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# A SHORT-CHAIN ESTER FROM THE SEED OIL OF *CARDIOSPERMUM HALICACABUM* L.*†

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**Abstract**—Methyl esters prepared from the seed oil of *Cardiospermum halicacabum* L. yielded a small amount of a volatile ester, $C_9H_{18}O_5$. The molecular weight was confirmed by the mass spectrum. Examination by i.r. and NMR spectra indicated the probable structure as methyl 4,4-dimethoxy-3-(methoxymethyl)butyrate (I). Comparison of the spectra with those of methyl 9,9-dimethoxynonanoate and evidence of a lactone on hydrolysis provided additional support for this structure. The compound is presumed to exist in the oil as part of a glyceride or glyceryl ether and to be converted into the dimethylacetal ester during the interesterification treatment.

## INTRODUCTION

During a study of the fatty acids of glyceride oils in various seeds, the seed oil of *Cardiospermum halicacabum* L. (family Sapindaceae) was examined. The composition of the long-chain acids has been reported.[1] The oil was converted into methyl esters by direct transesterification with methanol and dry hydrogen chloride. On fractional distillation of the esters, an unusual low-boiling fraction was obtained. Because of the possibility that this fraction might be an intermediary metabolite in the biosythesis of fatty acids, it was of interest to examine its composition.

## RESULTS

The substance was purified by preparative gas–liquid chromatography (GLC), after which it gave a single peak on the chromatogram. It was a colorless liquid. The empirical formula by analysis was $C_9H_{18}O_5$ and the molecular weight, from the mass spectrum, was 206. The low refractive index, $n_D^{25}$ 1·4260, indicated that it was neither aromatic nor unsaturated. It was optically active, $[\alpha]_D^{25} + 3{\cdot}3$ (*ca.* 10 per cent in MeOH). There was no u.v. absorption maximum in the range 230–400 nm.

The i.r. spectrum (in $CS_2$) gave evidence of one or more ether groups (1075–1125 $cm^{-1}$), an ester (1740 $cm^{-1}$) and a short carbon chain (2950 $cm^{-1}$). Absence of a band near 700 $cm^{-1}$ showed that the chain was very short or branched. Hydroxyl absorption was negligible. There was no ordinary olefinic unsaturation (*ca.* 1650 $cm^{-1}$). This was confirmed by a Wijs iodine value determination and by the NMR spectrum.

Thus the preliminary examination showed that the substance was saturated, aliphatic, that it had ester and ether functions and probably a branched chain.

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[1] M. J. Chisholm and C. Y. Hopkins, *Can. J. Chem.* 36, 1537 (1958).

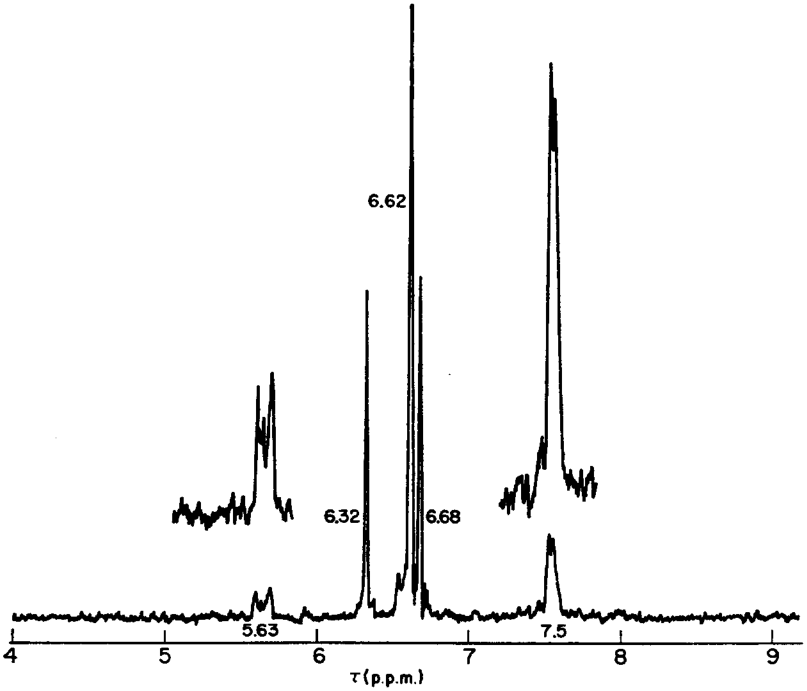


FIG. 1. THE NMR SPECTRUM OF THE COMPOUND AT 60 Mc/s IN $CDCl_3$.

