^1H and ^{13}C N.M.R. Studies on the Positional Isomers of Methyl Thialaurate and Methyl Thiastearate

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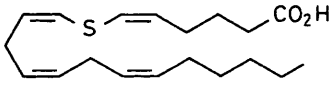
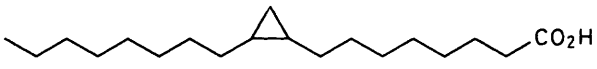
^1H N.m.r. analysis of methyl thialaurate and methyl thiastearate shows that the sulphur atom in the alkyl chain has a significant deshielding effect (α-effect) on the chemical shift of the protons of the adjacent methylene and methyl groups, and also on the methyl protons of the methoxycarbonyl (ester) function. It was possible to identify seven of the positional isomers in each series by examining the chemical shifts and the splitting pattern of the signals in the ^1H n.m.r. spectra of these fatty acid ester analogues. The ^{13}C n.m.r. results showed that this S atom has an interesting effect (α-, β-, and γ-effect) on the chemical shift of the carbonyl carbon atom of the methoxycarbonyl (ester) function: the sulphur atom exerts a significant shielding effect in the case of the 2-, 3-, and 4-thia isomers. In the remaining isomers the sulphur atom causes a strong deshielding effect on the methylene or methyl carbon atoms on either side of the sulphide linkage. These results permitted all positional isomers of methyl thialaurate to be identified by this technique, while 10 out of the 16 positional isomers of methyl thiastearate could be characterized.

Corey *et al.* have synthesized 7-thia-arachidonic acid (**1**), which has been found to be a potent irreversible inhibitor of leukotriene (lipoxin) biosynthesis.[1] This observation suggests a possible means of control of inflammatory diseases caused by an overproduction of leukotrienes in the human body.[2] The antibacterial activity of numerous sulphur-containing organic molecules is well documented and evidenced by the extensive



(**1**)



(**2**)

use of such compounds as antibiotics in medicine.[3] In the field of fatty acid chemistry, only a small number of long-chain fatty acids containing a sulphur atom has been synthesized in order to study their chemical, physical, or biological properties.[4–9] Pascal and Ziering have recently prepared the 9- and 10-thiastearic acid isomers and have reported on the inhibitory effects of these fatty acid analogues on dihydrosterculic acid (**2**) biosynthesis and on the growth of a protozoan species, *Crithidia fasciculata*.[10,11] Buist's group has been engaged in the study of the metabolism of thiastearic acids by yeast cells, *Saccharomyces cerevisiae*, and have noted a highly chemo-, regio-, and stereo-selective introduction of a (*Z*)-ethylenic bond into such thia fatty acid analogues.[12] In view of these important developments in the chemistry and biochemistry of thia analogues of long chain fatty acids, we have synthesized two complete series of thia fatty acid esters, *viz.* methyl thialaurate and methyl thiastearate, in an effort to study their spectroscopic and biological properties. In this paper we describe in brief the strategies adopted for the synthesis of these analogues, and report the results of the ^1H and ^{13}C n.m.r. properties of the positional isomers of methyl thialaurate and methyl thiastearate.

Results and Discussion

The ^1H n.m.r. chemical shift values of methyl thialaurate and methyl thiastearate isomers are presented in Tables 1 and 2. From the ^1H n.m.r. analysis of methyl 6-, 7-, and 8-thialaurate and methyl 6- to 14-thiastearate, the sulphur atom in the alkyl chain exerted a strong α-deshielding effect on the protons of the adjacent methylene groups, producing a characteristic signal at δ 2.50 (t, *J* 7.0 Hz) in the spectrum. The effect of the sulphur atom in the 2-thia isomers caused the chemical shift of the methyl protons of the ester function to appear at δ 3.80(s), while the C-3 methylene protons appeared downfield at δ 2.85, being additionally affected by the methyl ester function. The 3-thia isomers were readily differentiated from the other isomers by the appearance of a singlet at δ 3.20, due to the resonance shift of the protons of the C-2 methylene group. A partial overlap of three triplets in the spectrum of the 4-thia isomers was the result of the signals from the resonances of the protons of the C-2, C-3, and C-5 methylene groups. The 5-thia isomers were characterized by the appearance of a quintet at δ 1.98, due to the protons of the C-3 methylene group. In the isomers where the sulphur atom was located at the ω-1 position of the alkyl chain, the effect of the sulphur atom caused the methyl protons to shift to δ 2.08(s). The ω-2 isomers gave a signal at δ 1.24(t) for the terminal methyl protons; this signal was partially merged with the signal at δ 1.2–1.4 for the unperturbed methylene protons of the alkyl chain. In the case of the ω-3 isomers, the γ-effect of the sulphur atom on the protons of the terminal methyl group furnished a very distinct triplet at δ 0.9 as opposed to the distorted triplet signal commonly observed for methyl protons of long-chain fatty esters. By examining the chemical shifts and the splitting pattern of the signals in the ^1H n.m.r. spectra of these closely related positional isomers of thia fatty esters, it was possible to identify seven of the positional isomers in each series by this technique.

