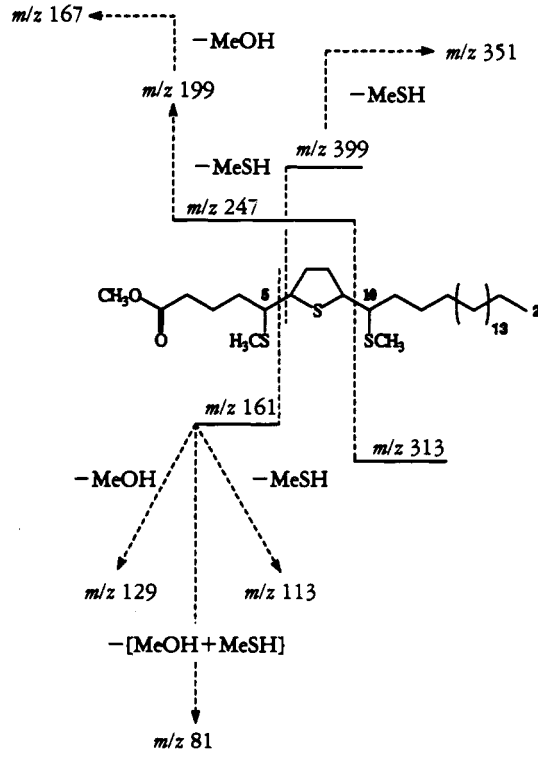



FIGURE 1. Mass spectral fragmentations for the 2-(4-methoxycarbonyl-1-methylthiobutan-1-yl)-5-(1-methylthiononadecan-1-yl)tetrahydrothiophene.

methylthio group α to the ring (11), thus allowing an unequivocal identification of the double bond positions. The principal mass spectral fragmentations for the DMDS derivative of methyl 5,9-octacosadienoate, [2-(4-methoxycarbonyl-1-methylthiobutan-1-yl)-5-(1-methylthiononadecan-1-yl)tetrahydrothiophene], are presented in Figure 1. Similar derivatives were obtained for the other $\Delta^{5,9}$ fatty acid methyl esters in the sponge.

The most interesting fatty acids in the sponge were a series of three brominated $\Delta^{5,9}$ demospongic acids. One of the fatty acids was immediately identified as the previously reported (2) 6-bromo-5,9-hexacosadienoic acid [**1a**]. All of the brominated fatty acids presented similar spectral characteristics. The $^1$H-nmr spectrum presented multiplets between 5.32 and 5.41 ppm for the C-9 and C-10 hydrogens, while the C-5 hydrogen

was observed as a triplet at 5.84 ppm. The gc/ms of the brominated fatty acid methyl esters showed strong mass spectral peaks at *m/z* 74 (due to the McLafferty rearrangement typical of fatty acid methyl esters) and $[M-79]^+$ peaks as molecular ions, indicating facile loss of bromine under electron impact.

The gc-ms of the corresponding brominated fatty acid pyrrolidides was critical for identifying the position of the bromine substituent. Compound **3b** afforded a strong peak at *m/z* 472 due to the loss of bromine. Similar peaks were observed for the pyrrolidides of the other fatty acids, differing by 14 amu depending on the chain length. However, common to all of these pyrrolidides were a base peak at *m/z* 113 due to McLafferty rearrangement, a peak not so prominent at *m/z* 180, which corresponded to a double allylic fragmentation between C-6 and C-9 with the loss of bromine, and a strong doublet of equal intensity at *m/z* 258 and *m/z* 260 due to the same fragmentation with the bromine substituent intact (Figure 2). This information limited the point of attachment of the bromine between C-1 and C-7.

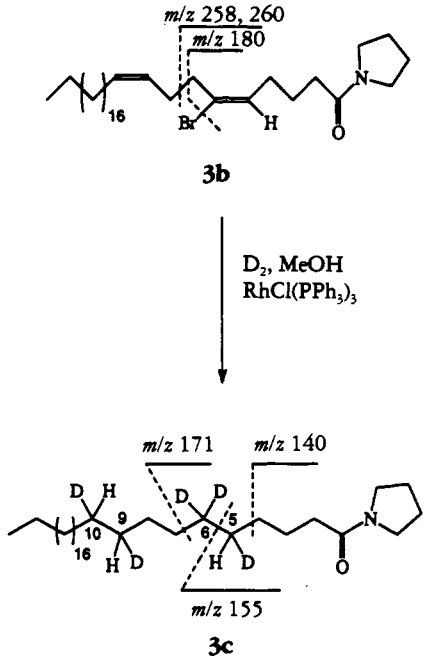



FIGURE 2. Key mass spectral fragmentations for the *N*-(5*E*,9*Z*)-6-bromooctacosa-5,9-dienoylpyrrolidine [**3b**] and the *N*-[5,6,6,9,10-$^2H_5$]-octacosanoylpyrrolidine [**3c**].