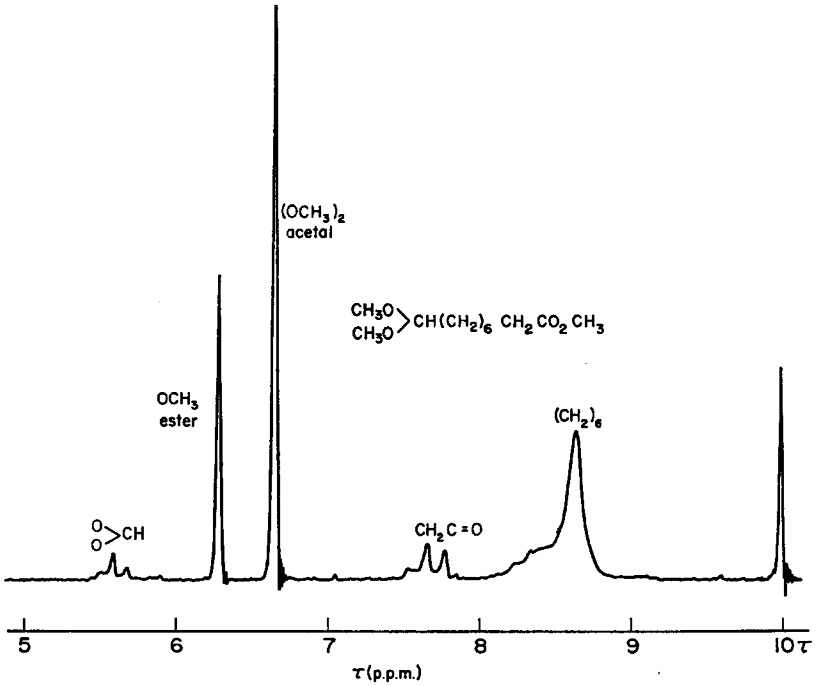


FIG. 2. THE NMR SPECTRUM OF A MODEL COMPOUND WITH A DIMETHYLACETAL GROUP, TAKEN AT 60 Mc/s IN $CDCl_3$.

## NMR *Spectrum*

The NMR spectrum in deuteriochloroform at 60 Mc/s (Fig. 1) had bands in the region $5\tau$ to $8\tau$ only. Thus the compound had no C-methyl group ($\sim 9\tau$), no olefinic protons ($3$–$5\tau$), and no aromatic protons ($\sim 2$–$3\tau$). The NMR data are given in Table 1.

TABLE 1. NUCLEAR MAGNETIC RESONANCE DATA (60 Mc/s)

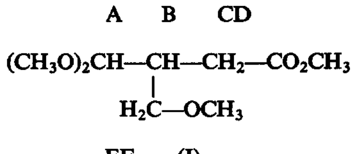
| Chemical shift, $\tau$, in $CDCl_3$ | in $C_6D_6$ | No. of protons* | Assignment | Type of band |
|---|---|---|---|---|
| 5·63 | 5·59 | 1 | H of acetal group | Doublet |
| 6·32 | 6·62 | 3 | $COOCH_3$ | Singlet |
| ~6·6 | ~6·55 | 2 | $—CH_2O—$ | See text |
| 6·62 | 6·86<br>6·88 | 6 | Two acetal $OCH_3$ | Doublet (see text) |
| 6·68 | 6·92 | 3 | Ether $OCH_3$ | Singlet |
| ~7·5 | ~7·4 | 3 | $\alpha CH_2$, $\beta CH$ | Multiplet |

* By measurement of the integration curve.