The ^{13}C n.m.r. chemical shift values of the series of methyl thialaurate and methyl thiastearate isomers are presented in Tables 3 and 4. In an effort to determine the incremental effects

Table 1. Values of δ_H(ppm) for all positional isomers of methyl thialaurate.

CH_3^a–CH_2^b–CH_2^c–CH_2^d–CH_2^e–CH_2^f–CH_2^g–CH_2^h–CH_2^i–CH_2^j–CH_2^k–$CO_2CH_3^l$ (CH_2 replaceable by S)

C_{12} Thia isomers	*a*	*b*	*c*	*d*	*e*	*f*	*g*	*h*	*i*	*j*	*k*	*l*
2-	0.88(t)					1.2–1.4(m)				2.85 (t, *J* 7 Hz)	S	3.80(s)
3-	0.88(t)				1.2–1.4(m)				2.62 (t, *J* 7 Hz)	S	3.20(s)	3.72(s)
4-	0.88(t)			1.2–1.4(m)				2.48 (t, *J* 7 Hz)	S	2.70 (t, *J* 6 Hz)	2.59 (t, *J* 6 Hz)	3.68(s)
5-	0.88(t)		1.2–1.4(m)				2.46 (t, *J* 7 Hz)	S	2.54 (t, *J* 7 Hz)	1.98 (qn, *J* 7 Hz)	2.44 (t, *J* 7 Hz)	3.67(s)
6-	0.89(t)			1.2–1.4(m)		2.50 (t, *J* 7 Hz)	S	2.50 (t, *J* 7 Hz)	1.2–1.4(m)		2.30 (t, *J* 7 Hz)	3.65(s)
7-	0.89(t)		1.2–1.4(m)		2.50 (t, *J* 7 Hz)	S	2.50 (t, *J* 7 Hz)		1.2–1.4(m)		2.30 (t, *J* 7 Hz)	3.65(s)
8-	0.91(t)		1.2–1.4(m)	2.50 (t, *J* 7 Hz)	S	2.50 (t, *J* 7 Hz)		1.2–1.4(m)			2.27 (t, *J* 7 Hz)	3.65(s)
9-	0.90 (t, *J* 7 Hz)	1.2–1.4(m)	2.48 (t, *J* 7 Hz)	S	2.48 (t, *J* 7 Hz)		1.2–1.4(m)				2.30 (t, *J* 7 Hz)	3.65(s)
10-	1.24 (t, *J* 6.8 Hz)	2.55 (q, *J* 6.8 Hz)	S	2.48 (t, *J* 7 Hz)			1.2–1.4(m)				2.30 (t, *J* 7 Hz)	3.65(s)
11-	2.08(s)	S	2.47 (t, *J* 7 Hz)			1.2–1.4(m)					2.30 (t, *J* 7 Hz)	3.65(s)

Table 2. Values of δ_H(ppm) for all positional isomers of methyl thiastearate.