Catalytic hydrogenation ($PtO_2$) of the brominated methyl esters yielded the corresponding saturated fatty acids. For example, 6-bromo-5,9-hexacosadienoic acid methyl ester afforded methyl hexacosanoate, 6-bromo-5,9-heptacosadienoic acid methyl ester afforded methyl heptacosanoate, and 6-bromo-5,9-octacosadienoic acid methyl ester afforded methyl octacosanoate. These results were confirmed by gc co-injection with authentic samples obtained from Sigma. This information now limited the point of attachment of the bromine between C-5 and C-6.

The exact location of the bromine substituent was confirmed by deuteration of the pyrrolidides utilizing Wilkinson's catalyst [$RhCl(PPh_3)_3$] in MeOH in order to avoid

deuterium scrambling (Figure 2). Five deuteriums were incorporated into the brominated fatty acid pyrrolidides. In all of the deuterated fatty acid pyrrolidides a difference of 15 amu was observed between C-4 and C-5, between C-8 and C-9, and between C-9 and C-10, indicating the addition of deuterium atoms to the double bonds. Moreover, a difference of 16 amu was detected between C-5 (*m/z* 155) and C-6 (*m/z* 171), clearly demonstrating an additional replacement of bromine with deuterium at C-6 (1).

The gc Ft-ir spectra of the original brominated fatty acid pyrrolidides displayed absorption peaks at 3018, 2932, 2868, 1682, 1413, 1264, 831, 726, and 648 $cm^{-1}$. No peak at 980–968 $cm^{-1}$ was observed, indicating cis rather than trans unsaturation for the $\Delta^9$ double bond, as is usual for demospongic acids (2). The stereochemistry at the $\Delta^5$ double bond was previously established as *E* by a microscale metal-halogen exchange reaction followed by protonolysis (2).

We can only speculate on the origin of the two new brominated acids, but it is possible that they originated by the action of a bromoperoxidase on their non-brominated counterparts, which were also found in this sponge (12). In fact, the biosynthesis of the 6-bromo-5,9-hexacosadienoic acid [**1a**] was previously investigated by Lam *et al.* (2) utilizing radiolabeled precursors, and they concluded that the bromination was the terminal step in the biosynthesis of these unusual acids. From the available data in the literature, we believe that the biosynthesis of the brominated acids follows the biosynthetic route shown below, as an example, for the 6-bromo-5,9-octacosadienoic acid [**3a**]:

$$16{:}0 \rightarrow 28{:}0 \rightarrow \Delta^5\text{- and } \Delta^9\text{-}28{:}1 \rightarrow \Delta^{5,9}\text{-}28{:}2 \rightarrow 6\text{-Br-}\Delta^{5,9}\text{-}28{:}2$$

Work is in progress trying to elucidate the origin of these unusual fatty acids in sponges.

## EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—The methyl esters were analyzed by gc-ms using a Hewlett-Packard 59970 MS ChemStation (Hewlett-Packard, Palo Alto, CA) equipped with a 30 m×0.32 mm nonpolar fused silica column (Supelco, Bellefonte, PA) with SPB™-1 as the bonded phase. Gc/Ft-ir spectra were recorded on a Nicolet (Madison, WI) 740 FT-IR spectrometer. The sponge was freeze-dried on a Labconco Freeze Dryer 4.5 (Model 77510; Kansas City, MO).

COLLECTION AND DESCRIPTION OF *PETROSIA* SP.—*Petrosia* sp. was collected August 1, 1992, near Mona Island, Puerto Rico, at a depth of 25 m. The sponge has been assigned the number M-10, and a voucher specimen is available at the Department of Chemistry of the University of Puerto Rico. Dr. Vance Vicente from the United States Department of the Interior Fish and Wildlife Service classified the sponge, and a specimen will also be deposited at the National Museum of Natural History, Smithsonian Institution. This new species is closely related to *Petrosia pellasarca* de Laubenfels 1934, having almost identical morphological characteristics with the exception that *P. pellasarca* displays a pink-red color and the present sponge a light green color.