The i.r. and NMR data together showed the presence of one methyl ester group and one ordinary methyl ether group, thus accounting for three of the oxygen atoms. The remaining two oxygens were judged to form part of an acetal group on the basis of the chemical shifts of the respective protons, which were in close agreement with those of a model acetal ester, methyl 9,9-dimethoxynonanoate (Fig. 2). The model compound had signals at $5·59\tau$ (one proton, H of acetal group) and 6·64 (six protons, acetal $OCH_3$ groups) in deuteriochloroform.

However, the single proton of the acetal group in the unknown compound was essentially a doublet (spacing 5·3 c/s), indicating that the adjacent carbon atom carried a single hydrogen. Similarly, the signal for protons alpha to the carbomethoxy group ($\sim 7·5\tau$) was essentially a doublet.

A partly hidden signal at about $6·6\tau$ (in $CDCl_3$) was presumed to arise from a $—CH_2O—$ group. On the basis of the analytical and spectral data, therefore, the compound is judged to have the structure I.



$$\overset{A}{(CH_3O)_2CH}—\overset{B}{\underset{\underset{EF}{H_2C—OCH_3}}{CH}}—\overset{CD}{CH_2—CO_2CH_3}$$

(I)

The signal for the two acetal $OCH_3$ groups was resolved when the spectrum was recorded in hexadeuteriobenzene (spacing 1·2 c/s at $6·87\tau$). This non-equivalence implies that the acetal group is attached to an asymmetric carbon atom, thus providing further evidence for structure I. The resolution of the acetal $OCH_3$ signals was confirmed by a spectrum taken at 100 Mc/s (Fig. 3). In an analogous structure, the two methoxyl groups of arabinose dimethylacetal were shown by the NMR spectrum to be non-equivalent.[2]

[2] BRIAN CAPON and DAVID THACKER, *Proc. Chem. Soc.*, 369 (1964).

In $CDCl_3$, $H_B$, $H_C$ and $H_D$ appeared in the spectrum as a complex multiplet centered at about 7·5τ. The multiplicity of the signal for the three protons ($H_B$, $H_C$, $H_D$) is plausible when it is considered that $H_B$, $H_C$ and $H_D$ form an *ABC* system, $H_A$ an $H_B$ an *AX* system, and $H_B$, $H_E$ and $H_F$ a possible *XAB* system.

$H_E$ and $H_F$ had an average chemical shift in $CDCl_3$ of about 6·6τ, which supports the presence of a $—CH_2O—$ group (Fig. 1). This signal was partially obscured by the signal for the acetal methoxyls. However, when the spectrum was determined at 100 Mc in $C_6D_6$ (Fig. 3), the signal for protons $H_E$ and $H_F$ was clearly evident as a doublet centered at 6·59τ (spacing 5·0 c/s).

The doublet structure for these protons requires the accidental equivalence of $H_E$ and $H_F$ and that the adjacent carbon have a single hydrogen. The spacing (5·0 c/s) is in the expected range for an average coupling constant for aliphatic protons on adjacent carbon atoms.

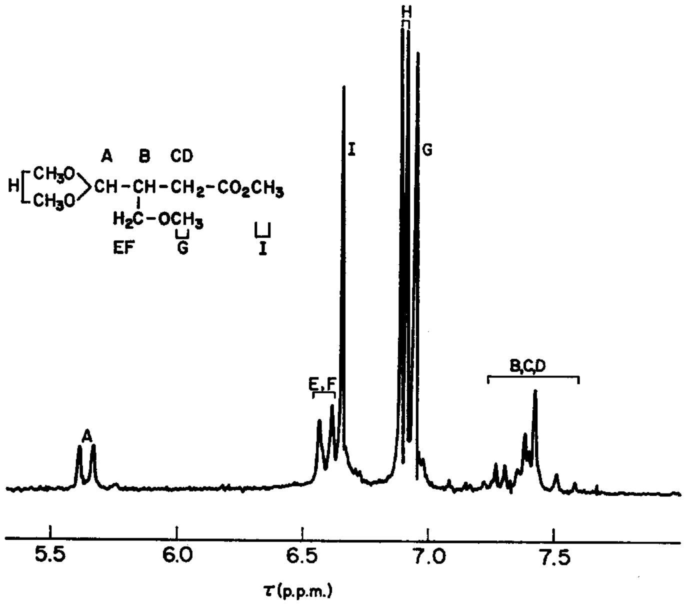
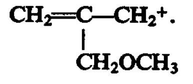