CH_3^a–CH_2^b–CH_2^c–CH_2^d–$(CH_2^e)_9$–CH_2^f–CH_2^g–CH_2^h–CH_2^i–$COOCH_3^j$ (CH_2 replaceable by S)

C_{18} Thia isomers	*a*	*b*	*c*	*d*	*e*	*f*	*g*	*h*	*i*	*j*
2-	0.88(t)					1.2–1.4(m)		2.85 (t, *J* 7 Hz)	S	3.80(s)
3-	0.88(t)				1.2–1.4(m)			2.62 (t, *J* 7 Hz)	S	3.20(s)	3.73(s)
4-	0.89(t)			1.2–1.4(m)			2.50 (t, *J* 7 Hz)	S	2.70 (t, *J* 6 Hz)	2.59 (t, *J* 6 Hz)	3.69(s)
5-	0.88(t)			1.2–1.4(m)		2.49 (t, *J* 7 Hz)	S	2.55 (t, *J* 7 Hz)	1.98 (qn, *J* 7 Hz)	2.42 (t, *J* 7 Hz)	3.67(s)
6- to 14-	0.88(t)		1.2–1.4(m)		2.50 (t, *J* 7 Hz)	S	2.50 (t, *J* 7 Hz)		1.2–1.4(m)		2.33 (t, *J* 7 Hz)	3.65(s)
15-	0.90 (t, *J* 7 Hz)	1.2–1.4(m)	2.49 (t, *J* 7 Hz)	S	2.49 (t, *J* 7 Hz)			1.2–1.4(m)			2.33 (t, *J* 7 Hz)	3.65(s)
16-	1.24 (t, *J* 6.8 Hz)	2.57 (t, *J* 6.8 Hz)	S	2.50 (t, *J* 7 Hz)			1.2–1.4(m)				2.30 (t, *J* 7 Hz)	3.66(s)
17-	2.08(s)	S	2.49 (t, *J* 7 Hz)			1.2–1.4(m)					2.31 (t, *J* 7 Hz)	3.65(s)

Table 3. Values of δ_C(ppm) for all positional isomers of methyl thialaurate.

$$^{12}CH_3(CH_2)_n\text{–}S\text{–}(CH_2)_m{}^{1}CO_2{}^{13}CH_3$$

C_{12} Thia isomers	*n*	*m*	C-1	C-2	C-3	C-4	C-5	C-6	C-7	C-8	C-9	C-10	C-11	C-12	C-13
2-	9	0	171.70	S	31.15	29.90	28.80	29.65[a]	29.60[a]	29.40	29.30	32.00	22.75	14.05	53.70
3-	8	1	170.85	33.60	S	32.90	29.55	28.85	29.30[a]	29.20[a]	29.30	31.95	22.75	14.05	52.05
4-	7	2	172.20	35.00	27.25	S	32.40	29.75	28.95	29.30	29.30	31.90	22.70	14.05	51.50
5-	6	3	173.45	32.95	25.00	31.60	S	32.20	29.80	28.95	28.95	31.80	22.65	14.00	51.40
6-	5	4	173.50	33.70	24.30	29.30[a]	31.60	S	32.35	29.85[a]	28.70	31.90	22.60	14.00	51.30
7-	4	5	173.70	33.95	24.70	28.45	29.50	32.10[a]	S	32.30[a]	29.50	31.20	22.35	13.95	51.25
8-	3	6	173.80	34.00	24.90	28.85[a]	28.60[a]	29.65	32.25	S	32.00	32.00	22.10	13.65	51.25
9-	2	7	173.90	34.10	24.95	29.10	28.95[a]	28.75[a]	29.80	32.30	S	34.45	23.15	13.50	51.25
10-	1	8	173.90	34.10	25.00	29.15	29.15	29.15	28.90	29.75	31.85	S	26.10	14.90	51.20
11-	0	9	173.85	34.10	25.00	29.25	29.25	29.25	29.30	28.85	29.40	34.45	S	15.50	51.20

[a] Interchangeable.