EXTRACTION AND ISOLATION OF PHOSPHOLIPIDS.—The sponge (40 g) was carefully cleaned of all nonsponge debris and cut into small pieces. Extraction with 250 ml of $CHCl_3$-MeOH (1:1) yielded the total lipids. The neutral lipids, glycolipids, and phospholipids (20 mg) were separated by cc on Si gel (60–200 mesh) using the procedure of Privett *et al.* (13). The phospholipid classes were fractionated by preparative tlc using Si gel 60 and $CHCl_3$-MeOH-$NH_4OH$ (65:35:5) as solvent.

PREPARATION AND ISOLATION OF FATTY ACID DERIVATIVES.—The fatty acyl components of the phospholipids were obtained as their methyl esters by reaction of the phospholipids with methanolic HCl (14) followed by cc purification eluting with *n*-hexane–$Et_2O$ (9:1). For the location of double bonds, pyrrolidides were prepared by direct treatment of the methyl esters with pyrrolidine-HOAc (10:1) in a capped vial (2 h at 100°) followed by ethereal extraction from the acidified solution and purification by preparative tlc (10). The double bond position of the monoenoic acids was elucidated by preparing the corresponding dimethyl disulfide derivatives by dissolving the esters (2 mg) in dimethyldisulfide (0.2 ml), adding a solution (0.05 ml) of iodine in $Et_2O$ (60 mg/ml), and heating the solution at 50° for 24 h, followed

by the standard workup (9). Hydrogenations were carried out in 10 ml of MeOH and catalytic amounts of $PtO_2$. Deuterations were carried out also in 10 ml of MeOH and catalytic amounts of Wilkinson's catalyst [$RhCl(PPh_3)_3$]. Deuterium was obtained from the reaction of Na and $D_2O$. Spectral data for the key fatty acids for this discussion follows.

*17-Pentacosenal dimethyl acetal.*—Ms *m/z* (rel. int.) $[M-31]^+$ 379 (0.8), 362 (0.5), 347 (0.5), 207 (0.2), 163 (0.4), 149 (0.9), 137 (1.4), 135 (1.9), 123 (2.8), 121 (2.6), 111 (4), 110 (4), 109 (6), 107 (1.2), 101 (1.6), 98 (2), 95 (11), $[C_3H_7O_2]^+$ 75 (100), 67 (14).

*17,18-Bis(methylthio)pentacosanal.*—Ms *m/z* (rel. int.) $[M]^+$ 458 (4), $[C_{18}H_{35}SO]^+$ 299 (100), 283 (1.7), 271 (2), 255 (3), 229 (1.5), 199 (1.4), 187 (1.3), 185 (1.2), 177 (1), $[C_9H_{19}S]^+$ 159 (65), 149 (3), 143 (4), 135 (5), 133 (3), 131 (4), 123 (6), 121 (6), 117 (4), 111 (12), 109 (15), 105 (8), 97 (16.6), 69 (86).

*17-Hexacosenal dimethyl acetal.*—Ms *m/z* (rel. int.) $[M-31]^+$ 393 (0.6), 361 (0.4), 209 (0.3), 207 (0.3), 190 (0.2), 177 (0.3), 163 (0.4), 157 (0.4), 149 (1), 137 (1), 135 (2.6), 123 (2.3), 121 (2.9), 108 (1.8), 107 (1.5), 101 (1.5), 98 (1), 97 (8.8), 95 (11), $[C_3H_7O_2]^+$ 75 (100), 67 (17).

*17,18-Bis(methylthio)hexacosanal.*—Ms *m/z* (rel. int.) $[M]^+$ 472 (3.4), 400 (1.4), 317 (2.6), $[C_{18}H_{35}SO]^+$ 299 (88), 271 (2), 255 (2.5), 225 (1.9), 213 (2.2), 201 (2), 199 (3.4), 197 (2), 193 (2), 187 (2), $[C_{10}H_{21}S]^+$ 173 (60), 167 (6), 159 (11), 151 (6), 143 (6), 141 (3), 139 (4), 111 (15), 109 (19), 105 (11), 97 (46).