FIG. 3. THE NMR SPECTRUM OF THE COMPOUND AT 100 Mc/s IN BENZENE-$d_6$, AND THE TENTATIVE STRUCTURE.

Decoupling experiments at 100 Mc/s, with frequency sweep, established that $H_A$, $H_E$ and $H_F$ were coupled to part of the multiplet representing $H_B$, $H_C$ and $H_D$. This supports the assignment of $H_B$, since it must be coupled to $H_A$, $H_E$ and $H_F$, as well as to $H_C$ and $H_D$.

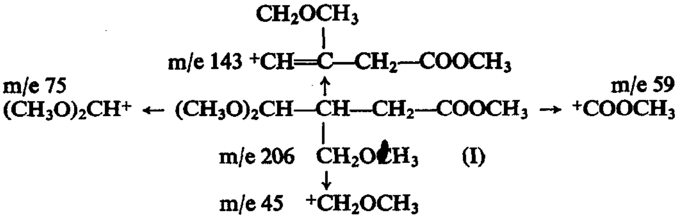
### *Mass Spectrum*

The mass spectra of the compound and the model compound were determined. The parent molecular ion of the compound appeared at m/e 206, although it was of low intensity. Major fragment peaks occurred at m/e 143, 101, 85, 75, 59 and 45. The base peak at m/e 75 was assigned to the acetal fragment $(CH_3O)_2CH^+$. The ion of m/e 85 was presumed to be



$$\underset{\displaystyle CH_2OCH_3}{CH_2{=}\underset{|}{C}{—}CH_2^+}.$$

Some of the other assignments were as follows:



$$\begin{tikzcd}[column sep=small] & \mathrm{CH_2OCH_3} & \\ & \text{m/e 143 } {}^{+}\mathrm{CH{=}C{-}CH_2{-}COOCH_3} & \\ \text{m/e 75 } \mathrm{(CH_3O)_2CH^{+}} & \mathrm{(CH_3O)_2CH{-}CH{-}CH_2{-}COOCH_3} \arrow[l] \arrow[r] \arrow[u] & \text{m/e 59 } {}^{+}\mathrm{COOCH_3} \\ & \text{m/e 206 } \mathrm{CH_2OCH_3} \arrow[d] & \\ & \text{m/e 45 } {}^{+}\mathrm{CH_2OCH_3} & \end{tikzcd}$$

(I)

The mass spectrum of methyl 9,9-dimethoxynonanoate gave corresponding fragment peaks at m/e 75 (acetal group) and 59 (ester group) but not at 85 or 143.

### *Chemical Reactions*

When the compound was hydrolyzed and then acidified, the product appeared to be a lactone. Its i.r. spectrum in $CS_2$ had a strong band at 1795 $cm^{-1}$ ($\gamma$-lactone) in place of the ester band. It had also an ether band at 1110 $cm^{-1}$.

When the compound was treated with 2,4-dinitrophenylhydrazine in acid solution, it gave a solid product, m.p. 156–157°, whose i.r. spectrum had the characteristics of a phenylhydrazone. However, this substance was not obtained analytically pure.