Table 4. Values of δ_C(ppm) for all positional isomers of methyl thiastearate.

$$^{18}CH_3(CH_2)_n\text{–}S\text{–}(CH_2)_m{}^{1}CO_2{}^{19}CH_3$$

C_{18} Thia isomers	*n*	*m*	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
2-	15	0	171.80	S	31.15	29.90	28.75				29.15–29.75							32.00	22.75	14.05	53.75
3-	14	1	170.95	33.05	S	32.80	29.75	28.80				29.10–29.75						32.00	22.75	14.10	52.20
4-	13	2	172.35	34.85	27.10	S	32.30	29.75	28.95				29.30–29.75					32.00	22.75	14.10	51.65
5-	12	3	173.55	32.85	24.80	31.40	S	32.05	29.70	28.95				29.30–29.70				31.95	22.70	14.10	51.50
6-	11	4	173.65	33.65	24.20	29.20[a]	31.75	S	32.25	29.75	29.00[a]				29.35–29.75			32.00	22.75	14.15	51.35
7-	10	5	173.80	33.95	24.85	28.45	29.90	32.00	S	32.30	29.90	28.45				29.05–29.70		32.00	22.75	14.15	51.35
8-	9	6	173.80	33.95	24.85	28.80[a]	28.60[a]	29.90	32.10	S	32.30	29.90	28.60				29.05–29.40	32.00	22.75	14.15	51.25
9-	8	7	173.90	34.00	24.90	29.10	28.95	28.75	29.85[a]	32.30	S	32.30	29.75[a]	28.80	29.60	29.35	29.35	31.95	22.70	14.10	51.30
10-	7	8	173.80	34.10	25.05	29.20	29.20	29.20	29.00	29.85	32.30	S	32.30	29.85	29.00	29.35	29.35	31.95	22.75	14.15	51.25
11-	6	9	174.00	34.10	24.95		29.00–29.40			29.00	29.80	32.30	S	32.30	29.80	29.00	29.00	31.85	22.70	14.10	51.30
12-	5	10	173.45	34.00	25.10			29.10–29.45			28.75	29.70[a]	32.30	S	32.30	29.95[a]	28.75	31.70	22.75	14.10	51.10
13-	4	11	173.90	34.10	25.05				29.35–29.50			29.00	29.85	32.25	S	32.25	29.50	31.20	22.45	14.05	51.25
14-	3	12	174.05	34.10	25.00					29.20–29.60			29.00	29.80	32.25	S	31.90	31.90	22.10	13.70	51.30
15-	2	13	174.05	34.10	25.00						29.20–29.65			29.00	29.85	32.20	S	34.35	23.10	13.50	51.30
16-	1	14	174.15	34.10	24.95						29.15–29.65				29.00	29.70	31.75	S	25.95	14.85	51.30
17-	0	15	174.10	34.10	24.95							29.30–29.70				28.85	29.70	34.40	S	15.95	51.25

[a] Interchangeable.

due to the sulphur atom on the chemical shifts of the adjacent α-, β-, and γ-located methylene carbon nuclei, it became apparent that the incremental effects exerted by the sulphur atom depended considerably on the position of the sulphur atom in the alkyl chain of the fatty ester molecule. On the whole the sulphur atom in the alkyl chain induced a deshielding effect (*ca.* +2.70 ppm) on the shift of the α-methylene carbon atom located on either side of the sulphide linkage. The effect of the sulphur atom on the shift of the β-methylene carbon atoms was a weak deshielding effect (*ca.* +0.5 ppm), while the γ-methylene carbon atoms experienced a weak shielding effect (*ca.* −0.5 ppm).

An interesting phenomenon was observed in the case of the methyl 2- and 3-thia isomers, where the sulphur atom caused a strong shielding effect on the carbonyl carbon of the methoxycarbonyl (ester) group, but at the same time exerted a strong deshielding effect on the α-methylene carbon atom on the other side of the sulphide linkage. The shielding effect on the carbonyl carbon of the ester group in the methyl 2-thia isomers was probably due to a conjugative electronic effect involving the lone pair of electrons of the sulphur atom and the polarizable nature of the carbonyl system. In the methyl 3-thia isomers the shielding effect was more likely the product of a steric effect with the lone pair of the sulphur atom causing an effective shielding of the carbonyl carbon nucleus of the ester group. Another interesting observation in the case of the methyl 3-thia isomers was that the methylene carbon atom at the C-2 position was only very slightly shielded by the sulphur atom. In the methyl 4-thia isomers the shielding effect of the sulphur on the carbonyl carbon of the ester group was much less than that observed for the methyl 3-thia analogues, and the sulphur atom appeared to cause a deshielding effect on the α-methylene carbons located on either side of the sulphide linkage. This deshielding effect on the α-methylene carbon resonance by the sulphur atom prevailed in all the remaining positional isomers in both series of thia fatty ester analogues. Although it was not possible to establish a general set of shielding or deshielding effects due to the sulphur atom on the neighbouring carbon atoms, the ^{13}C n.m.r. spectral analysis of these compounds showed the possibility of identifying each of the methyl thialaurate isomers, and ten out of 16 positional isomers of methyl thiastearate by this technique.