*Methyl 19-heptacosenoate.*—Ms *m/z* (rel. int.) $[M-32]^+$ 390 (6), 333 (1.2), 306 (1), 292 (1), 222 (1), 207 (1), 179 (1), 165 (2), 149 (2.8), 143 (3), 137 (3), 135 (2), 127 (4), 125 (5), 123 (4), 121 (4), 115 (3), 111 (13), 107 (3), 101 (4), 98 (17), 97 (33), 96 (20), 95 (18), 87 (28), 83 (46), 81 (25), 75 (62), 74 (49), 69 (58).

*Methyl 19,20-bis(methylthio)heptacosanoate.*—Ms *m/z* (rel. int.) $[M]^+$ 516 (1), $[C_{21}H_{41}SO_2]^+$ 357 (15), $[C_{20}H_{37}SO]^+$ 325 (10), $[C_9H_{19}S]^+$ 159 (26), 111 (19), 109 (15), 95 (26), 87 (44), 83 (29), 81 (40), 75 (12), 74 (24), 69 (63), 67 (55), 59 (21).

*Methyl (5E,9Z)-6-bromo-5,9-heptacosadienoate.*—Ms *m/z* (rel. int.) $[M-Br]^+$ 419 (7), $[M-Br-MeOH]^+$ 387 (2.6), 278 (5), 221 (2.5), 2.07 (2), 195 (2), 191 (2.4), 181 (3.8), 179 (4), 177 (2.8), 174 (2.6), 167 (3), 161 (11.5), 159 (9), 149 (11), 147 (10.8), 141 (14), 139 (14), 135 (14), 133 (8), 131 (4), 121 (12), 119 (11.6), 111 (10), 109 (21), 107 (17), 105 (15), 97 (27), 95 (25), 93 (23), 91 (25), 85 (12), 83 (35), 81 (56), 79 (54), 77 (12.7), 74 (26), 71 (23), 69 (46), 67 (52).

*N-(5E,9Z)-6-Bromoheptacosa-5,9-dienoylpyrrolidine.*—Ms *m/z* (rel. int.) $[M-Br]^+$ 458 (27), 260 (13), $[C_{11}H_{17}NOBr]^+$ 258 (18), $[C_{11}H_{18}NO]^+$ 180 (5), 126 (9), $[C_6H_{11}NO]^+$ 113 (100), 98 (28), 85 (18).

*Methyl (5E,9Z)-6-bromo-5,9-octacosadienoate.*—Ms *m/z* (rel. int.) $[M-Br]^+$ 433 (7.6), $[M-Br-MeOH]^+$ 401 (2.5), 383 (1), 359 (1), 293 (1), 292 (5.5), 250 (1.2), 221 (2), 219 (1.3), 207 (1.3), 194 (2.3), 191 (2), 179 (4), 177 (2.5), 167 (3.5), 161 (11), 159 (10), 149 (10.5), 147 (10.3), 141 (14), 139 (16), 135 (11), 133 (7.6), 131 (4.4), 121 (12.4), 119 (10.5), 111 (10.3), 109 (21), 107 (15), 105 (13), 97 (27.6), 95 (26), 91 (20), 85 (14), 83 (36.4), 81 (54), 79 (52.5), 74 (21), 71 (24), 69 (49), 67 (54).

*N-(5E,9Z)-6-Bromooctacosa-5,9-dienoylpyrrolidine.*—*Ms m/z* (rel. int.) $[M-Br]^+$ 472 (29), 260 (18), $[C_{11}H_{17}NOBr]^+$ 258 (17), $[C_{11}H_{18}NO]^+$ 180 (5), 126 (8.6), $[C_6H_{11}NO]^+$ 113 (100), 98 (21), 85 (15).

*2-(4-Methoxycarbonyl-1-methylthiobutan-1-yl)-5-(1-methylthiononadecan-1-yl)tetrahydrothiophene.*—Ms *m/z* (rel. int.) $[M-CH_3SH]^+$ 512 (6), $[C_{24}H_{47}S_2]^+$ 399 (9), $[C_{23}H_{43}S]^+$ 351 (51), $[C_{20}H_{41}S]^+$ 313 (8), $[C_{11}H_{19}S_2O_2]^+$ 247 (24), $[C_{10}H_{15}SO_2]^+$ 199 (82), $[C_9H_{11}SO]^+$ 167 (75), $[C_7H_{13}SO_2]^+$ 161 (23), $[C_6H_9SO]^+$ 129 (33), $[C_6H_9O_2]^+$ 113 (15.7), $[C_5H_5O]^+$ 81 (91).

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