## DISCUSSION

The tentative structure (I) of the compound was deduced mainly from the NMR data, along with other spectroscopic and some chemical evidence. The presence of an acetal group is shown by the NMR signals, and is confirmed by the mass spectrum. Further evidence is provided by the formation of a phenylhydrazone, and by the formation of a lactone on hydrolysis. The i.r. band for the lactone, at 1795 $cm^{-1}$, is near the upper limit of the range for $\gamma$-lactones (1740–1800 $cm^{-1}$).[3] However, instances of the $\gamma$-lactone band occurring in this region (e.g. 1790 $cm^{-1}$) are not unusual.[4, 5]

In the NMR spectra, the two acetal $OCH_3$ groups were shown to be non-equivalent in $C_6D_6$ solution but this was not apparent in $CDCl_3$ solution at 60 Mc/s. The enhanced non-equivalence in $C_6D_6$ is probably due to a conformation redistribution, occasioned by the solvent. These observations indicate that the total intrinsic asymmetry of the system is low (cf. Raban[6]).

The NMR and mass spectra define the structure of the branch ($—CH_2OCH_3$). All of the evidence leads to the conclusion that the compound is methyl 4,4-dimethoxy-3-(methoxymethyl) butyrate (I).

A compound of this type has not been isolated previously from seed oils. Its presence in an oil suggests that it may be combined with glycerol, as an ester or ether. If so, it would be converted into the dimethylacetal ester by the interesterification treatment.

There is some similarity in structure to such natural substances as apiose and cordycepose, which have a $CH_2OH$ branch, and to mevaldic acid. The $—CH_2O—$ branch is also known in a fatty acid isolated by Jefferies and Knox from a plant source.[7]

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[3] L. J. Bellamy, *Infrared Spectra of Complex Molecules*, 2nd edition, pp. 186–187. Methuen, London (1958).

[4] C. Y. Hopkins, M. J. Chisholm and L. Prince, *Lipids* **1**, 118 (1966).

[5] J. D. Bu'Lock, *Quart. Rev. London* **10**, 376 (1956).

[6] M. Raban, *Tetrahedron Letters* 3105 (1966).

[7] P. R. Jefferies and J. R. Knox, *Aust. J. Chem.* **14**, 628 (1961).

## EXPERIMENTAL

Spectral measurements and GLC were carried out as follows: u.v. spectra in cyclohexane; i.r. spectra in $CS_2$; NMR spectra with tetramethylsilane as internal standard in solvents as stated and at 60 or 100 Mc/s; mass spectrum by means of a Hitachi magnetic deflection spectrometer at an electron energy of 75 eV; GLC with a polyester liquid phase, thermal conductivity detector, and column temperature 135°.

### *Isolation of the Compound*

The seeds were ground and extracted with light petroleum (30–60°) in a Soxhlet apparatus. After removal of the solvent, the oil was dissolved in dry MeOH containing 2 per cent by weight of dry HCl and refluxed for 16 hr. Most of the MeOH was removed by distillation, water was added and the esters were extracted with ethyl ether. The ether was removed and the residual esters were distilled in a Podbelniak spinning band column at 0·5 mm pressure. The fraction distilling at 58–63° at 0·5 mm (0·5 per cent of the total esters) was examined by GLC. It gave two peaks of relative areas 5:1. The portion represented by the larger peak was collected in a cooled receiver containing glass wool wetted with MeOH and the condensate was recovered.

### *Properties of the Compound*

The compound gave a single peak on GLC. It was a colorless liquid with a slight odor. On the basis of the data and discussion above, it was tentatively identified as *methyl*-4,4-*dimethoxy*-3-(*methoxymethyl*)*butyrate*. (Found: C, 52·57; H, 8·75. $C_9H_{18}O_5$ required: C, 52·42; H, 8·80 per cent.) It had b.p. 58–60°/0·5 mm and $n_D^{25}$ 1·4260. The i.r., NMR, and mass spectra have been described. There was slight end absorption in the u.v. (280–230 nm) but no maximum in the range 400–230 nm. There was negligible absorption of iodine from Wijs solution (iodine value 2·9).

Hydrolysis of the compound in acid or alkaline solution gave a water-soluble product. It was extracted with ether as a colorless liquid with a mild fruity odor. This product had $\nu_{max}$ 1110 (ether), 1795 (lactone), and 2940 (carbon chain) cm$^{-1}$.

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