Experimental

^{13}C N.m.r. spectra were obtained with a JEOL FX90 instrument operating at 22.62 MHz with proton noise decoupling. The spectra (3 000 to 6 000 accumulations; 27° pulse; 30 °C) were obtained from solutions in $CDCl_3$ (0.2–0.3 mol dm^{-3}), which also served as an internal deuterium lock. All spectra were calibrated against $SiMe_4$ as internal standard.

The synthesis of both series of C_{12} and C_{18} thia fatty acids was readily accomplished by treatment of n-alkanethiols with the corresponding ω-bromoalkanoic acids in the presence of KOH in ethanol,[10] except in the case of the 2-thia isomers. The 2-thia isomers were directly obtained as their methyl ester derivatives by reaction of the appropriate n-alkanethiol with methyl chloroformate in the presence of triethylamine in benzene, as the 2-thia-alkanoic acid decomposed rapidly on contact with dilute mineral acids.[13] Conversion of the other thia fatty acids to the methyl ester derivatives was achieved by refluxing with BF_3–methanol complex.

Short chain n-alkanethiols (C_1–C_6) were commercially available, while long chain n-alkanethiols were prepared by reacting n-bromoalkanes with thiourea.[14] n-Bromotetradecane and n-bromohexadecane were obtained by hydride reduction of methyl myristate and methyl palmitate respectively, followed by bromination using concentrated HBr in the presence of sulphuric acid. n-Bromopentadecane and n-bromotridecane were produced by the modified Hunsdiecker reaction of palmitic and myristic acid respectively.[15] Most of the shorter chain ω-bromoacids were commercially available, but could also be readily obtained by partial bromination of α,ω-alkanediols followed by $KMnO_4$ oxidation.[16,17] 15-Bromopentadecanoic acid was prepared from cyclopentadecanone by the method described by Bidd *et al.*[18]

General Method for the Preparation of Methyl Thia Fatty Esters as Exemplified by the Synthesis of Methyl 12-*Thiastearate.*—A mixture of hexanethiol (6 g, 51 mmol), 11-bromoundecanoic acid (11.5 g, 43 mmol), KOH (5.6 g, 0.1 mol) and ethanol (150 cm^3) was refluxed for 5 h under nitrogen. Water (75 cm^3) was added to the cooled reaction mixture which was then extracted with hexane (2 × 40 cm^3). The aqueous layer was acidified with conc. HCl and extracted again with diethyl ether (2 × 75 cm^3). The ethereal extract was washed with water (50 cm^3) and saturated NaCl solution (50 cm^3), then dried over anhydrous Na_2SO_4. Evaporation of solvent gave 12-thiastearic acid (10.9 g, 84%). The latter was refluxed with methanol (25 cm^3) and BF_3–MeOH (15%, w/w; 5 cm^3) for 15 min. Water (50 cm^3) was added to the cooled reaction mixture and extracted with hexane (2 × 30 cm^3). The extract was washed, dried and evaporated to dryness. Chromatography on silica gel with hexane–diethyl ether (95:5) as the eluant gave methyl 12-thiastearate (10.2 g, 89%).

General Method for the Preparation of Methyl 2-*Thia Fatty Esters as Exemplified by Methyl* 2-*Thiastearate.*—Methyl chloroformate (2.4 g, 25 mmol) was added to a mixture of hexadecanethiol (4.5 g, 17 mmol), triethylamine (3.5 g, 35 mmol) and benzene (75 cm^3) at 0 °C under nitrogen and stirred for 3 h at 0–10 °C. The reaction mixture was stirred at room temperature for a further 60 h. The reaction mixture was filtered and the solvent removed under reduced pressure. Chromatography on silica gel with hexane–diethyl ether (95:5) as the eluant gave methyl 2-thiastearate (4.5 g, 82%).

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